Enhancing resilience at systems level through breeding for diverse cropping systems (T3.2)

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Authors	**Pedro Mendes Moreira, Agnes Bruszik, Edwin Nuijten, Ambrogio Costanzo, Monika Messmer, Linda Legzdina, Mariateresa Lazzaro**

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Contents

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Executive Summary

In order to enhance resilience at systems level through breeding for diverse cropping systems, the LIVESEED project identified three levels of investigation:

- crop level (through more diverse cultivars, cultivar mixtures and heterogeneous material)
- field level (through various types of mixtures of annual and perennial crops) and
- systems level (by increasing crop number and complexity of its relationships, such as done in agroforestry systems).

LIVESEED partners developed and/or continued and analysed field trials under these three levels, applying breeding methods and co-design approaches to optimise annual crop mixtures, perennial crops, and agroforestry crops.

Altogether, 26 field trials carried out by 12 institutions in 10 different EU countries have been analysed in this document with regards to their applied breeding strategies, applied methodology, analysed traits, their multi-actor approaches, and their results and contribution to the diversification of cropping systems and the increase of resilience. Six potential overarching hypotheses that were collaboratively identified by the project partners, as well as several additional, individual hypotheses have been tested with these trials. The individual trials have been described in depth in the trials' full reports in Annex I.

Based on this work, our results suggest:

1) **Concerning the traits and characteristics involved in general or specific mixing ability of cereal cultivar mixtures, CCPs or cereal-legumes mixtures, perennial grass-legume mixtures, and in crop adaptation in agroforestry systems**

An important question was how to predict mixing ability for cultivar mixtures with a focus on cereal crops. Several partners have been comparing cultivar mixtures with the same cultivars in monoculture. Unfortunately, no generalisations can be made based on trial results. In the case of barley trials, combining mixtures with components differing for three target traits did not ensure the expected effect in all cases and therefore could not be generalized. Stable mixtures arose by combining components with different adaptability, but not in all cases. For spring wheat, the cultivar mixtures overall did not show a better performance than the best performing cultivars. Winter wheat participatory trials confirmed that the participatory ideotyping approach is the best way to predict the mixing ability of cultivars.

Another question was to better understand the potential benefits of composite cross populations (CCPs) compared to cultivar mixtures and cultivars in single stands. For barley, CCPs had a potential for organic and stress environments (for yield, yield stability, diseases and weed competition) and CCPs might be better suited than homogenous varieties. Similar results were observed for winter wheat in France. The results suggest that for developing populations with improved resilience, the components need to be contrasting genetically to traits related to disease resistance. The overall results suggest that it is best for CCPs to be developed under the same growing conditions as the target environment.

An important reason to grow legume-cereal mixtures is their improved competitiveness to weeds compared to a mono legume crop. Such cropping system fits well within organic farm management. In the trials oriented towards interspecific diversity, different combinations of cereals and leguminous crops (pea, lupin, alfalfa-grass mixtures) were tested based on the local agro-ecological conditions and

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farming contexts. An important question was how to breed for improved mixing ability. For the pea and lupin mixtures, plant type can determine whether breeding for mixing ability is necessary or not. The traits to look at in a crop for good mixing ability can be also very broad. For lupin, depending on the local context, specific plant types are more appropriate and determine whether breeding for mixing ability is necessary.

It was also concluded that it is important to define the objectives which performance and ecosystem functions should be improved, because in many cases it is not the total yield of the mixture, but the yield of the two components that will be processed separately. A mixed stand can improve the baking quality of wheat for instance when combined with a legume crop. However, in the case of food purposes, an important aspect is the quality of the grain separation to avoid the chances of allergenic reactions when some grains of a lupin crop remain in the cereal grains. Hence, the companion crop must also have market potential next to good agronomic potential in mixtures.

For perennial alfalfa-grass mixtures, using a nursery system with alfalfa and undersown lawn type grasses reduced weeding efforts in comparison to alfalfa pure stand. When working within a set of elite germplasm with reduced variation, it is recommended that the selection decision consider the cultivation system.

The agroforestry systems provide important opportunities for evolutionary and/or direct selection harnessing complex interactions that become even more complex when the level of genetic diversity (selection) of different perennial and annual crops is included. In these systems, plant-plant and soil-plant interactions provide to be more complex than previously assumed as spatial arrangement and microclimate play a major role. Thus, further research is required to define the plant traits best adapted to intercropping both for alleys and interrow traits and the best performance in specific temporal and spatial positions.

2) **New breeding methods, experimental designs, and selection tools for various mixed cropping systems**

Concerning breeding methods to increase intraspecific diversity, tailor-made variety mixtures were developed using a participatory ideotyping approach in France, where co-design assembly rules were tailored to the farmers' local contexts based on interviews and workshops with organic farmers. Another new method to enhance diversity in cereal populations is the so-called Diversified Oriented Population (DOP). The strategy is to request many accessions of a species from gene banks, which are then multiplied and observed (for mainly phenological and morphological criteria). Then the accessions are then mixed according to the farmer's choices based on various criteria. The results in LIVESEED trials showed that the breeding for inter-specific diversity (crop mixtures) needs to be context-specific, both at the agro-climatic and value-chain level. It is very important to have clear goals set for the plant type and the companion crop(s), which depend on the local context. When breeding for mixed cropping, it is necessary to evaluate both species in mixed stand, as the ranking in performance of pure stand crops can differ from the ranking in a mixed stand. When designing mixed stand cereal-legume evaluations, one or a few testers of the cereal crop (cultivars) can be sufficient when GMA effects are dominating, while there is no or little SMA. The type of companion crop also determines the crop phenotype to breed for.

For mixtures. it was relevant to develop synchronization of the crop cycles to make harvesting easier. To better cope with weed pressure, some companion crop proved useful in supporting the other, under unfavourable conditions. Agronomic aspects (such as plant density, seed ratio of the mixed crops, time of sowing, sowing depth, sowing method, soil type, and soil fertility) can be important

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factors that influence plant growth and hence the selection process and breeding progress. Moreover, the specific advantages of crop mixtures might vary considerably between agroecological zones, as local stress factors (e.g., competition for water, nutrients and light, different pest, and disease pressure) might have a greater influence.

Several tools for breeding for diversity were developed, tested, and implemented: in France, the history of the seed lots (parental material) in the creation of CCP, breeding and multiplication can be followed in a database called SHiNeMaS (Seeds History and Network Management System), that allows for transparency and knowledge transfer. Statistical tools matching diverse contexts and scales were identified and explored by partners. Training sessions on statistical tools were also held and training materials shared among breeders. IPC with the collaboration of FEUP (University of Oporto) assisted AREI to apply the methodology of random forest, CART and MARS for barley mixtures.

3) **Improving resilience of crops with inter- and intra-species genetic diversity on field specifically fitted for organic farming**

LIVESEED's approach was to evaluate how resilience can be improved at the following levels: (i) at the crop level through the creation of more diverse cultivars, cultivar mixtures and heterogeneous materials, (ii) at the field level through various types of mixtures of annual and perennial crops, and (iii) at system level through increasing crop numbers and the complexity of its relationships (agroforestry systems). Overall, the results suggest that increasing diversity is particularly useful to improve resilience in stress prone conditions. Through the participatory breeding experiments in France on winter wheat, the created genetic diversity enabled resilience for those populations. For other crops (e.g., barley, maize) diversity improved yield stability, proving that diversity incorporated in CCPs can improve yield stability over environments. Several CCPs or DOPs of the diverse crops have been developed in the scope of LIVESEED. Due to the new Organic Regulation (EU, 2018/848) about the simple notification of Organic Heterogeneous Material (OHM), these new sources of diversified organic seeds can be made commercially available to a larger number of organic farmers, which will contribute to resilience at the seed system level.

At the field level, improving resilience through crop mixtures proved more difficult compared to increasing intraspecific diversity. In the first place, in the case of crop mixtures it is important to attune the crop phenology, ripening time and plant density of the crops. Tillering ability, directly related to plant density, is important for improved resilience at monocrop level, but can hamper or compete crop growth of the companion crop under specific conditions. In the trials mixing leguminous crops with cereals, contrasting results were observed with regards to better coping with weed pressure and lodging resistance. The overall conclusion was that intercropping seems to be a good solution to prevent lodging of legumes (e.g., pea). This is particular the case for low-input conditions. The effect of the mixtures on resilience by offering a better disease resistance could not always be confirmed due to very low disease pressure. In some specific cases crop mixtures may induce a conducive environment for specific diseases. On the other hand, results revealed higher resilience of the lupin-cereal mixtures in comparison to the crops sown in pure stands concerning yield stability over years. In the perennial alfalfa-grass mixture trials and maize-beans intercropping trials no clear improvement of resilience of the system were observed.

4) **Novel approach to co-design with farmers' blends of cultivars or species mixtures specifically according to their local conditions, market, and socio-economic context**

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In the LIVESEED project, several co-design approaches (CCPs, DOPs, participatory ideotyping, involving farmers in the choice of the starting genetic material, culinary breeding, etc.) were applied, responding to different local pedo-climatic conditions, markets, seed systems, and socio-economic contexts. These differences may influence the acceptance of co-designed materials, and the co-creation process, e.g., CCPs may be more accepted in one country than in another. CCPs and DOPs were reported as materials that can be used easily for on-farm breeding, increasing the adaptability to a diversity of pedo-climatic conditions, and reducing the costs of breeding. In the ideotyping trial, the co-designed assembly rules implemented on a multi-criteria assessment tool helped farmers when designing mixtures. This work emphasises the interest and importance to joining up knowledge between actors form different disciplines and perspectives. In the maize trial in Portugal, it proved also important to keep farmers motivated and interested in germplasm evaluations to adapt their preferred populations to their systems. Culinary breeding networks could promote the use of traditional maize populations and underutilized maize landraces, as they have great grain quality traits that allow a wider range of products to be developed.

From the experiences it can be concluded that researchers/facilitators will play a crucial role in the near future in providing capacity building for farmers, to enable them to work with such materials on their own. For the time being, farmers require the link with scientists for access to genetic materials, for selection methods, evaluation and statistical analyses and coordination. Farm advisory services could build in such programs for on-farm breeding across Europe to facilitate uptake. Farmers are willing to sacrifice land from production and their time to develop improved cultivars suited for their local conditions.

5) **Prove of concept for breeding for more diverse cropping systems**

The results show that for developing cultivar mixtures, Diversity Oriented Populations (DOP) and Composite Cross Populations (CCP), a participatory ideotyping approach is advised. For breeding for mixed cropping, different breeding tools can be helpful, depending on the set goal and the local context. Based on statistical analysis of the individual cases, most efficient strategies can be developed (e.g., if cultivars bred for monocropping can be used or if specific breeding for mixed cropping needs to be set up). For breeding for more diversity, considering the local context, good collaboration with farmers, processors and other value chain actors is crucial to integrate farmer and expert knowledge and to embed the new populations (OHM) and mixtures in the food system.

Multi-actor approaches have a potential advantage of developing material better suited for the needs of the different actors with high adoption rate and a better distribution of the work reducing the financial costs. In the future, not only economic and ecological, but also social resilience of plant breeding will become more important as outlined in the systems-based breeding framework developed by LIVESEED (Lammerts van Bueren et al., 2018).

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Introduction

In organic agriculture, the first step for a successful crop is the use of quality seed of adapted germplasm. Adapted germplasm needs to meet the needs for Genetic x Environment x Societal interactions specific to the target organic farming system. Measures to improve plant growth during the season are limited, but also undesired from the perspective of a holistic approach which is at the basis of organic. Conventional varieties rarely meet the needs of organic and low-input agriculture, were environmental variation and stressors such as pests and diseases cannot be mitigated by synthetic inputs. Under those conditions, farmers must rely on the crop's own disease and pest resistance, weed suppression capacity and stability of production under low or irregular levels of soil fertility. Hence, crop varieties need to have good yield stability and general resilience, rather than maximum yield potential. Improved resilience can also help buffering against climate variability and climate change. An important challenge in organic breeding is to maintain a favourable balance between resilience, yield, and quality.

Following the principles[1] of organic agriculture, i.e., ecology, health, fairness and care, an important challenge is how to benefit from genetic diversity to improve aspects of resilience, local adaptation, yield stability and societal dimension through Genotype x Environment interactions.

The current document points out the concept of resilience at systems level and how breeding can help to enhance it to create optimised cropping systems.

Resilience's common meaning is 'the capacity to recover quickly from difficulties. It is also referred to as toughness, elasticity, or plasticity. The EU-funded project SOLIBAM (2010-2014) defines resilience as the capacity of an ecosystem to respond to a perturbation by resisting damage and recovering quickly. A resilient system will reorganize while undergoing change to still retain essentially the same function, structure, identity and feedbacks. Thus, resilience is linked to the adaptive capacity of a system in the face of change. Within the EU-funded project DIVERSIFOOD (2015-2019), the resilience concept was extended to the whole food system, including economic, social, political, and cultural dimensions. Thus, resilience of the food system calls for adaptive capacities of the food chain at the agro-ecological and socio-economic level to provide sufficient high-quality food and to maintain its cohesion over time. However, the building of resilience in agro-ecosystems needs further studies (Lin, 2011). An in-depth literature review by LIVESEED partners (Nuijten et al., 2020) provided us some of the key concept and background to better approach resilience considering its multidimensionality (Table 1).

The aims of the trials in the LIVESEED project's task 3.2 were to generate new insights on organic breeding approaches and contribute to our understanding on some of the aspects listed in Table 1. The task was divided into three subtasks:

- T3.2.1 New breeding methods and co-design approaches for optimised annual crop mixtures, with a focus on cereals
- T3.2.2: New breeding methods and co-design approaches for optimised perennial crop mixtures, with a focus on grass-legume mixtures
- T3.2.3: Development of concepts for co-breeding of agroforestry cropping systems

[1] https://www.ifoam.bio/why-organic/shaping-agriculture/four-principles-organic

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Therefore, the task focused on different types of mixtures: mixtures of two annual species, mixtures of two perennial species and complex agroforestry systems. Each subtask implemented in this 3.2 task had different goals, and hence the trials had a different dynamic. Also, the number of partners involved in each of the tasks and subtasks differed, and so did the number of years and the number of locations the trials were repeated, the stakeholders involved, the trial designs applied, and the statistical methods used.

Table 1. Aspects of resilience identified in LIVESEED Deliverable 3.5 based on literature review by Nuijten et al. (2020).

Sections	Topics
Section 1: Broader perspectives on organic breeding	Topic 1) Incorporating cultural and ethical aspects into breeding
	Topic 2) How organic breeding deals with IPR, patents, and geographical indication schemes (PDO and PGI)
	Topic 3) Collaborative approaches in breeding (such as community-based breeding, chain based breeding and participatory approaches)
	Topic 4) Breeding for diversity based on agro-ecological principles: Concepts of genetic diversity, resilience, local adaptation, Examples on GxExM interaction; and GxExS interaction
	Topic 5) Financing organic breeding initiatives
Section 2: Breeding strategies for organic breeding	Topic 6) Trade-offs between resilience, yield, and quality in the breeding process
	Topic 7) Breeding for complex systems (such as mixed cropping, agroforestry)
	Topic 8) Balanced plant – microbe interaction (such as plant communication and defence strategies)
Section 3: Specific breeding tools for organic	Topic 9) direct versus indirect selection, and indirect parameters for complex traits like NUE and WUE
	Topic 10) Use and efficiency of molecular tools, such as MAS, and genomics
	Topic 11) Inheritance of resistance to seed and soil borne diseases: From a gene-based to a cropping-systems perspective
	Topic 12) Breeding for integrated weed management
	Topic 13) Challenges and perspectives in rootstock and scion organic fruit tree breeding
	Topic 14) Efficient breeding methods: Decentralized-Participatory Breeding for Organic Agriculture

This diversity allowed a **comparative approach and analysis grounded in local contexts**, with the aim to maximise the applicability of the results to a wide range of crops and contexts, but also, next to detailed analysis per trial with different species, cultivar types and crop traits, allowed a more general analysis comparing these different systems, better understanding what type of traits are important for each type of crop mixture or comparison on the possibilities of using multi-actor approaches in different types of trials and their outcomes.

The LIVESEED trials involved twelve partners from ten countries in which seven crops per se and more than 14 crop combinations were used, including combinations with perennial crops and agroforestry (Table 2-3, page 9-10). The cultivar mixtures were all food crops and can also be used as feed in the

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case of barley, wheat, maize, and white lupin. In the case of the crop mixtures most have dual suitability for feed/food, with some exemptions being the feed for perennial grass and food for the beans or tomato. In most of the trials of cultivar mixtures, there has been a concern for crop improvement. In 60% of the trials were carried out on-farm. The years of the trials were for most cases between 2017 and 2020, however there are cases of partners whose work goes back to 2015 during other projects, for culture mixtures, such as ITAB + INRAE Rennes (e.g., COBRA bread wheat populations), AREI (e.g., spring barley), and IPC (e.g., Project VASO on PPB maize). In the case of crop mixtures, we have the example of FIBL-CH (e.g., blue lupin x cereals and white lupin x cereal or grasses or other crops). Most partners have obtained in the framework of the LIVESEED project trial data for 3 years in the period 2017 and 2020.

1. Experimental Trials

1.1. The objectives of the trials and research goals

The general objectives were the following:

- To identify the best cereal-legume crop combinations, considering agronomic and socio-economic contexts.
- To quantify benefits and drawbacks of crop mixtures compared to pure stands.
- To identify best varieties of each crop to associate with the partner crop, and traits involved in these mixing abilities.
- To explore whether it is possible to predict mixing ability based on a set of identified key-traits important for mixing ability.
- To identify best combinations of varieties from the two crops for particular purposes.

An important objective formulated was to increase knowledge on how to build the diversified populations and crop or variety mixtures:

- Can we characterise varieties for their mixing ability, what is the range of mixing ability depending on the origin and breeding method of the varieties?
- Can we identify traits to be associated with predictability for mixing ability (in a “ideotype” or “idea-pop” approach)?
- In the case of Dynamic Populations versus Composite Cross Populations: what are the advantages and disadvantages of mixing vs crossing the components?

To research breeding for **intra-specific diversity**, the LIVESEED project partners worked with cultivar mixtures and various types of heterogeneous populations.

The objectives of the work done on cultivar/genotype mixtures were: (1) to design variety mixtures specifically adapted to the local farmers’ production context (INRAE, winter wheat), (2) to identify traits important for mixed stands, to find out whether it is possible to predict which (type of) cultivars are suitable for use in mixed stands (LBI, spring wheat), and (3) to compare mixtures and components in pure stand and to conclude on the effect of specific component traits on mixture performance (AREI, spring barley).

The research on populations was aimed to (1) to compare two breeding strategies: Composite Cross populations and Dynamic populations in terms of diversity level and adaptation (ITAB, winter bred wheat), (2) to create different CCPs in order to fit different types of market of the farmers of the

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cooperatives: on-farm baking or selling grain to on-farm bakeries, as well as long chain markets, and also suitable for poor and productive lands (UBIOS and INRAE, winter wheat), (3) to create and distribute to farmers Diversified Oriented Populations (DOPs) and start their evaluation (ITAB, underutilized species - rivet wheat, spelt and oat), (4) to identify traits important for populations (LBI, spring wheat), (5) compare agronomic performance of populations with different diversity level to check varieties in order to recommend to farmers (AREI, spring barley). (6) In addition, completely uniform doubled haploid lines were compared to traditionally selected pure lines to find out if there is a difference in adaptability between both types of lines (AREI, spring barley). We also aimed at the (7) regeneration, multiplication, characterization and agronomic evaluation under breeder and farmer conditions of germplasm collection of Azores Islands and VASO germplasm to provide varieties adapted to organic and agroecological agricultural systems by recovering underutilized populations of Portuguese maize germplasm with gastronomic potential for use in organic farming and low-input systems (IPC, maize), and (8) to test the performance of a composite cross population of white lupin in terms of yield production and susceptibility to anthracnose (FiBL).

The research objectives for **inter-specific diversity** were mostly on cereal-grain legume mixtures and formulated by partners as following: (1) to compare how the yield and yield components change when peas, beans and vetch are grown in pure stand and in mixtures (AREI, grain legume mixtures with spring wheat or spring triticale), (2) screening of genetic resources for performance under mixed cropping for breeding purposes (GZPK, pea mixtures with triticale or barley), (3) to study effects on yield of different combinations between normal and semi-leafless types of winter peas of different plant length with three winter triticale of different length (CULTIVARI), (4) to identify traits important for mixed cropping of cereals and grain legumes (LBI, different cereals/pea, spring wheat/white lupin, spring wheat/ faba bean), (5) to find out what are the differences in yield, diseases, plant development and ripening between the cultivars in mixed and pure stand and are commercial varieties suitable enough for mixed cropping and if there a difference between varieties in how they adapt to mixed cropping (LBI, spring wheat/ faba bean), (6) to assess the productivity of various cereal-lupin mixtures and to indicate the most favourable variants of these mixtures in conditions of organic farming, especially for poor/sandy soils and evaluate traits involved in general mixing ability (IUNG-PIB, spring triticale and oat/ blue lupin and yellow lupin), (7) to test whether mixed cropping (lupin with another cash crop) or under-sowing (lupin with a subsidiary crop not to be harvested at the end of the season) are useful practices to optimise lupin cultivation in Switzerland (FiBL-CH, blue lupin and white lupin/ different cereals or subsidiary crops), (8) to assess the extent of accession-by-cultivation system interaction, to determine a suitable selection system for selection in mixture, and to produce seeds of experimental populations that have been selected in the three systems to assess realized selection gain in later plot trials for forage mixtures (AGROSCOPE and NARDI, alfalfa/ tall fescue and red fescue).

Trials on **agroforestry** were aimed (1) to continue to promote Successional Agroforestry Systems (SAFs) in Portugal (IPC), (2) develop a maize CCP in co-breeding for Agroforestry Intercropping Systems (IPC), and (3) to understand the main drivers of microbiome structuration of on-farm selected tomato in contrasted agroforestry conditions and to get inspiration for organic selection in order to improve resilience with holistic breeding strategies relying on plant and microorganism's co-evolution (INRAE).

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1.2. Overview of the trials I – Intra-specific diversity

In the Table 2 and 3 below, we are presenting an overview of the case studies of the Task 3.2. Whereas some partners focused on cultivar mixtures, others focused on crop mixtures. Some partners conducted several trials with different objectives in parallel. The locations of the trials, and hence the pedo-climatic conditions differed widely.

Table 2. LIVESEED partners involved, respective crops and typology of trials targeting intra-specific diversity

							Intraspecific diversity		Typology				Value Chain		On-farm	Years of trials (green) and years of data analysed from trials (diagonal stripes)						Data on Pure Stands
Trial nr.	Trial code	LIVESEED Partner involved	Country	Subtask in LIVESEED	crop type	Trial crops	Cultivar mixtures	Organic Heterogeneous population	Land-races	Open pollinated Variety	Inbred (self-pollinating)	Doubled Haploid	Main purpose	Crop improvement part of experiment		2015	2016	2017	2018	2019	2020	
1	1I_WW	INRA + GQE-Le Moulon	France	3.2.1	winter wheat	Participatory on-farm breeding for diverse wheat mixtures							food									
2	2I_WW	INRA + GQE-Le Moulon	France	3.2.1	winter wheat	Participatory ideotyping (co-design and assessment) of wheat mixtures							food									
3	3I_WW	UBIOS+INRAE-Le Moulon	France	3.2.1	winter wheat	Development of winter wheat CCP							food									
4a	4aI_WW	ITAB + INRA Rennes	France	3.2.1	winter wheat	Bread wheat							food									
4b	4bI_WR	ITAB + INRA Rennes	France	3.2.1	winter rivet wheat	DOP-rivet							food									
4c	4cI_WW	ITAB + INRA Rennes	France	3.2.1	winter spelt wheat	DOP-spelt							food									
4d	4dI_Ot	ITAB + INRA	France	3.2.1	winter oat	DOP-oat							food									
5	5I_SW	LBI	Netherlands	3.2.1	spring wheat	Spring wheat							food									
6	6I_Br	AREI	Latvia	3.2.1	barley	Spring barley							feed/food									
7	7I_Z	IPC	Portugal	3.2.1	maize	Maize landraces, improved populations and CCP							food/feed									
8	8I_WL	FiBL-CH	Switzerland	3.2.1	white lupin	Lupin CCP							feed/food									

1.3. Overview of the trials II – Inter-specific diversity

Table 3. Partners involved, respective crops and typology of trials targeting inter-specific diversity

					Interspecific diversity	Trial characteristics			Value Chain		On-farm	Years of trials						Data on Pure Stands
Trial nr.	**Trial code**	**LIVESEED Partner involved**	**Country**	**Subtask in LIVESEED**	**crop mixtures**	**annual/perennial**	**crop type**	**Trial crops**	**Main purpose**	**Crop improvement part of experiment**		**2015**	**2016**	**2017**	**2018**	**2019**	**2020**	
9	9IE_P	AREI	Latvia	3.2.1		annual	pea	spring wheat x spring pea	feed/food									
10	10IE_P	FiBL-CH-GZPK	Switzerland	3.2.1		annual	pea	spring triticale/barely - pea	feed/food									
11	11IE_P	Cultivari	Germany	3.2.1		annual	pea	winter triticale x winter pea (no wheat)	feed									
12	12IE_P	LBI	Netherlands	3.2.1		annual	pea	different cereals/pea	food/feed									
13	13IE_FB	LBI	Netherlands	3.2.1		annual	faba bean	spring wheat x faba bean	food/feed									
14	14IE_FB	AREI	Latvia	3.2.1		annual	faba bean	bean	feed/food									
15	15IE_BL	IUNG	Poland	3.2.1		annual	blue lupin	mixed cropping spring triticale x blue lupin	feed									
16	16IE_BL	IUNG	Poland	3.2.1		annual	blue lupin	mixed cropping spring oat x blue lupin	feed									
17	17IE_BL	FiBL-CH	Switzerland	3.2.1		annual	blue lupin	mixed cropping blue lupin - different partners	food/feed									
18	18IE_WL	FiBL-CH	Switzerland	3.2.1		annual	white lupin	mixed cropping white lupin - different partners	food/feed									
19	19IE_WL	LBI	Netherlands	3.2.1		annual	white lupin	spring wheat x lupine	food/feed									
20	20IE_YL	IUNG	Poland	3.2.1		annual	yellow lupin	mixed cropping spring trirticale x yellow lupin	feed/food									
21	21IE_YL	IUNG	Poland	3.2.1		annual	yellow lupin	mixed cropping spring oat x yellow lupin	feed/food									
22	22IE_V	AREI	Latvia	3.2.1		annual	vetch	spring wheat/triticale x spring vetch	green fodder									
23a	23aP_Gss	AGROS	Switzerland	3.2.2		perennial	alfalfa	grass-legume mixtures	feed									
23b	23bP_Gss	NARDI	Romania	3.2.2		perennial	alfalfa	grass-legume mixtures	feed									
24	24P_Ph	IPC	Portugal	3.2.3		annual	beans	maize x beans	food									
25	25SAF	INRA	France	3.2.1, 3.2.3		perennial	tomato	tomato	food									
26	26SAF	IPC	Portugal	3.2.3		perennial SAF	maize	successional agroforestry early selection of maize under coevolution, no results still	food									

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1.4. Methodology

In this chapter we will introduce the main aspects concerning how the work was organised within the consortium of the LIVESEED partners.

1.4.1. Meetings and steps of implementation

An overview of the main steps and meetings leading to the preparation of current document is presented in Figure 1 below.

Figure 1. Workflow of Task 3.2 in LIVESEED between 2017-2021

As it can be seen from the workflow, many occasions were taken for discussions due to the complexity of the tasks to target synthesis and overarching frameworks in which the contributing diverse trials can be analysed. We held several meetings addressing the statistical methods and to assess the needs of partners to carry out more complex analysis to fit the larger conceptual frameworks.

1.4.2. Monitoring progress in the trials - Annual Reports

A very detailed yearly reporting template was developed (see Annex I trial reports) with the contribution of several project partners and was used by the partners to explain the trial's objectives, location(s); the genetic materials and list of accessions used (including their origin and any relevant background information available), the list of traits assessed, what hypothesis were used/tested/evolved, and how the research question and hypothesis have been developed. The yearly report was important to maintain a good overview on the activities conducted in Task 3.2.

Information was also collected on how the trials contribute to diversity management within plant breeding activities on cereals and/or enhances resilience at systems level through breeding for diverse cropping systems. Partners reported if the experiment would result in 1) lists of traits and characteristics important for general or specific mixing ability of cereal cultivar mixtures, CCPs or cereal-legumes mixtures; 2) new breeding methods, experimental designs and selection tools for various mixed cropping systems; 3) novel approaches to co-design with farmers blends of cultivars or species mixtures specifically adapted to their local conditions, market and socio-economic context; and 4) if improved plant populations were selected suited for mixtures and/or pure stands.

Concerning trial design and management, partners reported on how the trial was sown, grown, harvested, which experimental design and which additional factors (randomised blocks, incomplete blocks, etc) were adopted.

Next to the statistical analyses conducted, partners were asked to explain the main results from the trials and provide a brief discussion interpretating the results. For the present document, the partners were also requested to explain the main lessons learned with regards to the trials, and how they thought the information gained could be transferred to other situations.

1.4.3. Trial data collection

Due to the diversity of the trials and their contexts, the project partners used a broad range of methods for data collection and comparison, including a set of meetings, the annual reports of the trials, and questionnaires. Next to the annual reporting templates, a yearly data and metadata template was also used (developed by FiBL-CH and AREI), in line with the project's Data Management Plan. Partners uploaded their data files to the LIVESEED project's internal Sharepoint. Furthermore, during LIVESEED internal workshops and various European conferences (EUCARPIA Conference organic and low input 2018, 2021; Crop Diversification 2019; Intercropping for Sustainability 2021), researchers active in other research projects such as ReMIX and DIVERSIFY joined the discussions for common learning and the exchange of new insights and methodologies. Data of field trials which are already published were made available in open repositories, such as raw data from trials on alfalfa in mixture with grasses[2] or on wheat variety mixtures[3].

1.4.4. Data analysis

For data analysis also a range of statistical methods were used by the partners. These have been summarized per trial in Table 4. In the annual report, partners listed the traits for which the trial they made information available, and the types of data available (e.g., qualitative, quantitative t/ha, rank, scoring, etc.). Additional information in excel spreadsheets were provided as supplementary material if the datasets were too long or too complex.

2 Grieder, C. 2021. Raw Data: Breeding alfalfa (Medicago sativa L.) in mixture with grasses
Link to the data: https://zenodo.org/record/5035280#.YN3f1EyxVZU

3 Forst, E., Enjalbert, J., Allard, V., Ambroise, C., Krissaane, I., Mary-Huard, T., & Goldringer, I. (2019). A generalized statistical framework to assess mixing ability from incomplete mixing designs using binary or higher order variety mixtures and application to wheat. Field Crops Research, 242, 107571. Link to the data: https://zenodo.org/record/3999270#.YN1-xkyxVZV
The data have been published together with the article. The link to the data is mentioned in the article (https://github.com/cambroise/lme4-adapt-for-variety-mixture

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Table 4. Overview of statistical methods used by project partners

Trial nr.	Partner involved	Crop (or crops)	crop	Type of diversity	Desciptive statistics	Boxplots	ANOVA and post-hoc test	Regression	Factor interaction	LER (land equivalent ratio)	Ranking (genotypes in pure and mixed stand)	Spearman rank correlation	Pearson correlation	Mixing effect for mixtures	Nei diversity index	Other Diversity indexes (shannon, etc)	PCA	Multivariate adaptive regression splines (MARS)	Random Forest	Classification and regression tree (CART)
1	INRA + GQE-Le Moulon	Participatory on-farm breeding for diverse wheat mixtures	winter wheat	intraspecific																
4a	ITAB + INRA Rennes	bread wheat	winter wheat	intraspecific																
5	LBI	spring wheat	srping wheat	intraspecific																
6	AREI	spring barley	barley	intraspecific																
7	IPC	maize landraces, improved populations and CCP	maize	intraspecific																
8	FiBL-CH	lupin CCP	white lupin	intraspecific																
9	AREI	spring wheat x spring pea	pea	interspecific																
11	Cultivari	winter triticale x winter pea (no wheat)	pea	interspecific																
12	LBI	different cereals/pea	pea	interspecific																
13	LBI	spring wheat x faba bean	faba bean	interspecific																
14	AREI	s.wheat/s. triticale x faba bean	faba bean	interspecific																
15	IUNG	mixed cropping spring triticale x blue lupin	blue lupin	interspecific																
16	IUNG	mixed cropping spring oat x blue lupin	blue lupin	interspecific																
17	FiBL-CH	mixed cropping blue lupin - different partners	blue lupin	interspecific																
18	FiBL-CH	mixed cropping white lupin - different partners	white lupin	interspecific																
19	LBI	spring wheat x lupine	white lupin	interspecific																
20	IUNG	mixed cropping spring trirticale x yellow lupin	yellow lupin	interspecific																
21	IUNG	mixed cropping spring oat x yellow lupin	yellow lupin	interspecific																
22	AREI	s. wheat/triticale x s. vetch	vetch	interspecific																
23a	AGROS	grass-legume mixtures	alfalfa	interspecific																
23b	NARDI	grass-legume mixtures	alfalfa	interspecific																
24	IPC	maize x beans	beans	interspecific																
25	INRA	tomato	tomato	interspecific																

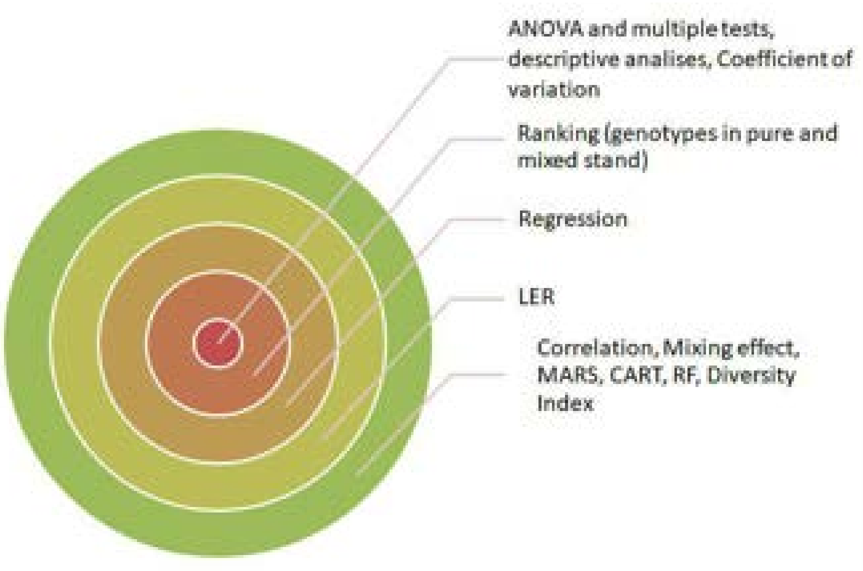

Figure 2. Popularity of statistical methods from central to marginal among LIVESEED partners

Figure 2 above visualises how popular statistical analytical methods were among partners. The centre of the circle represents ANOVA, which was used by most partners as central methodology while extending towards the external circles one finds methodology less used.

1.4.5. Common frame of analysis

The diversity of partners but also the germplasm that they have used provided different sets of data of crop and cultivar characterization. Data of characterization can be organized at crop level (through more diverse cultivars, cultivar mixtures and heterogeneous material), field level (through various types of mixtures of annual and perennial crops) or systems level (by increasing crop number and complexity of its relationships, such as done in agroforestry systems), see Figure 3 below.

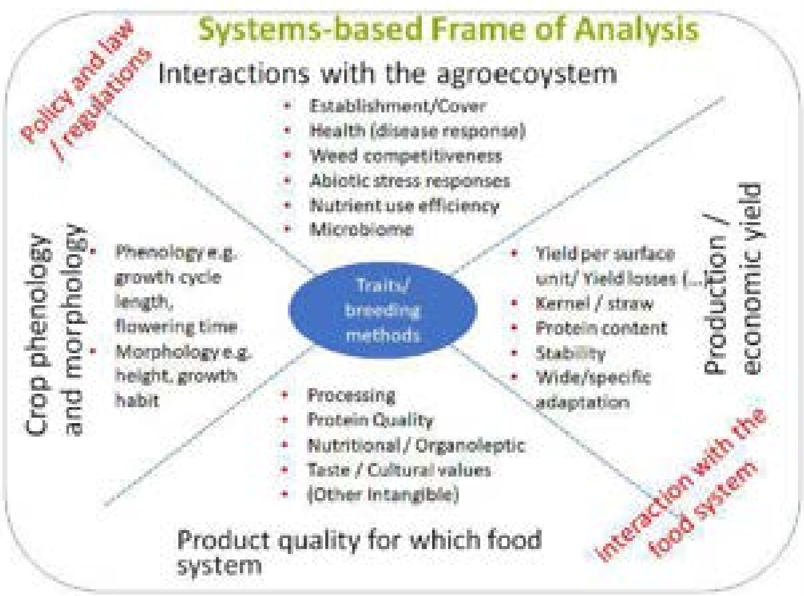

Figure3. Systems-based frame of analysis

Based on the systems-based frame of analysis, the importance of characteristics contributing to breeding for resilience was assessed, based on partners' reports and their validation.

The assessment was based on a scale of numbers associated with colours. In this way, the values were considered:
-2 (Means a negative relationship between a certain trait and increased diversity in **red colour**),
-1 (Means a negative trend between a certain trait and increased diversity in **light red colour**),
0 (Means no or no clear relationship between a certain trait and increased diversity **in yellow**),
1 (Means a positive trend between a certain trait and increased diversity in **light green colour**),
2 (Means a positive relationship between a certain trait and increased diversity **in green colour**).

According to a systems-based frame of analysis, data were grouped into 4 categories:
1) plant descriptors (e.g., uniform phenological states of the different crops using the BBCH scale (Zadoks et al., 1974; Meyer, 2001). The BBCH-scales were developed based on similar growth stages of each plant of different crops correspond to the same code,
2) Agroecosystem performance (e.g., biotic stresses, weed response),
3) Productive performance (e.g., yield and LER), and
4) Quality performance (e.g., baking quality, beta-glucan content in grain).

This approach allowed the identification of discriminating characteristics, some of which will be illustrated by the case studies and will help the breeders in their choices.

In the next pages, Tables 5-13 show the results of this exercise for intra-specific diversity trials and Tables 14-21 for inter-specific diversity trials.

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Table 5. Phenology and morphology results of trials on intra-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Phenology (BBCH stages)									Morphology				
					10 emergence	20 lateral shoots	30 elongation	40 flowerhead formation	50 flowerhead emergence	60 flowering	70 fruit formation	80 fruit ripening	90 harvest	Growth habit	Height	Seedhead characteristics	Uniformity	Other
Trial nr.	**Trial code**	**Partners**	**Trials**	**crop**	**10_E**	**20_LShoot**	**30_Elog**	**40_FHForm**	**50_FHForma**	**60_Fl**	**70_FrutF**	**80_FrutR**	**90_Harv**	**GHab**	**Height**	**SeedHCh**	**U**	**Other**
1	1I_WW	INRA + GQE-Le Moulon	Participatory on-farm breeding for diverse wheat mixtures	winter wheat										0	2	2		2
2	2I_WW	INRA + GQE-Le Moulon	Participatory ideotyping (co-design and assessment) of wheat mixtures	winter wheat											0			
3	3I_WW	UBIOS+INRAE-Le Moulon	Development of winter wheat CCP	winter wheat														
4a	4aI_WW	ITAB + INRA Rennes	COBRA (bread wheat)	winter wheat											0	0		1
4b	4bI_WR	ITAB + INRA Rennes	DOP-rivet	winter rivet wheat	0	0							0	1		0		0
4c	4cI_WW	ITAB + INRA Rennes	DOP-spelt	winter spelt wheat														
4d	4dI_Ot	ITAB + INRA Rennes	DOP-oat	winter oat														
5	5I_SW	LBI	spring wheat	spring wheat									-1	-1	-1			
6	6I_Br	AREI	spring barley mixtures and populations	barley					0					0	0			
7	7I_Z	IPC	maize	maize						0					1	2	0	
8	8I_WL	FiBL-CH	lupin CCP	white lupin	0								0	0	0			

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**). In the group of plant descriptors, it was adapted the BBCH stages for phenology that allowed the comparison of different crops. The third line is the abbreviation of the traits indicated in the second line of the table.

Table 6. Agroecosystem performance results of trials on intra-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Agroecosystem performance									
					Abiotic stress response	Biotic stress response	Biomass production	Establish ment	Establishment companion crop	Cover	Cover companion crop	Standing ability	Vigour	Weed response
Trial nr.	**Trial code**	**LIVESEED Partner involved**	**Trial crops**	**Crop**	AbStressR	BStressR	BiomassP	Estb	EstCompCrop	Cover	CoverComC	StandAb	Vigour	WeedRes
1	1I_WW	INRA + GQE-Le Moulon	Participatory on-farm breeding for diverse wheat mixtures	winter wheat			0					1		
2	2I_WW	INRA + GQE-Le Moulon	Participatory ideotyping (co-design and assessment) of wheat mixtures	winter wheat			0	0		0				0
3	3I_WW	UBIOS+INRAE-Le Moulon	Development of winter wheat CCP	winter wheat										
4a	4aI_WW	ITAB + INRA Rennes	bread wheat	winter wheat		0								
4b	4bI_WR	ITAB + INRA Rennes	DOP-rivet	winter rivet wheat		0		0		1		0		
5	5I_SW	LBI	spring wheat	spring wheat										
6	6I_Br	AREI	spring barley	barley		1				0	0	0	0	0
7	7I_Z	IPC	maize landraces, improved populations and CCP	maize								2		
8	8I_WL	FiBL-CH	lupin CCP	white lupin		-2		0						

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 7. Productive performance results of trials on intra-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Productive performance									
					Yield					**Yield components**				
					Main crop / Cereal	**Companion crop / grain legume**	**Overall intercrop yield**	**Yield stability**	**Mixing efficiency (LER)**	**Seedhead density**	**Seedhead size**	**Seedhead fertility**	**Grain number**	**Grain weight**
Trial nr.	**Trial code**	**LIVESEED Partner involved**	**Trial crops**	**Crop**	**MainC**	**CompC**	**OverallInterCY**	**YS**	**LER**	**SHDens**	**SHSize**	**SHFert**	**GN**	**GW**
1	1I_WW	INRA + GQE-Le Moulon	Participatory on-farm breeding for diverse wheat mixtures	winter wheat	1	na	na	1	1	na	2	2	2	-1
2	2I_WW	INRA + GQE-Le Moulon	Participatory ideotyping (co-design and assessment) of wheat mixtures	winter wheat	1	na	na	1	1	0				
3	3I_WW	UBIOS+INRAE-Le Moulon	Development of winter wheat CCP	winter wheat										
4a	4aI_WW	ITAB + INRA Rennes	Bread wheat	winter wheat	0	na	na		na		0	0		1
4b	4bI_WR	ITAB + INRA Rennes	DOP-rivet	winter rivet wheat		na	na		na					1
4c	4cI_WW	ITAB + INRA Rennes	DOP-spelt	winter spelt		na	na		na					
4d	4dI_Ot	ITAB + INRA Rennes	DOP-oat	winter oat		na	na		na					
5	5I_SW	LBI	Spring wheat	spring wheat	2	na	na		na					
6	6I_Br	AREI	Spring wheat	barley	2	na	na	2	na					
7	7I_Z	IPC	Maize landraces, improved populations and CCP	maize	1	na	na		na		1			2
8	8I_WL	FiBL-CH	Lupin CCP	white lupin	-2	na	na		na					

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 8. Crop quality performance results of trials on intra-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)								
					Baking quality	Beta-glucan content in grain	Crude protein content in grain	Falling number wheat	Grain characteristics (colour, thousand kernel weight)	Grain volume weight	Moisture Maize	Starch content in grain	Zeleny wheat
Trial nr.	**Trial short name**	**LIVESEED Partner involved**	**Trial crops**	**Crop**	**BakingQuality**	**Beta-glucan_G**	**Crude_protein_grain**	**FallingNW**	**GCha**	**GVolWeight**	**Moisture**	**Starch_G**	**ZelenyW**
1	1I_WW	INRA + GQE-Le Moulon	Participatory on-farm breeding for diverse wheat mixtures	winter wheat			0						
2	2I_WW	INRA + GQE-Le Moulon	Participatory ideotyping (co-design and assessment) of wheat mixtures	winter wheat	2		0						
4a	4aI_WW	ITAB + INRA Rennes	bread wheat	winter wheat	0				1				
4b	4bI_WR	ITAB + INRA Rennes	DOP-rivet	winter rivet					0				
5	5I_SW	LBI	spring wheat	spring wheat			0	0					0
6	6I_Br	AREI	spring barley	barley		0	0			0		0	
7	7I_Z	IPC	maize landraces, improved populations and CCP	maize							1		

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 9. Additional information on crop quality performance (crop development, part I.) results of trials on intra-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Crop development									
					Early vigor at tillering	Plant type at tillering	Canopy height at beginning of	Days from sowing to	Days from sowing to	Ground cover of Pea	Ground cover of	Lodging at ripening	Days to flowering	Number of fertile nodes
Trial nr.	**Trial short name**	**LIVESEED Partner involved**	**Trial crops**	**Crop**	EarlyVTill	Plant type at tillering	Canopy height at beginning of stem elongation	Days from sowing to start of heading	Days from sowing to ripening	Ground cover of Pea	Ground cover of Triticale	Lodging at ripening	Days to flowering (FLO).	Number of fertile nodes (FNOD)
1	1I_WW	INRA + GQE-Le Moulon	Participatory on-farm breeding for diverse wheat mixtures	winter wheat										
2	2I_WW	INRA + GQE-Le Moulon	Participatory ideotyping (co-design and assessment) of wheat mixtures	winter wheat	0									
3	3I_WW	UBIOS+INRAE-Le Moulon	Development of winter wheat CCP	winter wheat										
4a	4aI_WW	ITAB + INRA Rennes	Bread wheat	winter wheat										
4b	4bI_WR	ITAB + INRA Rennes	DOP-rivet	winter rivet wheat		1						0		0
4c	4cI_WW	ITAB + INRA Rennes	DOP-spelt	winter spelt wheat										
4d	4dI_Ot	ITAB + INRA Rennes	DOP-oat	winter oat										
5	5I_SW	LBI	Spring wheat	spring wheat										
6	6I_Br	AREI	Spring barley	barley	0	0	0	0	0	0	0	0		
7	7I_Z	IPC	Maize landraces, improved populations and CCP	maize									2	
8	8I_WL	FiBL-CH	Lupin CCP	white lupin										

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 10. Additional information on crop quality performance (crop development, part II.) results of trials on intra-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Crop development II												
Trial nr.	**Trial short name**	**LIVESEED Partner involved**	**Trial crops**	**Crop**	**Height in June**	**Plant Height Maize**	**Plant development lupin, architecture**	**Plant development wheat**	**Plant height**	**Ripening plants**	**Spikes' characteristics**	**Tillering**	**Germination**	**Field emergence lupin**	**Plant height at flowring-lupin**	**Maturation stage 30.7. in Rümikon**	**Percentage of healthy lupin plants in mid June**
1	1I_WW	INRA + GQE-Le Moulon	Participatory on-farm breeding for diverse wheat mixtures	winter wheat					2		1						
2	2I_WW	INRA + GQE-Le Moulon	Participatory ideotyping (co-design and assessment) of wheat mixtures	winter wheat													
3	3I_WW	UBIOS+INRAE-Le Moulon	Development of winter wheat CCP	winter wheat													
4a	4aI_WW	ITAB + INRA Rennes	Bread wheat	winter wheat					0		0						
4b	4bI_WR	ITAB + INRA Rennes	DOP-rivet	winter rivet wheat	0						0	0	0				
4c	4cI_WW	ITAB + INRA Rennes	DOP-spelt	winter spelt wheat													
4d	4dI_Ot	ITAB + INRA Rennes	DOP-oat	winter oat													
5	5I_SW	LBI	Spring wheat	spring wheat				-1	-1	-1							
6	6I_Br	AREI	Spring barley	barley													
7	7I_Z	IPC	Maize landraces, improved populations and CCP	maize		1											
8	8I_WL	FiBL-CH	Lupin CCP	white lupin			0							0	0	0	0

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 11. Additional information on crop quality performance (crop development, part III.) results of trials on intra-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

Crop development results of trials on intra-specific diversity III, in terms of the importance of the characteristics contributing to breeding for resilience.

					Crop development III												
Trial nr.	**Trial short name**	**LIVESEED Partner involved**	**Trial crops**	**Crop**	**Plant vigor**	**Threshing behaviour**	**Precocity**	**plant health**	**Weed biomass**	**Wheat biomass**	**Ear height Maize**	**Uniformity Maize**	**Leaf Angle**	**Tassel Branching Maize**	**Ear placement Maize**	**Root lodging Maize**	**Stalk lodging Maize**
1	1I_WW	INRA + GQE-Le Moulon	Participatory on-farm breeding for diverse wheat mixtures	winter wheat													
2	2I_WW	INRA + GQE-Le Moulon	Participatory ideotyping (co-design and assessment) of wheat mixtures	winter wheat					0	0							
3	3I_WW	UBIOS+INRAE-Le Moulon	Development of winter wheat CCP	winter wheat													
4a	4aI_WW	ITAB + INRA Rennes	Bread wheat	winter wheat	0			0									
4b	4bI_WR	ITAB + INRA Rennes	DOP-rivet	winter rivet wheat	1	0	0	0									
4c	4cI_WW	ITAB + INRA Rennes	DOP-spelt	winter spelt wheat													
4d	4dI_Ot	ITAB + INRA Rennes	DOP-oat	winter oat													
5	5I_SW	LBI	Spring wheat	spring wheat													
6	6I_Br	AREI	Spring barley	barley													
7	7I_Z	IPC	Maize landraces, improved populations and CCP	maize							2	2	2	2	2	2	2
8	8I_WL	FiBL-CH	Lupin CCP	white lupin													

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 13. Additional information on crop quality performance (interactions with the environment) results of trials on intra-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

			Interactionswith the ecosystem																		
Trial short name	**Trial crops**	**Crop**	**Crop ground cover**	**Crop ground cover at tillering**	**Crop ground cover at stem elongation**	**Soil coverage SC**	**Weed cover**	**Weed suppression ability at stem elongation**	**Weed suppression ability at flowering**	**Weed suppression ability at ripening**	**Productive tillering coefficient**	**Loose smut infected plants**	**Net blotch estimation**	**Netblotch AUDPC value**	**Powdery mildew estimation**	**Number of sick plants in May (Anthracnose, lupin)**	**Anthracnose, several dates**	**Powdery mildew AUDPC value**	**Disease's lupin**	**Disease's wheat**	**Bread making test (in 2018)**
1I_WW	Participatory on-farm breeding for diverse wheat mixtures	winter wheat																		0	
2I_WW	Participatory ideotyping (co-design and assessment) of wheat mixtures	winter wheat	0				0													0	2
3I_WW	Development of winter wheat CCP	winter wheat																			
4aI_WW	Bread wheat	winter wheat																		0	
4bI_WR	DOP-rivet	winter rivet wheat	1	1		0	0														
4cI_WW	DOP-spelt	winter spelt wheat																			
4dI_Ot	DOP-oat	winter oat																			
5I_SW	Spring wheat	spring wheat																		2	
6I_Br	Spring barley	barley		0	0			0	0	0	0	0	2	2	0						
7I_Z	Maize landraces, improved populations and CCP	maize																0	0		
8I_WL	Lupin CCP	white lupin														-1	2				

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 14. Phenology and morphology results of trials on inter-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Phenology (BBCH stages)									Morphology				
					10 emergence	20 lateral shoots	30 elongation	40 flowerhead formation	50 flowerhead emergence	60 flowering	70 fruit formation	80 fruit ripening	90 harvest	Growth habit	Height	Seedhead characteristics	Uniformity	Other
Trial nr.	Trial short name	Partners	Trials	crop	10_E	20_LShoot	30_Elog	40_FHForm	50_FHForma	60_Fl	70_FrutF	80_FrutR	90_Harv	GHab	Height	SeedHCh	U	Other
9	9IE_P	AREI	spring wheat x spring pea	pea						0			1		1			
10	10IE_P	FiBL-CH-GZPK	spring triticale/barely - pea	pea						1					1			
11	11IE_P	Cultivari	winter triticale x winter pea (no wheat)	pea					0	0			2	2	0			
12	12IE_P	LBI	different cereals/pea	pea									2	2	2			
13	13IE_FB	LBI	spring wheat x faba bean	faba bean									2	2	2			
14	14IE_FB	AREI	spring wheat/s. triticale x faba bean	faba bean														
15	15IE_BL	IUNG	mixed cropping spring triticale x blue lupin	blue lupin	0	0			0									
16	16IE_BL	IUNG	mixed cropping spring oat x blue lupin	blue lupin	0	0			0									
17	17IE_BL	FiBL-CH	mixed cropping blue lupin - different partners	blue lupin	2													
18	18IE_WL	FiBL-CH	mixed cropping white lupin - different partners	white lupin	2													
19	19IE_WL	LBI	spring wheat x lupine	white lupin														
20	20IE_YL	IUNG	mixed cropping spring triticale x yellow lupin	yellow lupin									2	2	2			
21	21IE_YL	IUNG	mixed cropping spring oat x yellow lupin	yellow lupin	0	0			0									
22	22IE_V	AREI	spring wheat/triticale x spring vetch	vetch	0	0			0									
23a	23aP_Gss	AGROS	grass-legume mixtures	alfalfa						0								
23b	23bP_Gss	NARDI	grass-legume mixtures	alfalfa										0				0
24	24P_Ph	IPC	maize x beans	beans										0				0
25	25SAF	INRA	tomato	tomato														
26		IPC	successional agroforestry early selection of maize under coevolution, no results still	maize														
26	26SAF	IPC	maize CCP: To develop a maize CC adapted to a successional agroforestry experiment	beans														

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**). In the group of plant descriptors, it was adapted the BBCH stages for phenology that allowed the comparison of different crops. The third line is the abbreviation of the traits indicated in the second line of the table.

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Table 15. Agroecosystem performance results of trials on inter-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Agroecosystem performance									
					Abiotic stress response	Biotic stress response	Biomass production	Establishment	Establishment companion crop	Cover	Cover companion crop	Standing ability	Vigour	Weed response
Trial nr.	**Trial short name**	**Partners**	**Trials**	**crop**	AbStressR	BStressR	BiomassP	Estb	EstCompCrop	Cover	CoverComC	StandAb	Vigour	WeedRes
9	9IE_P	AREI	spring wheat x spring pea	pea						2		1		
10	10IE_P	FiBL-CH-GZPK	spring triticale/barely - pea	pea								1		
11	11IE_P	Cultivari	The	pea	0					0		2	0	2
12	12IE_P	LBI	different cereals/pea	pea		2								
13	13IE_FB	LBI	spring wheat x faba bean	faba bean		2								
14	14IE_FB	AREI	spring wheat/s. triticale x faba bean	faba bean						1				
15	15IE_BL	IUNG	mixed cropping spring triticale x blue lupin	blue lupin				0					0	
16	16IE_BL	IUNG	mixed cropping spring oat x blue lupin	blue lupin				0					0	
17	17IE_BL	FiBL-CH	mixed cropping blue lupin - different partners	blue lupin		0		2	1	2		0		
18	18IE_WL	FiBL-CH	mixed cropping white lupin - different partners	white lupin		0		-2	-1	0	-1	0		
19	19IE_WL	LBI	spring wheat x lupine	white lupin		2								
20	20IE_YL	IUNG	mixed cropping spring tritricale x yellow lupin	yellow lupin				0					0	
21	21IE_YL	IUNG	mixed cropping spring oat x yellow lupin	yellow lupin				0					0	
22	22IE_V	AREI	spring wheat/triticale x spring vetch	vetch						0				
23a	23aP_Gss	AGROS	grass-legume mixtures	alfalfa		0							0	
23b	23bP_Gss	NARDI	grass-legume mixtures	alfalfa		0							0	
24	24P_Ph	IPC	maize x beans	beans										
25	25SAF	INRA	tomato	tomato										
26		IPC	successional agroforestry early selection of maize under coevolution, no results	maize										
26	26SAF	IPC	maize CCP: To develop a maize CC adapted to a successional agroforestry experiment	beans										

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in light red colour), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in light green colour), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 16. Productive performance results of trials on inter-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Productive performance									
					Yield					Yield components				
					Main crop / Cereal	Companion crop / grain legume	Overall intercrop yield	Yield stability	Mixing efficiency (LER)	Seedhead density	Seedhead size	Seedhead fertility	Grain number	Grain weight
Trial nr.	**Trial short name**	**Partners**	**Trials**	**crop**	**MainC**	**CompC**	**OverallInterCY**	**YS**	**LER**	**SHDens**	**SHSize**	**SHFert**	**GN**	**GW**
9	9IE_P	AREI	spring wheat x spring pea	pea	-1				na	1			0	0
10	10IE_P	FiBL-CH-GZPK	spring triticale/barely - pea	pea	1	1	1		na					1
11	11IE_P	Cultivari	winter triticale x winter pea (no wheat)	pea	2			2	na					2
12	12IE_P	LBI	different cereals/pea	pea	2	2	2		na					
13	13IE_FB	LBI	spring wheat x faba bean	Faba bean	2	2	2							
14	14IE_FB	AREI	spring wheat/s. triticale x faba bean	Faba bean	-1				na					0
15	15IE_BL	IUNG	mixed cropping spring triticale x blue lupin	blue lupin	2	2	2		2					
16	16IE_BL	IUNG	mixed cropping spring oat x blue lupin	blue lupin	2	2	2		2					
17	17IE_BL	FiBL-CH	mixed cropping blue lupin - different partners	blue lupin	1	1	1							
18	18IE_WL	FiBL-CH	mixed cropping white lupin - different partners	white lupin	-1	0	0							
19	19IE_WL	LBI	spring wheat x lupine	white lupin	2	2	2							
20	20IE_YL	IUNG	mixed cropping spring tirticale x yellow lupin	yellow lupin	2	2	2		2					
21	21IE_YL	IUNG	mixed cropping spring oat x yellow lupin	yellow lupin	2	2	2		2					
22	22IE_V	AREI	spring wheat/triticale x spring vetch	vetch	-1				na					0
23a	23aP_Gss	AGROS	grass-legume mixtures	alfalfa					na					
23b	23bP_Gss	NARDI	grass-legume mixtures	alfalfa					na					
24	24P_Ph	IPC	maize x beans	beans	2		2							
25	25SAF	INRA	tomato	tomato					na					

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 17. Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial) results of trials on inter-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)						
					Beta-glucan content in grain	Crude protein content in grain	Falling number wheat	Grain volume weight	Starch content in grain	Thousand grain weight	Zeleny wheat
Trial nr.	**LIVESEED Partner involved**	**Trial short name**	**Trial crops**	**Crop**	**Beta-glucan_G**	**Crude_protein_grain**	**FallingNW**	**GVolWeight**	**Starch_G**	**ThousandGW**	**ZelenyW**
9	9IE_P	AREI	spring wheat x spring pea	pea		0					
10	10IE_P	FiBL-CH-GZPK	spring triticale/barely - pea	pea		2					
11	11IE_P	Cultivari	winter triticale x winter pea (no wheat)	pea		2					
12	12IE_P	LBI	different cereals/pea	pea	0	2		0	0	2	
13	13IE_FB	LBI	spring wheat x faba bean	faba bean		2					
14	14IE_FB	AREI	spring wheat/s. triticale x faba bean	faba bean		2	2				2
15	15IE_BL	IUNG	mixed cropping spring triticale x blue lupin	blue lupin		0					
20	20IE_YL	IUNG	mixed cropping spring trirticale x yellow lupin	yellow lupin		2					

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 18. Additional information on crop quality performance (crop development, part I.) results of trials on inter-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Crop development I													
Trial nr.	Trial code	Partners	Trials	crop	Early vigor at tillering EarlyVTill	Plant type at tillering	Canopy height at beginning of stem elongation	Days from sowing to full emergence	Days from sowing to tillering (2-3 tillers detectable)	Days from sowing to start of heading	Days from sowing to end of heading (cereals)	Days from sowing to ripening	Lodging at ripening	Soil coverage SC	Days to flowering (FLO).	Plants: length (PLG)	Number of fertile nodes (FNOD)	Winter hardiness
9	9IE_P	AREI	spring wheat x spring pea	pea									1	2	0	1	0	
11	11IE_P	Cultivari	winter triticale x winter pea	pea	0	2	0			0		2	2					0
14	14IE_FB	AREI	spring wheat/spring triticale x faba bean	faba bean										1	0			
15	15IE_BL	IUNG	mixed cropping spring triticale x blue lupin	blue lupin				0	0		0							
16	16IE_BL	IUNG	mixed cropping spring oat x blue lupin	blue lupin				0	0		0							
17	17IE_BL	FiBL-CH	blue lupin - different partners	blue lupin									0	2				
18	18IE_WL	FiBL-CH	mixed cropping white lupin - different partners	white lupin									0	0				
20	20IE_YL	IUNG	mixed cropping spring tirticale x yellow lupin	yellow lupin				0	0		0							
21	21IE_YL	IUNG	mixed cropping spring oat x yellow lupin	yellow lupin				0	0		0							
22	22IE_V	AREI	spring wheat/triticale x spring vetch	vetch										0	0			

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 19. Additional information on crop quality performance (crop development, part II.) results of trials on inter-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Crop development II									
Trial nr.	**Trial short name**	**Partners**	**Trials**	**crop**	**Plant height of pea at beginning of shooting of triticale**	**Beginning of flowering of pea**	**End of flowering of pea**	**Plant length at beginning of flowering**	**Plant length at end of pea flowering**	**Height in April**	**Plant Height Maize**	**Plant development lupin, architect**	**Plant development wheat**	**Plant height**
10	10IE_P	FiBL-CH-GZPK	spring triticale/barely - pea	pea	1	1	1	1						
11	11IE_P	Cultivari	winter triticale x winter pea (no wheat)	pea	0			0	0	0				
12	12IE_P	LBI	different cereals/pea	pea								2	2	2
13	13IE_FB	LBI	spring wheat x faba bean	faba bean								2	2	2
19	19IE_WL	LBI	spring wheat x lupine	white lupin							2	2	2	2

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 20. Additional information on crop quality performance (crop development, part III.) results of trials on inter-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Crop development III									
Trial nr.	**Trial short name**	**Partners**	**Trials**	**crop**	**Field emergence lupin**	**Field emergence companion crop**	**Lodging at beginning of pea flowering**	**Lodging at end of pea flowering**	**Plant vigor**	**leaf-to-stem ratio**	**stem diameter**	**plant habitus**	**Disease resistance (downy mildew)**	**Date of ear emergence**
10	10IE_P	FiBL-CH-GZPK	spring triticale/barely - pea	pea			1	1						
11	11IE_P	Cultivari	winter triticale x winter pea (no wheat)	pea										0
17	17IE_BL	FiBL-CH	mixed cropping blue lupin - different partners	blue lupin	2	1								
18	18IE_WL	FiBL-CH	mixed cropping white lupin - different partners	white lupin	2	-1								
23a	23aP_Gss	AGROS	grass-legume mixtures	grass					0	0	0	0	0	
23b	23bP_Gss	NARDI	grass-legume mixtures	grass					0	0	0	0		

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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Table 21. Additional information on crop quality performance (interactions with the environment) results of trials on inter-specific diversity, in terms of the importance of the characteristics contributing to breeding for resilience.

					Interactionswith the ecosystem																			Microbiome structuration			
Trial nr.	**Trial short name**	**Partners**	**Trials**	**crop**	**Crop ground cover at tillering**	**Crop ground cover at stem elongation**	**Soil coverage SC**	**Weed suppression ability at stem elongation**	**Weed suppression ability at flowering**	**Weed species composition and abundance (In 2018)**	**Weed suppression ability at ripening**	**Productive tillering coefficient**	**Loose smut infected plants**	**Net blotch estimation**	**Netblotch AUDPC value**	**Powdery mildew estimation**	**Crown and root rot**	**Anthracnose, several dates**	**Powdery mildew AUDPC value**	**Disease's lupin**	**Disease's wheat**	**Diseases faba/ pea**	**Drought tolerance**	**Farm**	**Year**	**Agroforestrial modality**	**Tomato variety**
9	9IE_P	AREI	spring wheat x spring pea	pea			2																				
10	10IE_P	FiBL-CH-GZPK	spring triticale/barely - pea	pea																			1				
11	11IE_P	Cultivari	winter triticale x winter pea (no wheat)	pea	0	0		2	2		0	0	0	0	0	0			0								
12	12IE_P	LBI	different cereals/pea	pea																	2	2					
13	13IE_FB	LBI	spring wheat x faba bean	faba bean																	2	2					
14	14IE_FB	AREI	spring wheat/spring triticale x faba bean	faba bean			1																				
15	15IE_BL	IUNG	mixed cropping spring triticale x blue lupin	blue lupin						0																	
16	16IE_BL	IUNG	mixed cropping spring oat x blue lupin	blue lupin						0																	
17	17IE_BL	FiBL-CH	mixed cropping blue lupin - different partners	blue lupin			1				1							0									
18	18IE_WL	FiBL-CH	mixed cropping white lupin - different partners	white lupin			0											0									
19	19IE_WL	LBI	spring wheat x lupine	white lupin																2	2						
20	20IE_YL	IUNG	mixed cropping spring trirticale x yellow lupin	yellow lupin						0																	
21	21IE_YL	IUNG	mixed cropping spring oat x yellow lupin	yellow lupin						0																	
22	22IE_V	AREI	spring wheat/triticale x spring vetch	vetch			0																				
23a	23aP_Gss	AGROS	grass-legume mixtures	grass												0	0										
23b	23bP_Gss	NARDI	grass-legume mixtures	grass												0	0										
25	25SAF	INRA	tomato	tomato																				1	2	2	1

The values were considered: -2 (a negative relationship between a certain trait and increased diversity in **red colour**), -1 (a negative trend between a certain trait and increased diversity in **light red colour**), 0 (no or no clear relationship between a certain trait and increased diversity **in yellow**), 1 (a positive trend between a certain trait and increased diversity in **light green colour**), 2 (a positive relationship between a certain trait and increased diversity **in green colour**).

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1.4.6. Overview of the comparative analysis of the traits

To provide a comparative analysis of the 4 categories of data (presented in the overview tables and tables 6-21), we have used the circular visualization by using the program CIRCOS (Krzywinski et al., 2009). CIRCOS application turns the data tables into chords diagrams as it is referred in Figure 4 below plus its interpretation. The original data (-2, -1, 0, 1, 2) were converted respectively (1, 25, 50, 75, 100) to facilitate the analyses by associating a negative relationship, negative trend, absent or no clear trend or relationship, positive trend, or positive relationship with the different and detectable widths of the cords. The analyses started with the type of diversity (intraspecific and interspecific), then a graph was shown with and without “absent or no clear values”, i.e., the value 50. Finally, we grouped some of the most often tested species such as winter wheat, pea, and lupins.

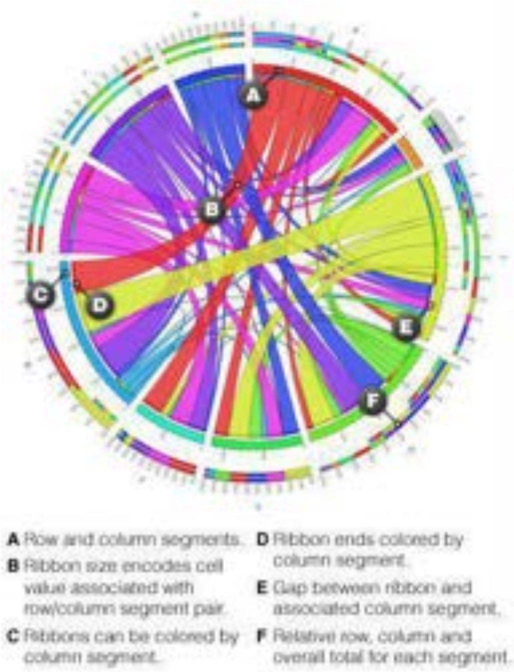

Figure 4. Visualization in CIRCOS diagrams and its interpretation[4].

To fit the CIRCOS graphics, we have developed the following nomenclature (Table 22).

Table 22. Nomenclature of crops and LIVESEED trials

Abbreviations	Meaning
CROPS	
WW	winter wheat
WR	rivet wheat
Ot	oat
SW	spring wheat
Br	barley
Z	maize
WL	white lupin

[4] CIRCOS table viewer - circular visualization of tabular data // with CIRCOS - circular genome data visualization (bcgsc.ca): http://mkweb.bcgsc.ca/tableviewer/visualize/

P	pea
FB	faba bean
BL	blue lupin
YL	yellow lupin
V	vetch
Gss	grass
Ph	beans
SAFs	successional agroforestry systems
TRIALS	
I	Intraspecific trial
1I_WW	Trial #1 winter wheat INRA + GQE-Le Moulon (Isabelle G.) (Participatory on-farm breeding for diverse wheat mixtures)
2I_WW	Trial #2 in winter wheat INRA + GQE-Le Moulon (Isabelle G.) (Participatory ideotyping (co-design and assessment) of wheat mixtures)
3I_WW	Trial #3 in winter wheat UBIOS+INRAE-Le Moulon (Development of winter wheat CCP)
4aI_WW	Trial #4a in bread wheat, w. wheat ITAB + INRA Rennes (DOP)
4bI_WR	Trial #4b in w. rivet wheat ITAB + INRA Rennes (DOP)
4cI_WW	Trial #4c in spelt, w. wheat ITAB + INRA Rennes (DOP)
4dI_Ot	Trial #4d in oat ITAB + INRA Rennes (DOP)
5I_SW	Trial #5 in spring wheat LBI
6I_Br	Trial #6 in spring barley AREI
7I_Z	Trial #7 in maize IPC
8I_WL	Trial #8 in white lupin CCP FIBL-CH
IE	Interspecific trials
9IE_P	AREI spring wheat x spring pea
10IE_P	GZPK+FiBL spring triticale/barley-pea, pea
11IE_P	CULTIVARI, winter triticale x winter pea
12IE_P	LBI, different cereals/pea, pea
13IE_FB	LBI, spring wheat x faba bean
14IE_FB	AREI, spring wheat/spring triticale x faba bean
15IE_BL	IUNG, mixed cropping spring triticale x blue lupin
16IE_BL	IUNG, mixed cropping spring oat x blue lupin
17IE_BL	FiBL-CH, blue lupin - different partners
18IE_WL	FiBL-CH, mixed cropping white lupin - different partners
19IE_WL	LBI, spring wheat x lupine, white lupin
20IE_YL	IUNG, mixed cropping spring triticale x yellow lupin
21IE_YL	IUNG, mixed cropping spring oat x yellow lupin
22IE_V	AREI, spring wheat/triticale x spring vetch
23aP_Gss	AGROS grass-legume mixtures
23bP_Gss	NARDI grass-legume mixtures
24P_Ph	IPC, maize x beans
25SAF	INRAE agroforestry - tomato
26SAF	IPC successional agroforestry early selection of maize under coevolution (no results yet)

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The traits used were also abbreviated to be presented in the graphic as indicated below (Table 23). To uniformize the plant descriptors used for the crops in the LIVESEED trials, we have used BBCH scale that is a system for a uniform coding of phenologically similar growth stages of all mono- and dicotyledonous plant species (Meyer, 2001). The BBCH-scales were developed based on similar growth stages of each plant of different crops correspond to the same code.

Table 23. Nomenclature of traits used

Abbreviations	Meaning
	PLANT DESCRIPTORS
10_E - 10 (emergence)	the principal stage 1 corresponds to leaf development (main shoot)
20_LShoot - 20 (lateral shoots)	the principal growth stage 2 corresponds to the formation of side shoots/tillering
30_Elog - 30 (elongation),	the principal growth stage 3 corresponds to the Stem elongation or rosette growth, shoot development (main shoot)
40_FHForm - 40 (flowerhead formation)	the principal growth stage 4 corresponds to the Development of harvestable vegetative plant parts or vegetatively propagated organs/booting (main shoot)
50_FHForma - 50 (flowerhead emergence)	the principal growth stage 5 corresponds to the Inflorescence emergence (main shoot)/heading
60_Fl - 60 (flowering)	the principal growth stage 6 corresponds to Flowering (main shoot) Other – it indicates some specific trait described in a trial
70_FrutF - 70 (fruit formation)	the principal growth stage 7 corresponds to development of fruit
80_FrutR - 80 (fruit ripening)	the principal growth stage 8 corresponds to Ripening or maturity of fruit and seed
90_Harv - 90 (harvest)	the principal growth stage 9 corresponds to Senescence, beginning of dormancy
GHab	Growth habit referring to plant habitus corresponds to erect to ascending and prostrate or decumbent behaviour in grass-legume mixtures, specially to trials 23a and 23b for AGROS and NARDI
Height	Plant height corresponds for most of the partner the height at flowering, corresponding to the end of the vegetative growth. In addition, Height also corresponds both to Height in April for trial 4b. ITAB + INRA Rennes DOP-rivet and trial 11 CULTIVARI winter triticale x winter pea. The trial 11 includes Plant Height, Height in April, and June because the value obtained is the same in all of them.
SeedHCh -	Seedhead characteristics such as awn, colour, and shape
U	Uniformity, it indicates the level of uniformity in the germplasm evaluated or characterized
	AGROECOSYSTEM PERFORMANCE
AbStressR	Abiotic stress response corresponding to winter hardiness and drought tolerance
BStressR	Biotic stress response it includes Loose smut infected plants Net blotch estimation and AUDPC value, powdery mildew estimation also with AUDPC, number of sick plants in May (anthracnose, lupin), crown and root rot, anthracnose monitored in several dates, and other diseases on lupin, wheat, faba bean, pea.
BiomassP	Biomass production (e.g., crop biomass produced at GS 20-25 rated from 1=low to 9=very high (Spring Barley for AREI) or sampling of 50cm x 50cm surfaces for wheat Trial 1)
Estb	Establishment, generally associated with days from sowing to full emergence

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EstCompCrop	Establishment companion crop: can be important to at identify suitable companion crops for mixed cropping and/or undersowing (e.g., Trial 17 and 18 mixed cropping of lupin with a cereal cash-crop or with an undersowing companion crop may reduce weed pressure. Mixed cropping and undersowing may reduce anthracnose pressure through the barrier that the companion crop poses to secondary infection.
Cover	Soil cover can be determined by visual observation in scores 1- poor soil coverage, 9 complete soil coverage (e.g., beginning of pea flowering stage (216-226 decimal code for the growth stages of peas by UPOV)); score 0: not covering; 5: very covering (e.g. in Trial 4a-d); visual assessment using ITAB scoring scale of % of covered soil (e.g., Trial 1); from 1 (very weak) to 5 (very strong) (e.g., Trial 5)
CoverComC	Cover companion crop aimed at identifying suitable companion crops for mixed cropping and/or undersowing, the scale used was de % of covered soil (e.g., blue, and white for Trial 17 and 18 FIBL-CH)
StandAb	Standing ability can also be measured by the trait lodging in several phases (e.g., on pea from the beginning to the end of flowering and ripening) and considering the stalk or root lodging (e.g., on maize)
Vigour	Vigour included Early vigour at tillering BBCH GS 31-39 (Trial 6) and Canopy height at beginning of stem elongation BBCH GS 31-39 and plant vigour. The scales used were 0: not vigorous, 5: very vigorous (Trial 4a to d) and a scale from 1 = high to 9 = low (Trial 23a to 23b)
WeedRes	The weed response includes weed biomass (weight of dry matter, species identification, Trial 1), weed cover of soil after harvest (only blue lupins on trial 17) (a Semi-quantitative scale was used to score 1-9), weed suppression ability at stem elongation BBCH GS 31-39, was calculated on the average weed ground cover estimated in % on plot level. Weed suppression ability for each crop genotype (Svar) was calculated as the difference between weed growth in plots for each genotype (Wvar) and the maximum weed growth (Wmax) from adjacent uncropped plot and expressed as a percentage: Svar = ((Wmax-Wvar)/Wmax) * 100 [Hoad et al., 2008] (trial 6). The weed suppression ability at flowering BBCH GS 59-65 was calculated on the basis of average weed ground cover estimated in % on plot level at crop the same methodology was used for weed suppression ability at ripening GS 87-92 for trial 6. For peas a better suppression ability was found with leafed varieties however they tend to lodge (Trial 9). Intercropping with white lupins had also a weed suppression (trial 19) can be also applied. In addition, the weed community can also represent Weed species composition and abundance.
PRODUCTION PERFORMANCE	
MainC	Main crop/cereal - it corresponds to the grain yield of the main crop if per se or to the cereal in a crop mixture
CompC	Companion crop/grain legume - it corresponds to the grain yield of the crop that is in the mixture, generally grain legume.
OverallInterCY	Overall intercrop yield, provides the global yield
YS	yield stability was calculated according to regression analysis and ranking using data from nº growing seasons (nº environments) (trial 6).
LER	Land equivalent ratio allows to evaluate the mixing efficiency. It is calculated by (crop 1 in mixture/crop 1 per se) + (crop 2 in mixture / crop 2 per se)
SHDens	Seedhead density indicates the number of spikes per m^2
SHSize	Seedhead size - is the spike length average, having the basis of the numbers of kernels per row
SHFert	Seedhead fertility indicates the number and weight of grains per spike and can be also considered as weight of grains per spike.
GN	Grain number
GW	Grain weight is the average grain weight, considering 100 or 1000 kernel weight
QUALITY PERFORMANCE	
GCha	Grain characteristics (colour, thousand kernel weight)
Moisture	Moisture of grain

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Crude_protein_grain	Crude protein content in grain, estimated in percentage with Near infrared Spectroscopy (NIRS) in trial 11 on trial 6 it was used INFRATEC 1241 grain analyser and data were expressed in % in dry matter
FallingNW	Falling number wheat
ZelenyW	Zeleny wheat
Starch_G	Starch content in grain (analysed by INFRATEC 1241 grain analyser for trial 6)
Beta-glucan_G	Beta-glucan content in grain spring barley (analysed by INFRATEC 1241 grain analyser for trial 6)
GVolWeight	Grain volume weight (analysed by INFRATEC 1241 grain analyser for trial 6)
BakingQuality	Baking quality
ThousandGW	Thousand grain weight

1.5. Results

1.5.1. Main outcomes of the experimental trials

The main outcomes regarding research on breeding for **intra-specific diversity** existing in cultivar/genotype mixtures were:

(1) An on-farm study on 28 mixtures developed by the farmers, with 2-16 components, tended to confirm the interest of mixtures. Response to farmers' mass selection was in general positive for productivity traits and was larger when selecting within mixtures then within components before mixing. Farmers' mass selection did not lead to a loss of phenotypic diversity within three years (Trial 1, winter wheat, INRAE-F).

(2) Using an ideotyping (2I_WW, winter wheat, INRAE) participatory approach based on workshops with a group of organic farmers from the Ile-de-France region, assembly rules and farmers' variety mixtures of 3 to 5 winter wheat varieties tailored to their local context were co-designed. The cultivar mixtures of 3-5 components showed a reduction of the risks of yield losses and a stable protein content compared with pure stands. The three-year on-farm agronomical assessment of these co-designed mixtures tends to confirm the interest of mixtures, especially in stressing conditions such as in organic systems.

(3) The cultivation of spring wheat cultivar mixtures in Trial 5 in the Netherlands had no positive effect on yield. A mixture consisting of four commercial spring wheat varieties stood out compared to the pure stand varieties for total yield. However, the yield of mixed treatments was only significantly higher compared to the lowest yielding variety in pure stand. Mixtures of two varieties showed in almost all cases no significant differences with the varieties in pure stand for yield. Whether cultivar mixtures lead to a higher resilience is not confirmed in this trial, due to the very low disease pressure (Trial 5, spring wheat mixtures, LBI-NL);

(4) Cultivar mixtures can provide advantages over growing the components in pure stand in respect to leaf diseases incidence and to lesser extent in terms of yield and weed suppression ability increase. The largest mixing effect for leaf diseases in barley cultivar mixtures in Latvia was found for net blotch. For this disease a reduction in incidence was found for 7 out of 8 tested mixtures. Smaller effects, spanning from positive to negative among mixtures and environments were detected for yield and competitive ability against weeds. From the experience in this trial, it is possible to obtain stable and good yielding mixtures by mixing components with distinctive disease resistance, yield potential and adaptability but it is not true for all cases. The strategy to combine mixtures with components differing for three target traits did not ensure expected effect in all cases and therefore cannot be generalized.

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Stable mixtures arose by combining components with different adaptability, broad adaptability, and adaptability to lower yielding environments, but not in all cases (Trial 6, Spring barley, AREI, LV).

Research on different types of populations provided the following outcomes:

(1) Comparing two population development strategies, Composite Cross Populations (generated by crossings) and Dynamic populations (generated by mixtures), no difference was observed in terms of their internal diversity. It was found that human selection has a greater impact on agronomical characteristics (phenotypic characteristics and yield components) than the breeding methods used to create the new populations and provided a positive impact on some agronomical characteristics and did not seem to reduce the global intra-varietal diversity (Trial 4a, 4aI_WW, ITAB, winter wheat);

(2) Around 30 different Diversity Oriented Populations (DOPs) were created from mixtures of several accessions from a pool of nearly 200 accessions and were tested by 35 farmers. Farmers are interested in this type of populations because they may choose relevant criteria answering to their conditions and market. According to farmers, in their fields the main traits of the populations correspond to the criteria they requested among the genetic resources (Trial 4b, winter rivet wheat ITAB + INRA Rennes, DOP-rivet, winter rivet wheat).

(3) The spring wheat populations tested in the Netherlands showed slightly lower yields compared to commercial cultivars and the question. The disease pressure was very low in the trial years, hence the question if populations have a higher biotic stressors resilience could not be answered (Trial 5, spring wheat populations, LBI, NL);

(4) Composite Cross Populations (CCPs) with higher genetic diversity ensured better yield performance and stability compared to those with lower diversity level. CCPs were stable yielding over different environments and provided comparable yield results to check varieties under organic farming and stress conditions. The diversity in phenotypic traits of organically cultivated sub-populations was slightly larger compared to the same original CCPs cultivated in conventional conditions in the case of more complex populations, but not for less diverse populations cultivated under the respective farming system for longer period (Trial 6, spring barley, AREI, LV);

(5) No effect of complete homogeneity of doubled haploid (DH) barley lines on adaptability was found. However, they yielded on average lower than the initial advanced lines (Trial 6, spring barley, AREI, LV).

(6) Testing a wide range of maize populations in low input organic conditions versus conventional systems increased farmers awareness of the availability of germplasm that can be used to increase agro-biodiversity and resilience of their ecosystem. One traditional maize population and one CCP had shown yield potential and stability across environments to be used in organic farming for gastronomic purposes and animal feeding (Trial 7, 7I_Z, IPC, maize).

(7) The white lupin Composite Cross Population developed by FiBL showed a decrease in yield, mainly due to the increasing anthracnose susceptibility in the population. Little information on anthracnose resistance of the parental lines was known at the moment of the crosses in 2015. This result showed that resilience through genetic diversity can only be achieved for white lupin in a genetic background containing considerable resistance for anthracnose that is the most important biotic stress for this crop in Swiss growing conditions. Outcrossing with bitter plants and selection for sweet plants probably enhanced the population breakdown of anthracnose resistance. However, these results should not lead to the conclusion that populations cannot be used for white lupin. In general, the

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development of composite cross populations can be worth in white lupin if based on parents with strong tolerance to anthracnose and with the same mutation for low-alkaloid content (Trial 8, white lupin, FIBL-CH).

The outcomes of research on breeding for **inter-specific diversity** were described by partners as following:

(1) Ranking of semi-leafless pea genotypes in a mixture with wheat and in the pure stand was similar. Leafed tall pea genotypes were more suitable for growing in a mixture with wheat under organic conditions than short genotypes. Adding of 20% of spring wheat to the sowing rate of semi-leafless pea genotypes, without the reduction of pea sowing rate, could be a viable option for growing of semi-leafless varieties under organic conditions. That would allow to increase weed suppressive ability by better soil coverage and to maintain the high productivity of pea. Growing field beans in mixture with spring cereals enhanced the soil coverage and could help to cope with weeds pressure. The species of the support crop did not have an impact on yield level of field beans. Field beans could be the best solution for spring vetch growing in mixture, to provide high seed yield under organic conditions for Trial 9 (spring wheat x spring pea, AREI-LV), Trial 14 (spring wheat/s. triticale x faba bean, AREI-LV) and Trial 22 (spring wheat/triticale x spring vetch, AREI-LV);

(2) Barley proved to be the best mixing partner for spring peas, providing stability against lodging and suppressing weeds, while ripening at the same time. Many pea gene bank accessions were found to have interesting traits, revealing accessions with traits valuable for future agriculture (Trial 10, spring triticale or barley/ spring pea, FIBL-CH-GZPK);

(3) To breed for better mixing ability, a medium to tall normal-leafed winter pea will be the first choice as result of the trials conducted in LIVESEED and a high yielding triticale with good compensation should be the one to combine with in yield trials for pea selection. As triticale has such an influence on the total yield, it should be given more research on more different varieties of triticale to be combined with medium to tall peas to increase the mixed yield with a focus on the pea share and the compensation ability of the triticale. In trials for searching better triticale genotypes for mixtures, a special focus should be given to morphological characters like broadness of leaves, date of ear emergence and rapidness of juvenile growth, plant length and ears per square meter to develop a 'triticale mixture type' (Trial 11, winter triticale/ winter pea, CULTIVARI-DE);

(4) Mixed stands of cereals with pea showed a higher yield compared to the yield of pure stand pea and pure stand cereal. Some mixed treatments showed more suppression of weeds compared to the pure stand pea treatments, especially the mixed treatment with barley and oats (Trial 12, different cereals/pea, LBI-NL);

(5) Mixed faba bean/wheat treatments showed in almost all cases a significant higher total yield compared to the pure stand faba bean treatments and some mixtures showed also a significantly higher total yield compared to the pure stand wheat treatments. Wheat in mixed cropping conditions had a significant higher protein content, compared to wheat in pure stand. The mixed faba bean/wheat treatments showed (for most treatments) significantly higher wheat plants compared to the pure stand wheat treatments and significantly lower faba bean plants compared to the pure stand faba bean treatments. (Trial 13, spring wheat/ faba bean, LBI, NL);

(6) Mixed cropping of blue lupins with oat or triticale is now recommended to farmers given the positive outcome of the trials in Switzerland, while for white lupins mixed cropping had no advantage under spring sowing conditions in Switzerland. It is difficult to find suitable companion crops and

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undersowing subsidiary crops for white lupin due to long vegetation period. Strong vigour of white lupins in spring - especially under drought conditions - and early canopy coverage due to larger leaf area compared to small leaved blue lupins resulted in suppression of less drought tolerant companion crops (Trial 17&18, respectively for blue lupin and white lupin/ different cereals or red fescue FIBL-CH);

(7) The high values of land equivalent ratio (LER) obtained for mixtures of oat/triticale with lupins demonstrated their good mixing ability. Mixtures of lupins with oat performed better (higher yields and lower variability in years) than mixtures with spring triticale in Poland. Yellow lupin in a mixture performed better than blue lupin (Trial 15, mixed cropping spring triticale x blue lupin, IUNG, PL; Trial 16, mixed cropping spring oat x blue lupin, IUNG-PL; Trial 20, spring triticale x yellow lupin), Trial 21 (spring oat x yellow lupin, IUNG-PL);

(8) Mixed treatments of spring wheat with white lupin showed a significantly higher total yield compared to the pure stand lupin treatments but no significant differences in total yield compared to the pure stand wheat treatments were found. Mixing of crops was found to be profitable. Wheat in mixed cropping conditions had a significantly higher protein content, compared to wheat in pure stand. The number of pods on the main stem was significantly lower in the mixed stands, compared to the pure stand lupin crop. General mixing ability (GMA) effect for lupin genotype was significant for lupin (direct effect) and wheat yield (indirect effect) but GMA effect for wheat genotype is only significant for wheat yield (direct), but not for lupin yield (indirect) and GMA effect for total yield is only significant for wheat genotype and not for lupin genotype. No significant specific mixing ability (SMA) effect was found (no significant wheat genotype*lupin genotype interaction) (Trial 19, spring wheat x white lupin, LBI, NL);

(9) Selection decisions for alfalfa in a nursery with undersown grasses to alfalfa in a nursery with bare soil differed depending on the trait. Interestingly differences were rather small for vigour related traits, but larger for growth type. No clear recommendations were found as correlations among the compared systems were rather high (low genotype-by-cultivation system interaction) in the complete set of accessions. However, indications were found when working within a set of elite germplasm with reduced variation, that selection decisions might again depend on the cultivation system (different in mixture to non-mixture) and direct selection in the mixture would be more straightforward. Using a nursery system with undersown lawn type grasses might be a good compromise: it is predictive for the system with forage type grasses (FORA) but offers easier phenotyping compared to FORA and easier trial handling (reduced weeding efforts) in comparison to the bare soil system (Trial 23a and Trial 23b alfalfa/ tall fescue and red fescue, by AGROSCOPE and NARDI).

(10) The LER results for the mixed cropping maize/beans in Portugal indicated potential use of the analysed beans genotypes for co-breeding work initiatives in a PPB context (Trial 24, maize /beans, IPC-PT).

Trials on **agroforestry** provided following outcomes:

(1) Clear evidence of the effect of agroforestry on biochemical and physical traits of tomato, with a direct impact of light gradient on the considered traits was found in one year. This study gives a first idea the drivers of plants and microbiome co-evolution through on-farm selection in an agroforestry context. These results could inspire organic breeding to integrate different levels of heredity when willing to select for resilient cropping systems and are first step toward a more holistic agriculture (Trial 25, tomato in agroforestry system, INRAE-FR).

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(2) In LIVESEED a maize trial was sown in syntrophic agriculture system in Portugal, using seeds coming from a CCP developed in and adapted to this system. This germplasm has high value as base to continue the work of selection and adaptation of the CCP 'Sinpre' to agroforestry system (Trial 26, maize in agroforestry system, IPC-PT).

1.5.2. Hypotheses validation

Connected to a set of common objectives, a few common hypotheses were developed at the second annual meeting (Figure 1, page 12). Below the results are presented of those partners who felt confident to either confirm or reject the hypotheses.

Objective No1: Improvement of yield stability by diversity
Hypothesis I - Homogeneous crops are less stable than CCP, OP, dynamic populations, or mixed varieties.
Of the six partners who worked on intraspecific diversity, three partners were able to test this hypothesis. IPC which worked on maize confirmed the hypothesis. Of the two partners who worked on spring wheat, AREI confirmed the hypothesis whereas LBI was not able to validate the hypothesis.

Hypothesis II - Yield of diversified material on average is higher in diverse cropping systems (e.g., organic, stress conditions)
Of the two partners who worked on intraspecific diversity, IPC validated, but LBI rejected the hypothesis. Of the six partners who worked on interspecific diversity (e.g., crop mixtures), three partners confirmed the hypothesis (AREI, IUNG, LBI). FIBL confirmed the hypothesis for crop mixtures with blue lupin but rejected the hypothesis for crop mixtures with white lupin. It is noteworthy to mention here that IUNG was not able to validate the hypothesis for blue lupin.

Objective No2: Special breeding programs/approaches are necessary for more diverse cropping systems
Hypothesis III - Ranking of genotypes is different in homogenous crops compared to heterogeneous crop (significant GxM)
Three partners were able to confirm this hypothesis for the following crops: maize (IPC), leafed pea (AREI) and grasses (AGROS). However, ARIEI rejected the hypothesis for semileafed pea.

Hypothesis IV - Indirect traits can be identified
Two partners were able to work with this hypothesis. IPC confirmed the hypothesis for maize. LBI confirmed the hypothesis for crop mixtures of spring wheat with faba bean and white lupin, whereas they rejected the hypothesis for spring wheat cultivar mixtures.

Objective No3: Participatory breeding approaches are better suited to meet holistic systems-based breeding concept including food network and socio-economic aspects

Hypothesis V - PPB enhance local commitment of stakeholders
This hypothesis was specifically confirmed by the trials on the maize landraces, improved populations and CCP experiments, and the agroforestry trial of IPC in Portugal. This work increased the level of commitment of involved actors, including farmers', citizens', and students and researchers, also when the involvement was at a regular basis and the given stakeholder is not directly benefiting from such systems or what is produced in such systems.

Hypothesis VI.: PPB result in more diverse cropping systems, which need to be adjusted to the local pedo-climatic and socio-cultural conditions

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This hypothesis was confirmed by IPC for maize and INRA and ITAB for the development of DOPs in rivet wheat, spelt wheat, and oat.

For hypothesis I to III partners had different outcomes: Most partners confirmed a hypothesis, but a few rejected the hypothesis. Only a few partners were able to validate hypothesis IV, V and VI. The results show that the main lesson learned here is that it not possible to have hypotheses that can be confirmed by all partners, and that it is important to have context-specific hypotheses.

Next to the common, overarching hypotheses agreed by the research partners, some partners have developed case specific hypotheses, categorized per crop in Table 24:

Table 24. Case specific hypotheses organised per partner

Partner	Hypothesis	Result
INRAE - w. wheat	For autogamous bread wheat, at the same generation, CCPs have a higher level on diversity than Dynamic Populations	Confirmed
INRAE - w. wheat	The selection location has an influence on the agronomic and diversity behaviour of the CCP and Dynamic Population selection strategies	Confirmed
INRAE - w. wheat	Human selection has a positive influence on the agronomic and diversity behaviour of the CCP and Dynamic Population selection strategies and tends to homogenise the populations	Confirmed
INRAE - w. wheat	The DOP selection strategy produces populations that are homogeneous for some specific traits but that keep an overall high diversity	Confirmed
AREI - s. barley	It is possible to obtain stable and good yielding mixtures by mixing components with distinctive disease resistance, yield potential, weed suppressive traits and adaptability	Partly confirmed
AREI - s. barley	Mixtures can provide advantages over growing in pure stand in respect to yield, stability, incidence of leaf diseases, weed suppression ability	Valid for net blotch disease, less for other traits
AREI - s. barley	Reduced diversity in DH lines affect yield stability negatively	Rejected
AREI - s. barley	diversity within a population is higher if maintained under organic vs conventional management	Partly confirmed
AREI - s. barley	CCP populations can provide comparable or better yields to existing varieties under organic and stress conditions	Confirmed
AREI - s. barley	CCPs perform comparably better under environment where it evolved	confirmed
CULTIVARI - pea	The type of leaf and the length of the pea plant has an influence on yield of pea and triticale	Partly confirmed
CULTIVARI - pea	A shorter or taller triticale can compensate missing peas better	Partly confirmed
CULTIVARI - pea	Plant length and leaf type in winter pea should be followed in breeding for best mixing with winter triticale	Partly confirmed
FIBL - w. lupin	Mixed cropping reduces weed pressure	Confirmed
FIBL - w. lupin	Mixed cropping reduces anthracnose pressure through the barrier that the mixing partner poses to secondary infection	Neither confirmed nor rejected
INRAE - tomato	On-farm selection induces changes in plant microbiome to enhance adaptation	Partly confirmed
INRAE - tomato	Microbiome structuration is an indicator of agroecosystem resilience	Partly confirmed

1.5.3. In-depth Results

The data were organized first into four groups (Plant descriptors, Agroecosystem performance, Production Performance and Quality performance) secondly by the division into intra and interspecific criteria, and finally by group of crops (see also Tables 9-21).

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Intraspecific analyses

Plant descriptors

In the group of plant descriptors, it was adapted the BBCH stages for phenology that allowed the comparison of different species[5].

The analyses indicated that **Height** increased for winter wheat on Trial 1 (w. wheat, INRA + GQE-Le Moulon, F) and the same tendency was observed on maize on Trial 7 (maize, IPC, Pt). This can indicate a better adaptation of the tailored populations winter wheat populations for farmers. For maize, this tendency is probably the result of heterosis among populations. Contrastingly lower height in the mixture was registered for spring wheat on Trial 5 (spring wheat, LBI, NL). Neutral responses were present on Trial 2 (w. wheat, INRA + GQE-Le Moulon, F), 4a (w. wheat, ITAB + INRA Rennes, F) (France), 6 (barley, AREI, LV), 8 (white lupin, FIBL, CH).

The **Seedhead** (e.g. inflorescence such as spike ear, or pod) characteristics, related with an awn, colour and shape, were had a positive relationship for the Trials 1 (w. wheat, INRA + GQE-Le Moulon, F) and 7 (maize, IPC, PT) in winter wheat and maize respectively following the same tendency for height, indicating diversity among the mixtures, but neutral for 4a (w. wheat, ITAB + INRA Rennes, F) and 4b (rivet wheat, ITAB + INRA Rennes, F).

90_Harvest, represents the principal growth stage 9 corresponding to Senescence and beginning of dormancy, that can be especially important for cycle length determination that showed a negative trend for Trial 5 (spring wheat, LBI, NL) of which the Mixtures of 2 varieties showed in almost all cases no significant differences with the varieties in pure yield support. A more neutral behaviour was observed for 4b (rivet wheat, ITAB + INRA Rennes, F) and 8 (WHITE LUPIN, FIBL, CH). A neutral response was observed in **10_Emergence** in Trial 4b (w.rivet wheat, ITAB + INRA Rennes, F) and Trial 8 (white lupin, FIBL, CH), **20 Lateral shoots** in Trial 4b (w.rivet wheat, ITAB + INRA Rennes, F), **50 Flowerhead emergence** in Trial 6 (barley, AREI, LV), **60_Fl** and **U (uniformity)** in Trial 7 (maize, IPC) indicating that for the above traits and cultivar mixtures (e.g., winter rivet, white lupins, barley and maize and locations) no differences existed among the material used (Figure 5 – all trials, Figure 6 - wheat and maize trials).

[5] https://en.wikipedia.org/wiki/BBCH-scale

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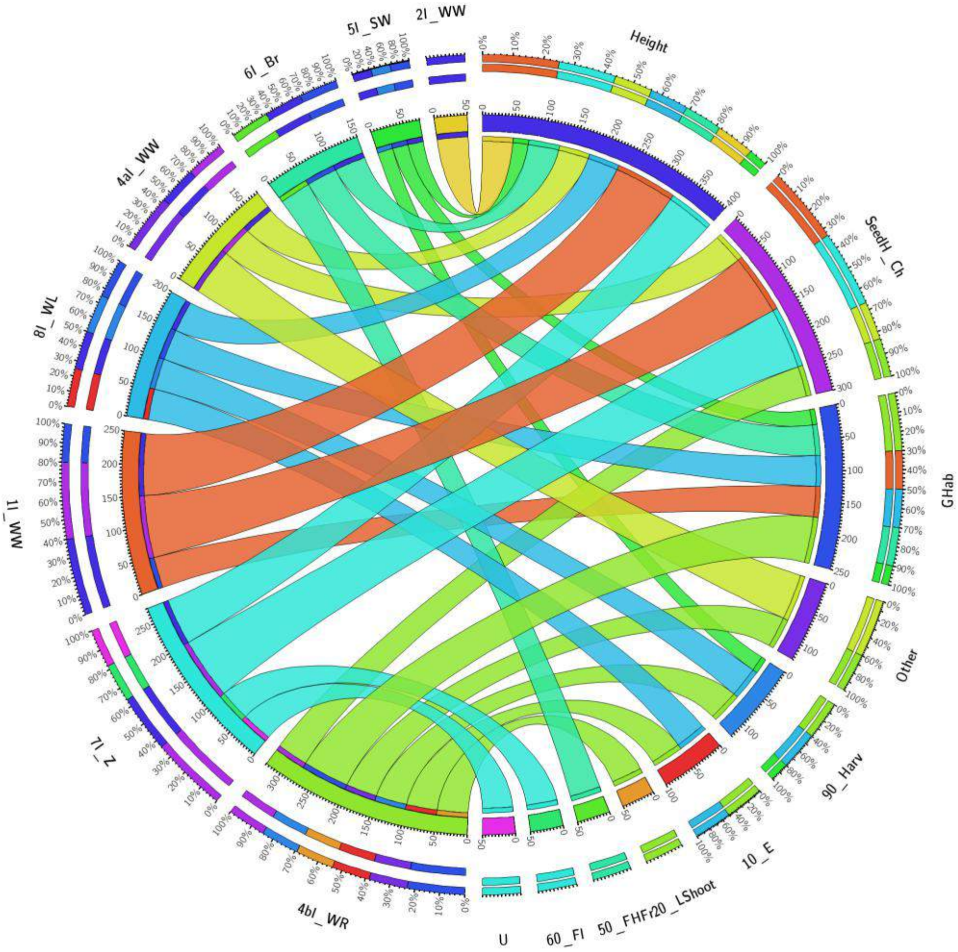

Figure 5. The figure represents intraspecific trials and the plant descriptors associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

The **growth habits**, i.e., *plant habitus*, from erect to ascending and prostrate or decumbent behaviour, can indicate an important way how plants interact among them in the population and adapted to a certain location. In trial 4b (rivet wheat, ITAB + INRA Rennes, F), the growth habit was moderately positive indicating that diversity on this trait exists across the 28 different populations. In contrast the spring wheat Trial 5 (spring wheat, LBI, NL) showed moderately negative results, that indicates less variation for this trait. Neutral results were observed for Trials 1 (w. wheat, INRA + GQE-Le Moulon, F), 6 (barley, AREI, LV) and 8 (WHITE LUPIN, FIBL, CH).

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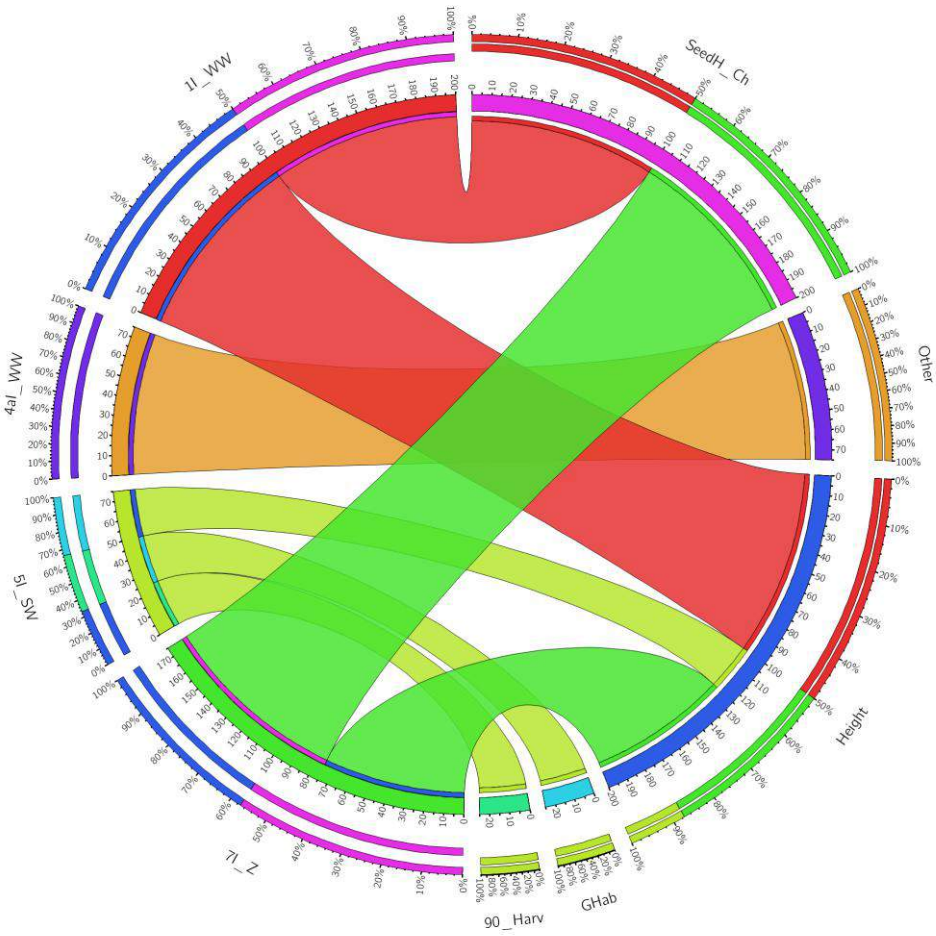

Figure 6. The figure represents intraspecific trials and the plant descriptors associated for wheat and maize only, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

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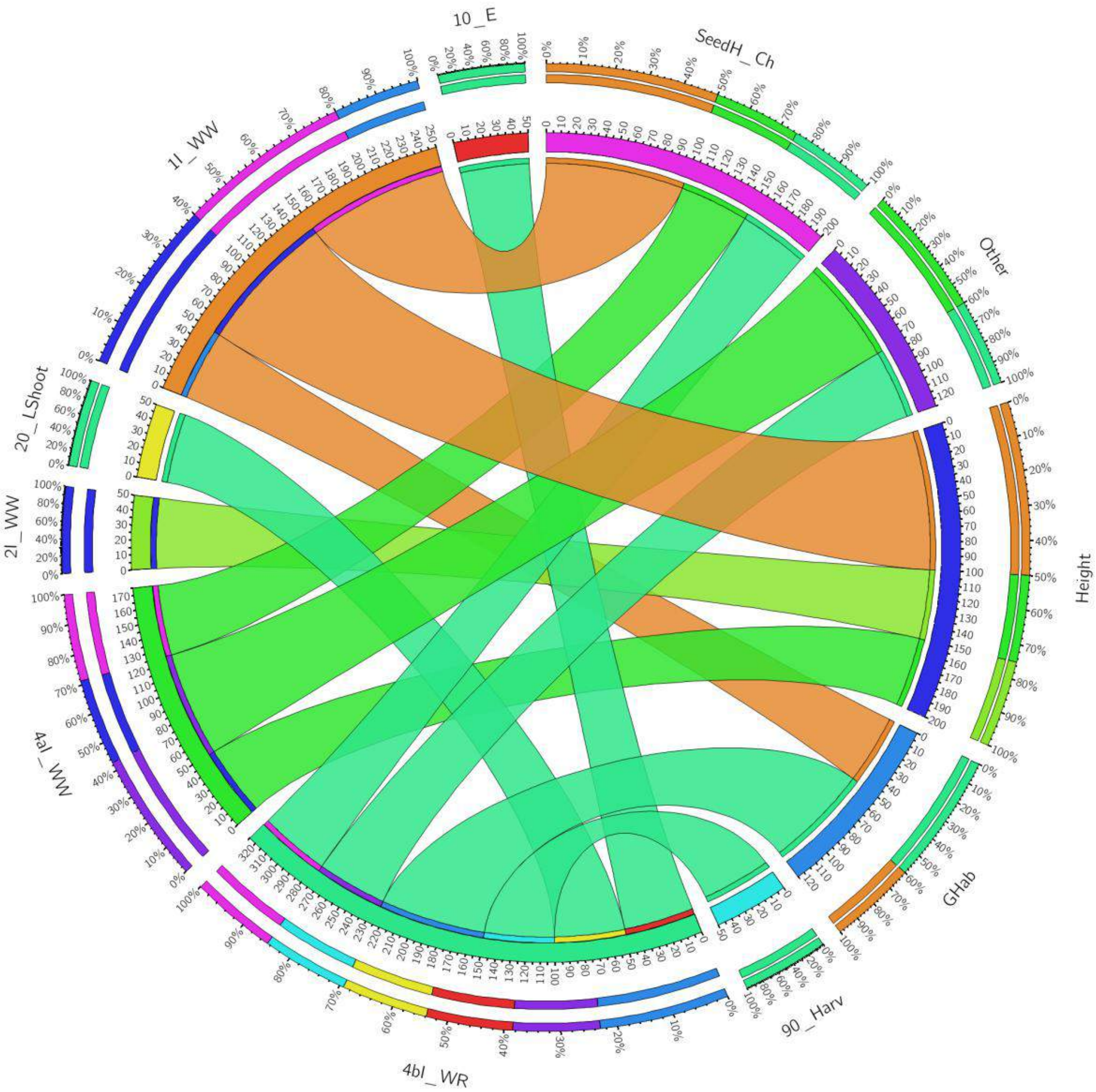

Figure 7. Relevant traits in winter wheat (Height and Seed Head Characteristics). The figure represents intraspecific trials and the plant descriptors associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials. Positive relationship traits for Height and Seed Head Characteristics for Trial 1 (w. wheat, INRA + GQE-Le Moulon, F) (Figure 7) and Growth habit for river wheat on trials 4b (w. rivet wheat, ITAB + INRA Rennes, F).

Agroecosystem performance

The **standing ability**, generally measured by the trait lodging in several phases, increases moderately for Trial 1 (w. wheat, INRA + GQE-Le Moulon, F) and increase for Trial 7 (maize, IPC, PT) indicating respectively that adapted winter wheat mixtures and maize CCPs had a less lodging and better performance. **Cover** had a positive trend in the case of Trial 4b (w. rivet wheat, ITAB + INRA Rennes, F) that matches with same trend observed for growth habit. The **Biotic stress response**, that includes the diseases caused by biotic factor (e.g., diseases), was moderately favourable for Trial 6 (barley, AREI, LV) in barley where mixtures can provide advantages over growing in pure stand in respect to

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incidence of leaf diseases. However, a contrast result was found for white lupin where an increase susceptibility to anthracnose was recorded (Figure 8 - all trials and Figure 9-contrasting cases). Neutral values which also indicates that mixtures had a similar result to parental lines, were observed for **weed response** and **cover** both for winter wheat in France Trial 2 (w. wheat, INRA + GQE-Le Moulon, F) and barley on Trial 6 (barley, AREI, LV).

No clear relationship were observed for the: **vigour**, **Cover companion crop**, on Trial 6 (barley, AREI, LV); for stand ability for Trial 4b (w.rivet wheat, ITAB + INRA Rennes, F) and Trial 6 (barley, AREI, LV); for **establishment** (generally associated with Days from sowing to full emergence), on Trials 2 (w. wheat, INRA + GQE-Le Moulon, F) and Trial 4b (w.rivet wheat, ITAB + INRA Rennes, F) and Trial 8 (white lupin, FIBL, CH); for **Biomass production** both Trial 1 (w. wheat, INRA + GQE-LE MOULON, F) and Trial 2 (w. wheat, INRA + GQE-Le Moulon, F); **weed response**, that can be obtained by scoring or measuring the biomass production of the crop and the weeds, on Trials 2 (w. wheat, INRA + GQE-Le Moulon, F) and Trial 6 (barley, AREI, LV).

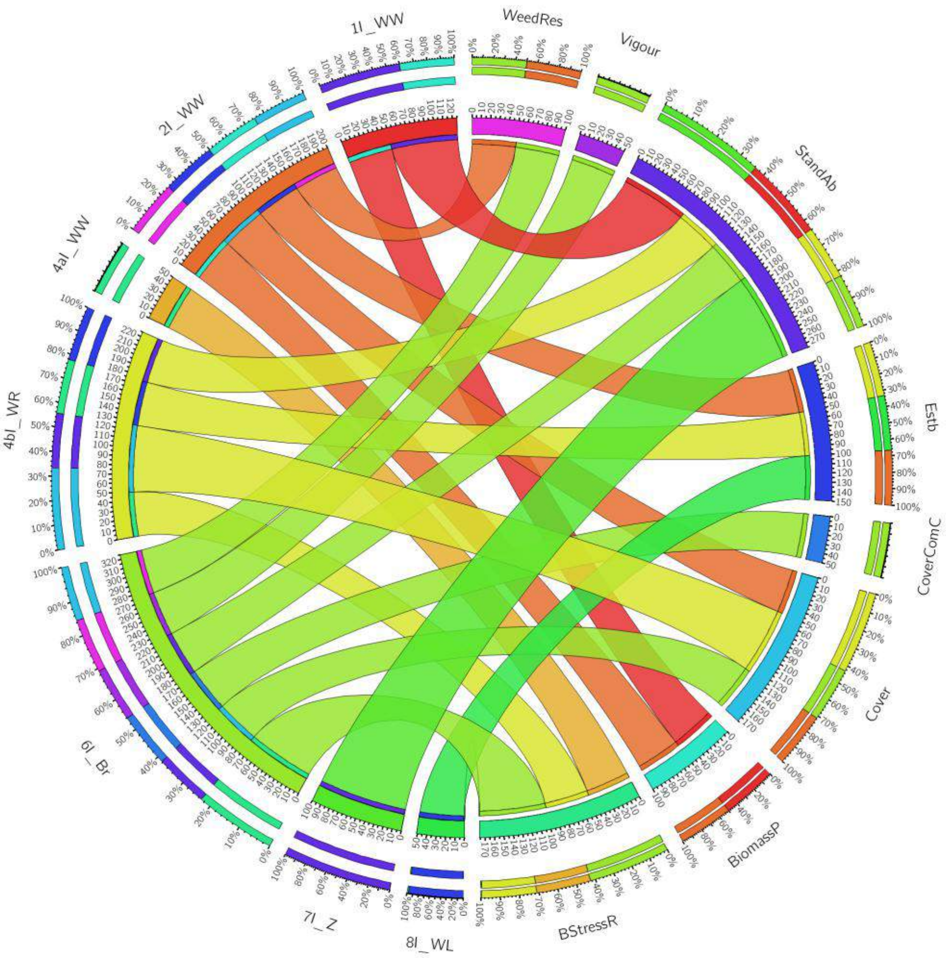

Figure 8. Traits from intraspecific trials and the agroecosystem performances associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

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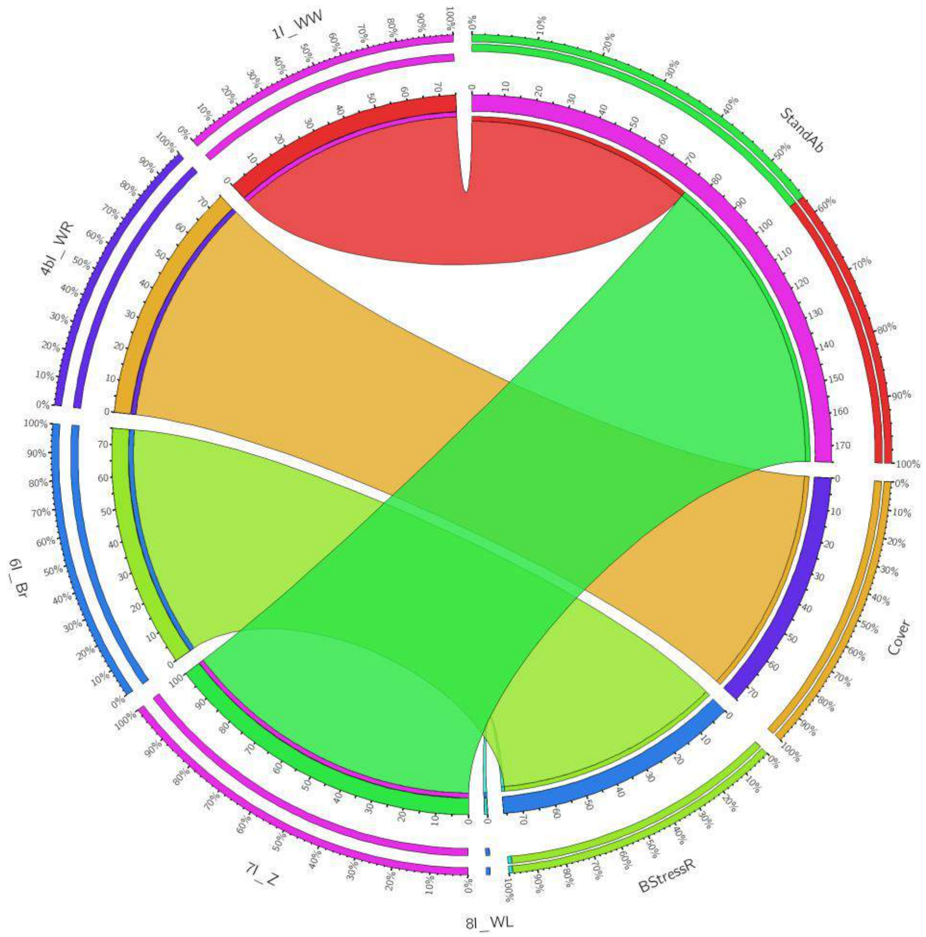

Figure 9. Selected traits (Biotic Stress, Cover and Stand ability) from intraspecific trials and the agroecosystem performances associated (for wheat, rivet wheat, barley, and maize), indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

No clear relationship were observed for the: **vigour**, **Cover companion crop**, on Trial 6 (barley, AREI, LV); for stand ability for Trial 4b (w.rivet wheat, ITAB + INRA Rennes, F) and Trial 6 (barley, AREI, LV); for **establishment** (generally associated with Days from sowing to full emergence), on Trials 2 (w. wheat, INRA + GQE-Le Moulon, F) and Trial 4b (w.rivet wheat, ITAB + INRA Rennes, F) and Trial 8 (white lupin, FIBL, CH); for **Biomass production** both Trial 1 (w. wheat, INRA + GQE-LE MOULON, F) and Trial 2 (w. wheat, INRA + GQE-Le Moulon, F); **weed response**, that can be obtained by scoring or measuring the biomass production of the crop and the weeds, on Trials 2 (w. wheat, INRA + GQE-Le Moulon, F) and Trial 6 (barley, AREI, LV). For **wheat group the** relationships are emphasized throughout Figure 10.

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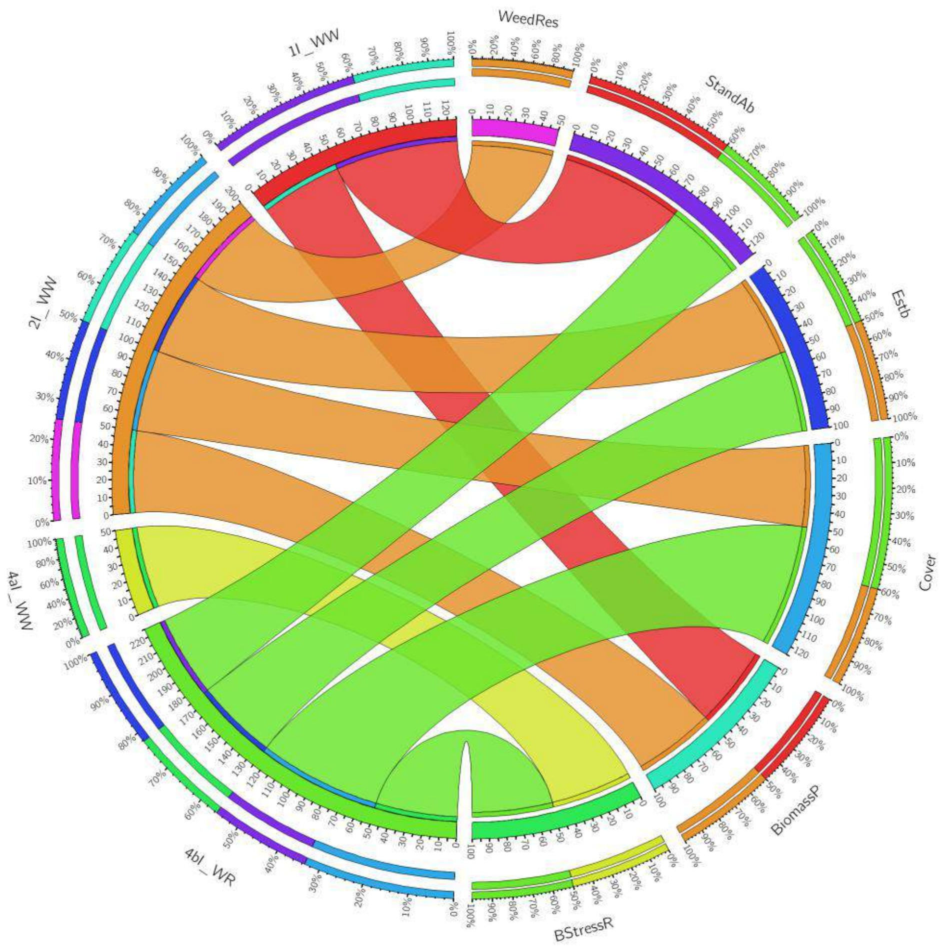

Figure 10. Traits from intraspecific trials and the agroecosystem performances associated for wheat only, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

Productive performance

Yield stability, (indicate across the years the rank production of a certain population, CCP or other accession), was observed from positive trend in winter wheat Trials 1 (w. wheat, INRA + GQE-Le Moulon, F) and 2 (w. wheat, INRA + GQE-Le Moulon, F) to positive relationship in barley Trial 6 (AREI, Latvia). The **Seed head size (**is the spike length average, having the basis of the numbers of kernels per row) had a positive relationship to Trial 1 (w. wheat, INRA + GQE-Le Moulon, F) and had a positive trend for maize in Portugal (Trial 7), indicating a bigger length of spike and ears. For six of the experiments the **Yield** of the mixtures had a positive relationship for Trial 5 (spring wheat by LBI, NL) and Trial 6 (barley AREI, Latvia). A positive trend was observed on Trial 1, (w. wheat, INRA + GQE-Le Moulon, F) Trial 2 (w. wheat, INRA + GQE-Le Moulon, F) and Trial 7 (maize for IPC, Portugal) indicating that more diverse germplasm can contribute to yield increase for organic. A contrasting result was also observed on trial 8 for winter lupin in which the yield decreased, that was related with the lack of resistance to diseases from the germplasm used in breeding. The cultivar **mixing ability** (indicated as **LER**) had a positive trend for both Trials 1 (w. wheat, INRA + GQE-Le Moulon, F) and 2 (w. wheat, INRA + GQE-Le Moulon, F) (Figure 11, all trials). **Grain weight** was relevant to had a positive trend for maize (Trial 7, IPC, Portugal) winter wheat and rivet wheat (respectively for Trial 4a (w. wheat, ITAB +

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INRA Rennes, F) and 4b (w. rivet wheat, ITAB + INRA Rennes, F) on DOPs, ITAB + INRA Rennes) in contrast for winter wheat (Trial 1, INRAE, GQE) it was not relevant.

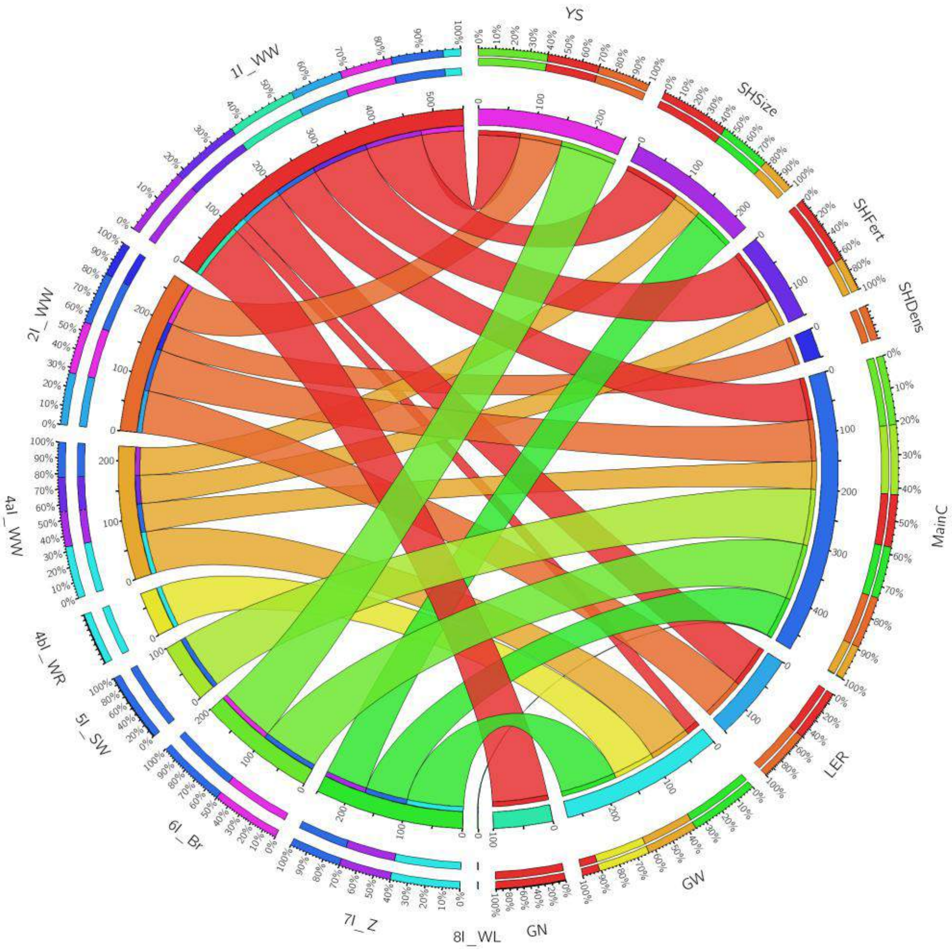

Figure 11. Traits from intraspecific trials and the productive performances associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

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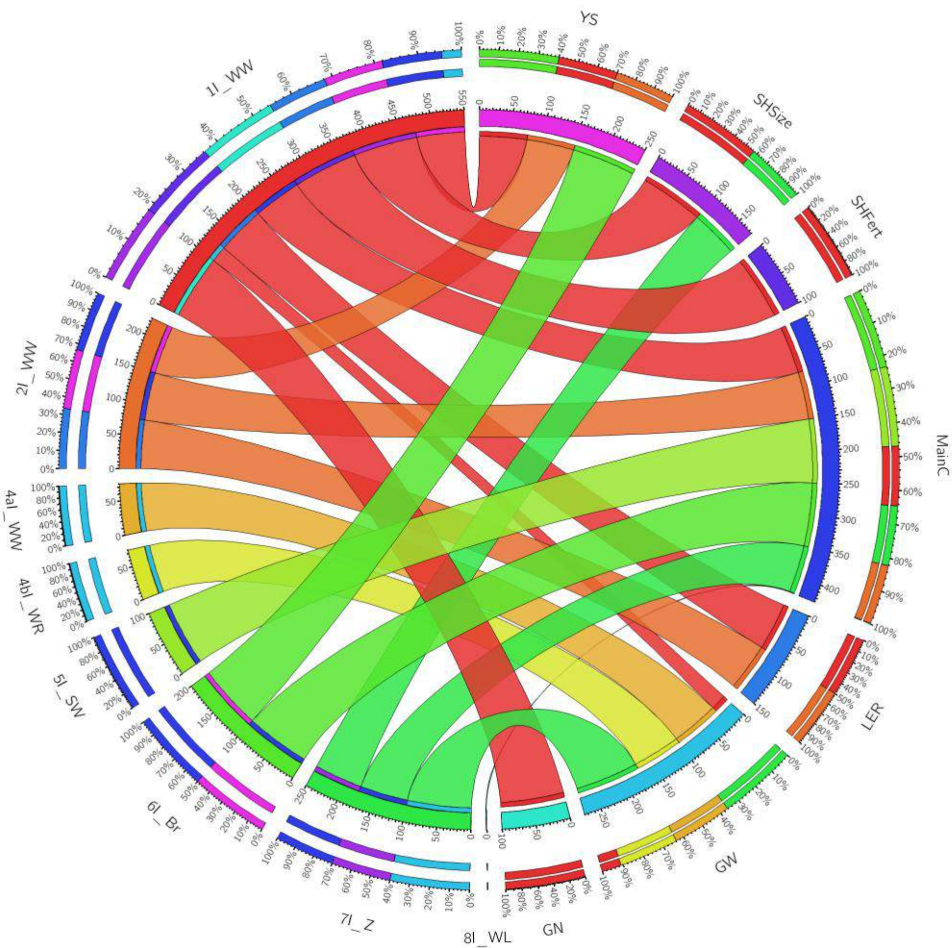

Figure 12. Selected traits from intraspecific trials on wheat, barley and maize, and the productive performances associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

The productive performance can be characterized by several traits for each of the trials. A positive relationship was obtained for **Seedhead size**, **Seedhead fertility**, **Grain number** on Trial 1 (w. wheat, INRA + GQE-Le Moulon, F) (Figure 12) indicating that germplasm used in Trial 1 had the capacity to increase of spike length average, number, and weight of grains per spike and grain number respectively. The **Seedhead size** had a positive trend for Trial 7 (maize, IPC-PT) indicating that diverse cultivars can contribute to increase this trait.
The grain weight (average grain weight, considering 100 or 1000 kernel weight) indicate that was a negative trend for Trial 1 (w. wheat, INRA + GQE-Le Moulon, F), a positive trend to Trials 4 (4a (w. wheat, ITAB + INRA Rennes, F) and 4b on DOPs, ITAB + INRA Rennes) and a positive relationship with Trial 7 (maize, IPC-PT).

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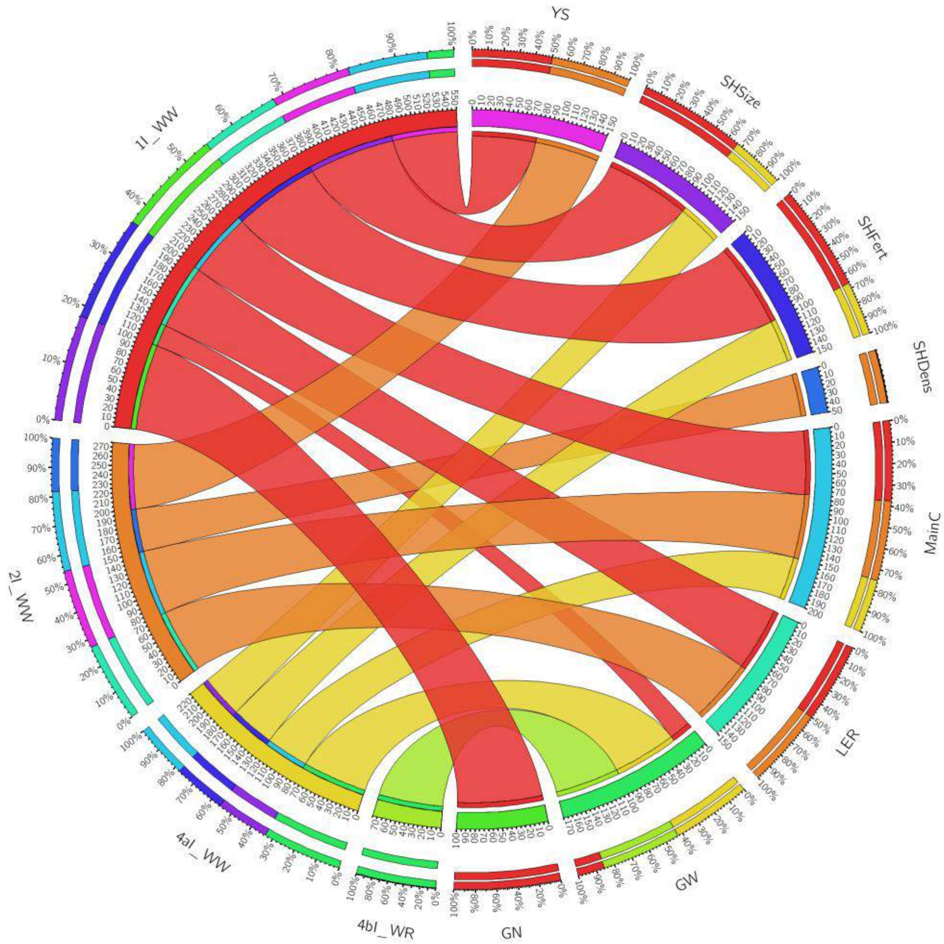

Figure 13. Traits from intraspecific trials (wheat only) and the productive performances associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

For wheat group the relationships are emphasized throughout Figure 13. The specific traits found for each of the trials had a positive relationship for Trial 1 (w. wheat, INRA + GQE-Le Moulon, F) the **Grain number** that probably explains the negative trend for Grain Weight; whereas there in no clear relationship between Trial 2 (w. wheat, INRA + GQE-Le Moulon, F) and **Seedhead density**.

Quality performance

In the quality performance it was observed that winter wheat mixtures of Trial 2 (w. wheat, INRA + GQE-Le Moulon, F) were related with **Baking quality**. It was also observed that **Moisture** had a positive trend for the maize composite (Trial 7, IPC), that is generally related with longer cycles and increases costs for drying. The **Grain characteristics** (colour, thousand kernel weight) had a positive trend for winter wheat Trial 4a (w. wheat, ITAB + INRA Rennes, F). In contrast, **Thousand grain weight** were moderately unfavourable for the Trial 1. In addition, many other neutral traits were found indicating that mixtures are similar to parental lines Figure 14 -all trials, Figure 15 -winter wheat and maize.

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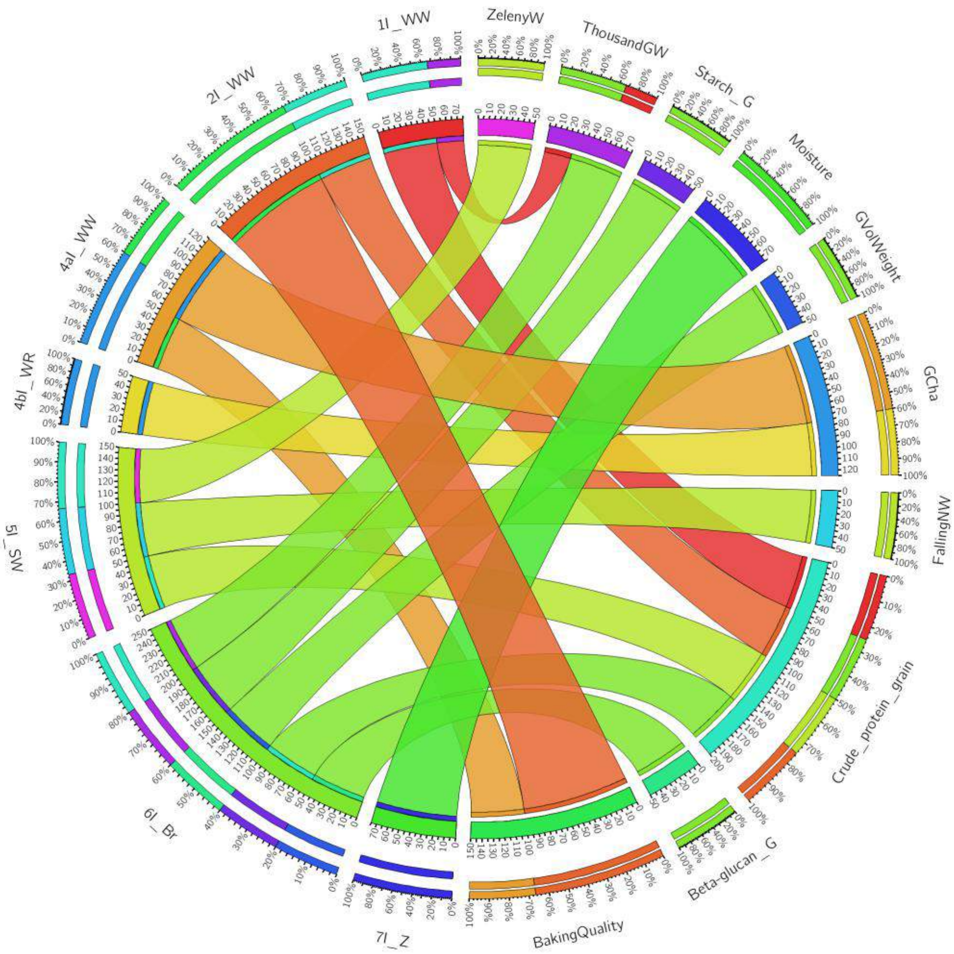

Figure 14. Traits from intraspecific trials and the quality performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

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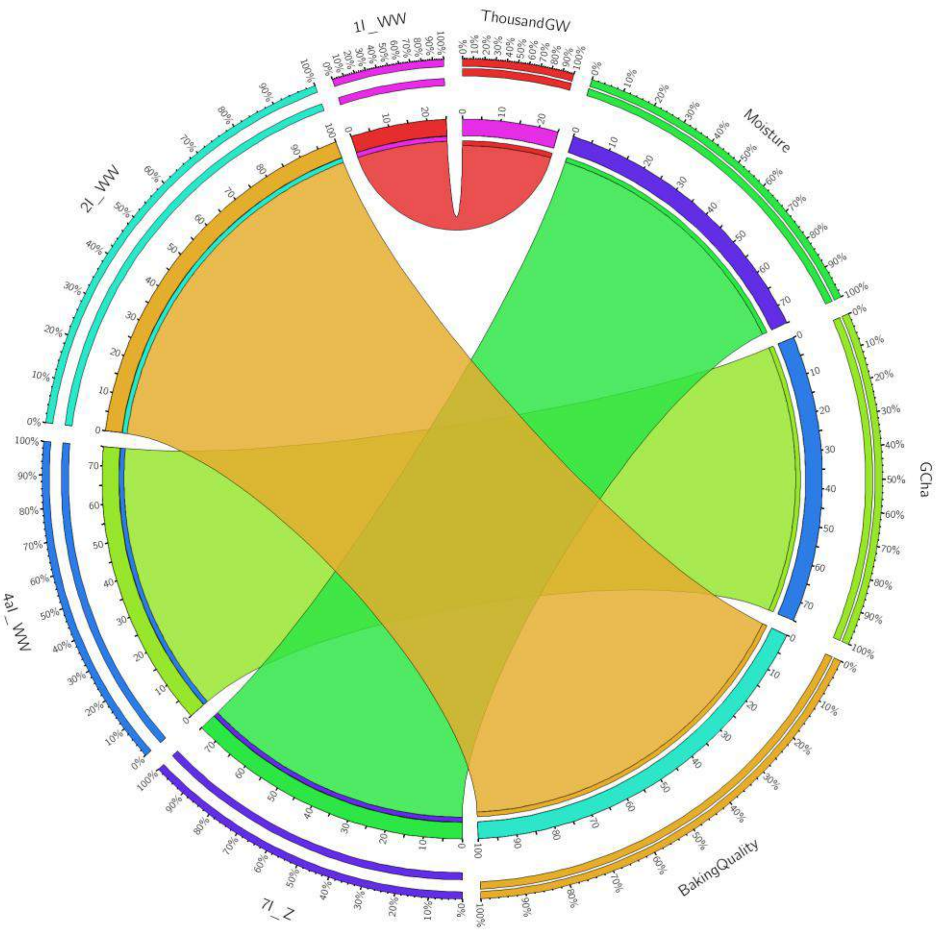

Figure 15. Selected traits from intraspecific trials on winter wheat and maize and the quality performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

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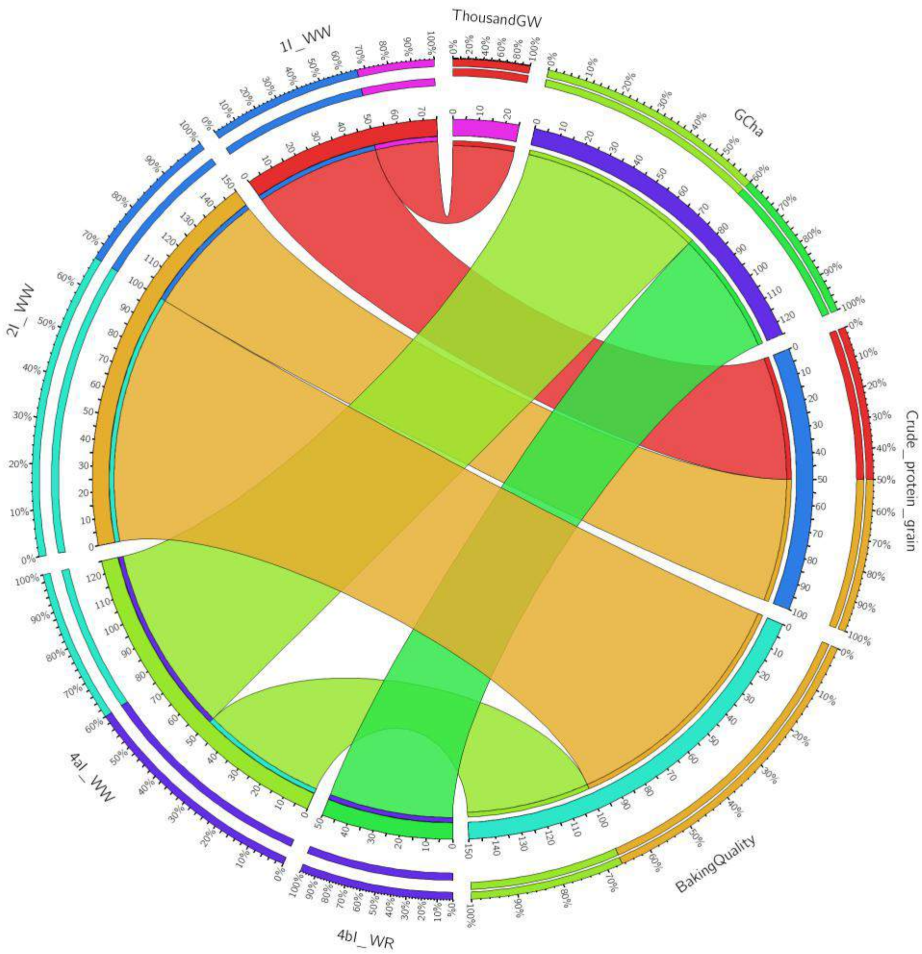

Figure 16. Traits from intraspecific trials (wheat only) and the quality performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

The winter wheat mixtures indicate to have a positive relationship for **Baking quality** for Trial 2 (w. wheat, INRA + GQE-Le Moulon, F) and also for **Grain characteristics** for Trial 4a (w. wheat, ITAB + INRA Rennes, F) (Figure 16).

Interspecific analyses (annuals)

Plant descriptors

The traits **Seedhead characteristics** (e.g. awn, colour and shape), **Height**, **Growth habit** (plant habitus corresponds to erect to ascending and prostrate or decumbent behaviour) for pea, faba bean and white lupin had a positive relationship for Trials 12 (different cereals x pea, LBI, NL), 13 (spring wheat x faba bean, LBI, NL) and 19 (spring wheat x white lupin, LBI, NL), indicating similar patterns for different legumes in similar ecogeographic areas for example it was understood that height and number of pods of the lupin variety are important factors to perform well in intercropping conditions with wheat (Trial 19 (spring wheat x white lupin, LBI, NL)).

The trait **20-lateral shoots** (the principal growth stage 2 corresponds to the formation of side shoots/tillering) had a positive relationship both for blue as for white lupin in Swiss growing conditions

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for Trial 17 (intercropping with blue lupin, FIBL-CH) and Trial 18 (intercropping with white lupin, FIBL-CH), which is probably related how the plants grow and its successful adaptation to the intercropping, as shown in Figure 17.

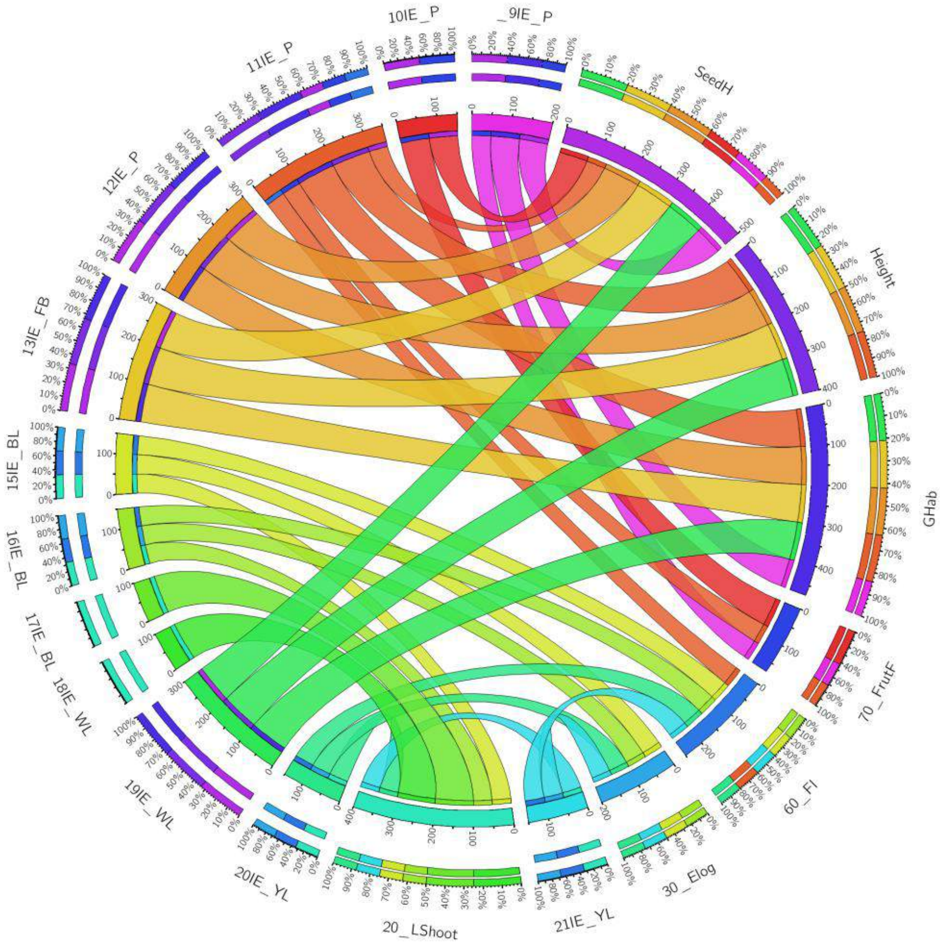

Figure 17. Traits from interspecific trials (pea, faba bean and white lupin) and the plant descriptors associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

In the case of Trial 11 (winter triticale x winter pea, CULTIVARI, D), a positive relationship was also found for **Height**, **Growth habit** similarly to the Trial 12 (different cereals x pea, LBI, NL) with exception of **Seed head characteristics**. A positive trend was found for **Seed head characteristics** in both Trials 9 and 10 (AREI, Latvia and GZPK+FIBL, CH). However, Trial 9 (spring wheat x spring pea, AREI, LV) had a positive trend with **Growth habit** and Trial 10 (spring triticale/barely - pea, FiBL-CH-GZPK) with **70-fruit formation** (the principal growth stage 7 that corresponds to development of fruit, see Figure 18) that was particularly relevant for searching the best mixtures, in which the synchronized fruit formation of the mixtures was particularly relevant for barley and pea mixture.

The **Height** and **Growth habit** had a positive relationship with Trial 11 (winter triticale x winter pea, CULTIVARI, DE) and Trial 12 (different cereals x pea, LBI, NL), for CULTIVARI's short peas were suppressed most, but a medium type in length could reach nearly up to the very tall pea varieties, which suppressed triticale most. For the medium length type of pea, the triticale seemed to be a more or less strong competitor. Breeder has been selecting to search better triticale genotypes for mixtures, indicating that some morphological characters should be considered such as broadness of leaves, date

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of ear emergence and rapidness of juvenile growth, plant length and ears per square meter to develop a 'triticale mixture type'. For LBI it is stressed that mixed cropping with peas is certainly possible in the Netherlands with the right pea varieties and cereals and some mixed treatments showed more suppression of weeds compared to the pure stand pea treatments, especially the mixed treatment with barley and oats. To better understand the variation in characteristics we have organised the data for peas and lupins in Figure 19 and 20 respectively.

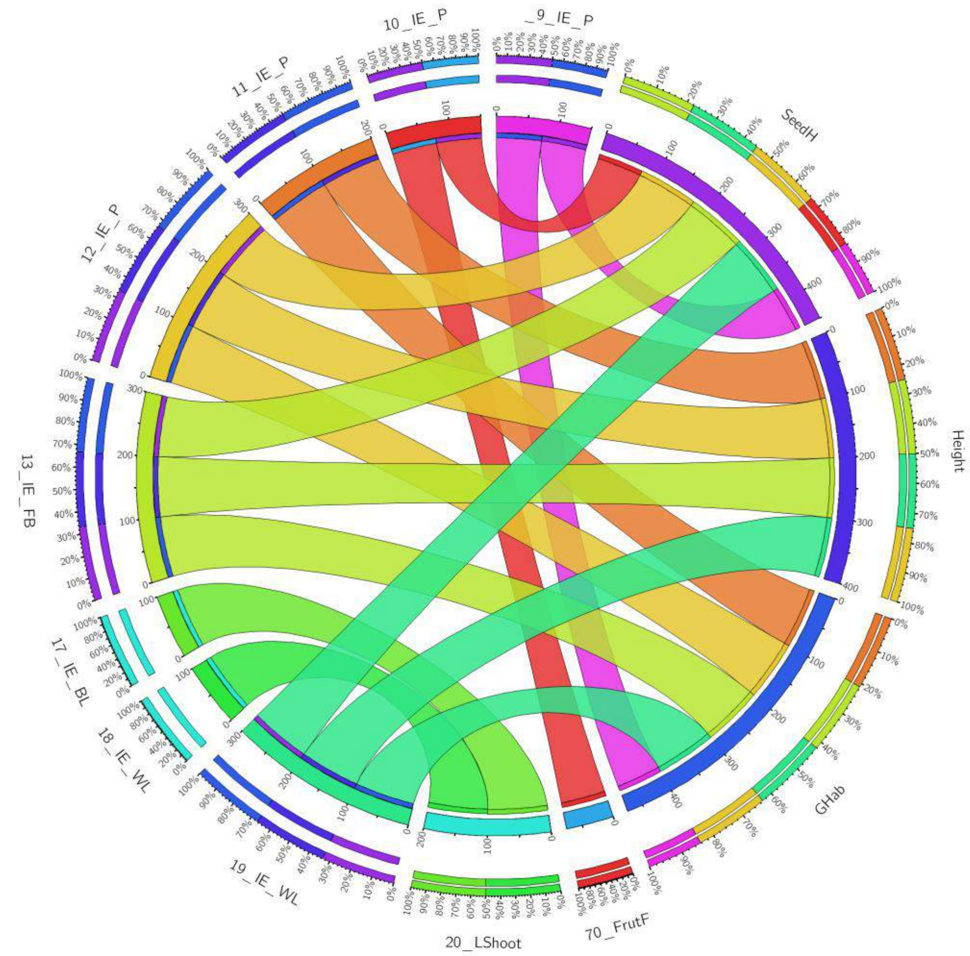

Figure 18. Traits from interspecific trials (pea, faba beans and lupins) and selected plant descriptors associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

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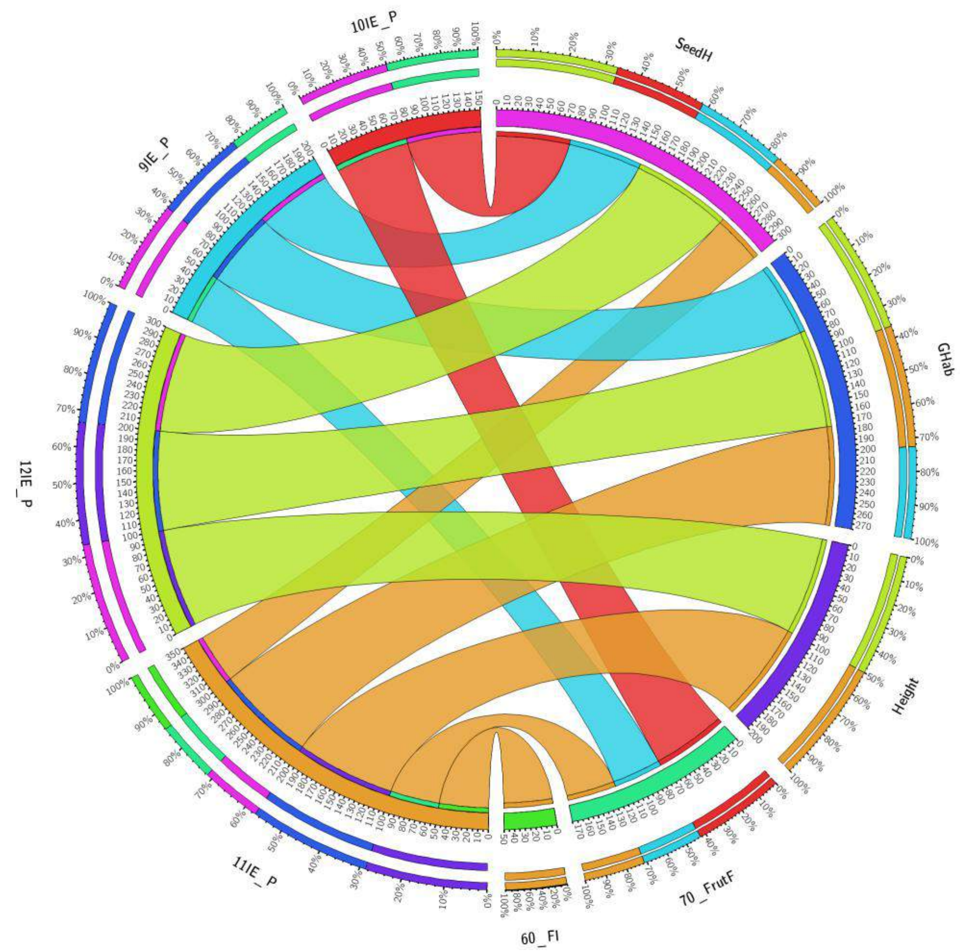

Figure 19. Traits from interspecific trials (only peas) and the plant descriptors associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials. ***Seedhead*** *characteristics indicate positive relationship for Trial 12 (different cereals x pea, LBI, NL) and positive trend to Trials 9 (AREI) and 10 (GZPK+FiBL). The* ***70-fruit formation*** *positive relationship indicates for Trial 10 (GZPK+FiBL) the extremely early flowering "Pisum sativum 75121"*

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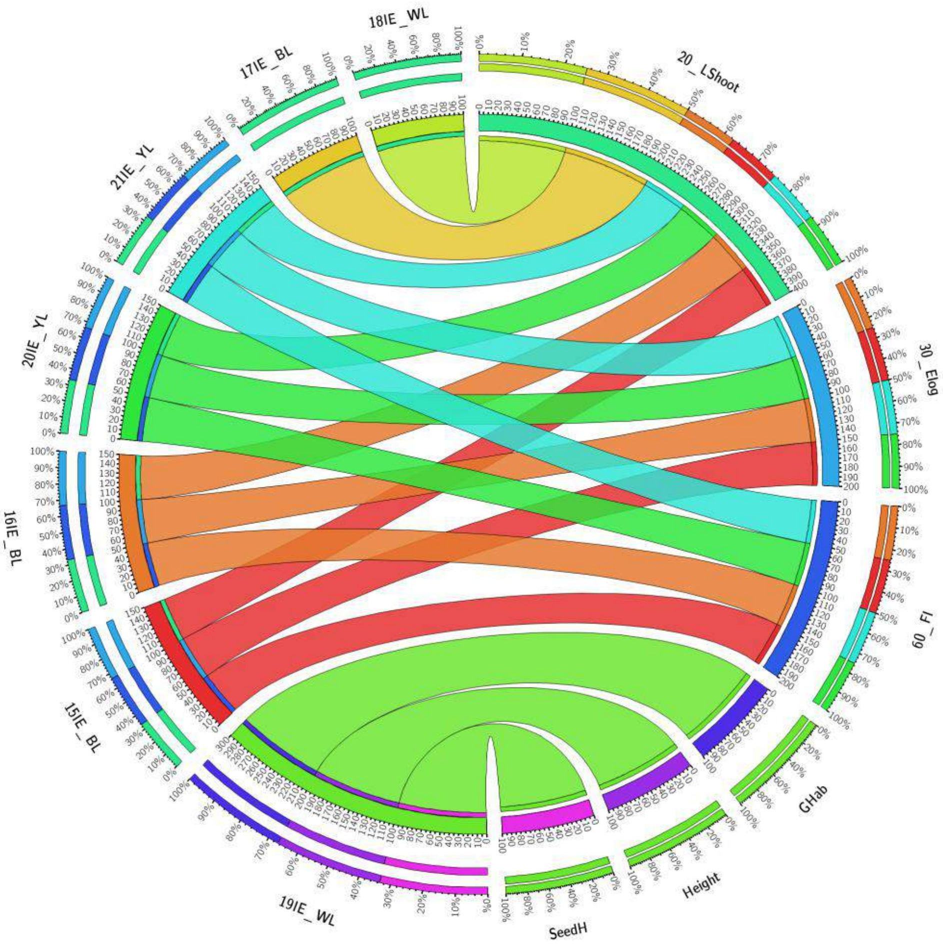

Figure 20. Traits from interspecific trials (lupin only) and the plant descriptors associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials. For the traits related with Trials 15 (spring triticale x blue lupin, IUNG, PL), 16 (spring oat x blue lupin, IUNG, PL) on blue lupin and Trials 20, 21 on yellow lupin by IUNG-PL, there was none or no clear relationship with 20-lateral shoots (the principal growth stage 2 corresponds to the formation of side shoots/tillering), 30-elongation (the principal growth stage 3 corresponds to the Stem elongation or rosette growth, shoot development) and 60-flowering. However, it was possible to establish a positive relationship with ***20-lateral shoots*** *in blue and white lupins for Trials 17 (intercropping with blue lupin, FIBL-CH) and Trial 18 (intercropping with white lupin, FIBL-CH) respectively, that stresses its vigour. The 70- fruit formation had a positive relationship for Trial 10 (GZPK) regarding the traits described as late flowering and maturity.*

Agroecosystem performance

A positive relationship was observed among **Biotic Stress Response** and the Trials 12 (different cereals x pea), 13 (spring wheat x faba bean) and 19 (spring wheat x white lupin) respectively for pea, faba bean and white lupin in The Netherlands by LBI (Figure 21). The hypothesis that genetic diversity in the field enhances resilience against diseases was not confirmed due to the low disease pressure in the wheat and legume crops. However, in the Trial 19 spring (wheat-lupin mixed crop), it was observed some brown rust in the lower parts of wheat plants in some plots, which was not present in the pure stand crop. This could be due to the higher relative humidity inside the mixed crop. This finding is contrary to the hypothesis, whilst only an indication.

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Cover had a positive relationship in Trial 9 (spring wheat x spring pea, AREI, LV) and Trial 17 (intercropping with blue lupin, FIBL-CH) and positive trend with Trial 14 (s. wheat/s. triticale x faba bean, AREI, LV). For example, on Trial 9 (spring wheat x spring pea, AREI, LV) without reduction the sowing rate of peas, but the adding 20% of wheat to the sowing rate of the semi leafless genotypes, the **Grain yield** did not decrease. Besides, the **Soil coverage** significantly increased which was highly beneficial for semi leafless genotypes, it could help to withstand with high weed pressure. In the case of Trial 17 blue lupins showed good mixing ability with oat or triticale for Swiss conditions.

A negative trend was found for **Cover companion crop** (Cover companion crop aimed at identifying suitable companion crops for mixed cropping and/or undersowing, the scale used was % of covered soil) in Trial 18 (intercropping with white lupin, FIBL-CH) where the mixed cropping rendered no improvement. **Weed suppression** decreases toward the end of the season when leaves have completely fallen down, a partner during this period that do not affect is still lacking. The most important target however for white lupin is the anthracnose reduction.

Establishment companion crop (can be important to at identify suitable companion crops for mixed cropping and/or undersowing (e.g., Trial 17 and 18)) had contrasting trends for the Trial 17 and 18 (intercropping with white lupin, FIBL-CH). Mixed cropping of blue lupins (*Lupinus angustifolius*) with oat or triticale is now recommended to farmers, while for white lupins (*Lupinus albus*) mixed cropping has no advantage under spring sowing conditions in Switzerland. Difficult to find suitable companion crops and undersowing plants for white lupin due to long vegetation period (mid-March – August/September). Strong vigour of white lupins in spring - especially under drought conditions - and early canopy coverage due to larger leaf areal compared to small leaved blue lupins results in suppression of less drought tolerant companion crops.

Establishment (Establishment, generally associated with Days from sowing to full emergence) had a positive relationship to a negative trend for respectively Trials 17 (intercropping with blue lupin, FIBL-CH) and 18 (intercropping with white lupin, FIBL-CH), mixed cropping blue lupin-cereal gave good results in the mixture contrarily to white lupin-cereal, because white lupin is more competitive, overgrows companion and undersowing plants.

Standing ability, can also be measured by the trait lodging in several phases (e.g., on pea from the beginning to the end of flowering and ripening), had a positive relationship for Trials 11 (winter triticale x winter pea, CULTIVARI, D) where is indicated that normal leaf type is also more sensitive for lodging, which might limit the use related to the fertility of the soil, the location, and the weather conditions. To breed for better mixing ability, a medium to tall normal-leafed winter pea will be the first choice and a high yielding triticale with good compensation should be the one to combine with in yield trials for pea selection.

A positive trend for trial 9 (spring wheat x spring pea AREI, LV) and Trial 10 (Pea GZPK+FiBL-CH). For Trial 9, the leafed varieties they have a poor lodging resistance, for this reason farmers associate a support crop, however it is not recommended short genotypes. In case of semi leafless pea genotypes which ranked high for GY in the pure stand, also ranked high when grown in a mixture with wheat. This finding allows the selection of semi-leafless genotypes to perform in a mixture within an organic breeding program and recommends the most suitable genotypes for a mix with spring wheat to the farmers for growing under organic conditions.
In the case of Trial 10, the effort done for Trial 10 on Pea, GZPK+FiBL allowed to identify interesting traits to introduce in the GZPK breeding programme to enhance resilience of this crop and identify interesting varieties for special uses (“niche variety”).

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Weed response had a positive relationship for Trial 11 (winter triticale x winter pea, CULTIVARI, D), the experiment could show the positive effect of a normal leafed pea to better supress weeds from the beginning of the growing season on and there seems to be a tendency, that normal leaf type of pea can reach better yield. But normal leaf type is also more sensitive for lodging, which might limit the use related to the fertility of the soil, the location, and the weather conditions. Therefore, more variety trials with mixtures at different locations would be preferable.

The Pea Trials indicated that **Stand ability** had positive relationship for Trial 11 (winter triticale x winter pea, CULTIVARI, D), that is corroborated with a positive relationship with **Weed response.** A positive trend for **Stand ability** with Trial 9 (spring wheat x spring pea, AREI, Latvia) and Trial 10 (S. triticale/barley-pea, GZPK+FiBL-CH) stresses the importance of semileafed and leafed plants in mixture and the need for more research regarding the mixtures. **Cover** had a positive relationship with Trial 9 (spring wheat x spring pea, AREI, Latvia) guiding the need both the type of peas as the varieties of the supported crop. **Biotic Stress Response** had a positive relationship with Trial 12 (different cereals x pea, LBI, NL) by preventing lodging and development of diseases.

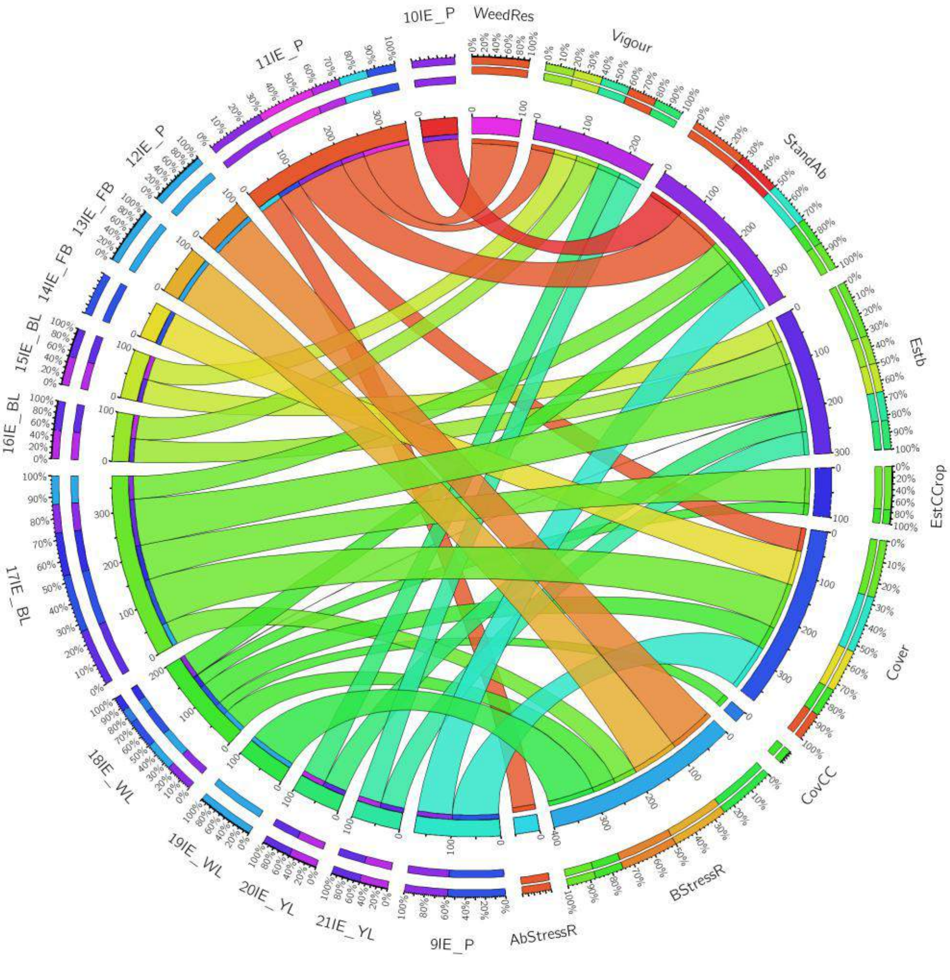

Figure 21. Traits from interspecific trials (legumes) and the agroecosystem performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

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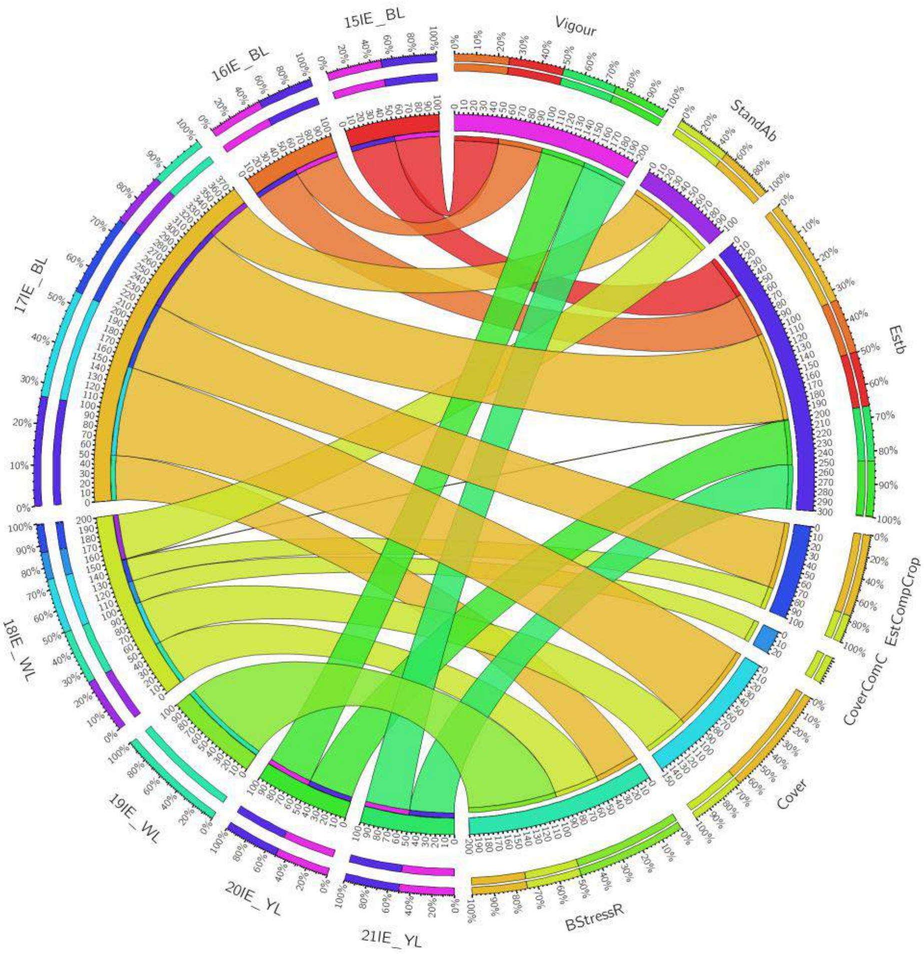

Figure 22. Traits from interspecific trials (lupins) and the agroecosystem performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

In the case of Lupins (Figure 22), the **establishment** had a negative relationship for Trials 18 (intercropping with white lupin, FIBL-CH) indicating that white lupin was not adapted to the intercropping due to its long cycle and vigorous behaviour. In contrast the Trial 17 (intercropping with blue lupin, FIBL-CH) was particularly successful, being adapted in Switzerland. The positive relationship is emphasised on **cover** for Trial 17. **Biotic Stress Response** had a positive relationship with Trial 19 (spring wheat x white lupin, LBI, NL) (spring wheat x white lupin, LBI, NL) by preventing lodging and development of diseases. **Establishment companion crop** had contrasting trends from positive in Trial 17 (intercropping with blue lupin, FIBL-CH) to negative trend in Trial 18 (intercropping with white lupin, FIBL-CH) which is emphasized with the same result for the trait **Cover companion crop**.

Production performance

A positive relationship for the traits **Overall intercrop Yield** (provides de global yield), **Main crop/cereal** (it corresponds to the grain yield of the main crop), **Companion crop/grain legume** (it corresponds to the grain yield of the crop that is in the mixture, generally grain legume) were simultaneous observed with a positive relationship for Trial 10 (Pea, GZPK+FiBL-CH) in which several favourable mixtures were tested, with Trial 12 (different cereals x pea, LBI, NL) indicates also the potential of the right mixtures between pea and cereals, Trial 13 (spring wheat x faba bean, LBI ,NL), the mixed Faba bean/wheat treatments showed (in most cases) a significant higher total yield compared to the pure stand treatments. Intercropping shows to be a good solution to achieve higher

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yields with less weed management and to increase the biodiversity on the field. It is interesting observe that in the intercropping wheat tend to have higher height and faba bean lower height comparing with crops per se; in Trial 15 (spring triticale x blue lupin , IUNG, PL) so as in Trial 16 (spring oat x blue lupin, IUNG, PL) indicate that mixtures of lupins with oat performed better (higher yields and lower variability in years) than mixtures with spring triticale it is referred that self-determinate cultivars have a good potential for their wider utilization in organic farming, mixtures of lupines with cereals can be recommended for organic farmers, especially on poor, sandy soils; Trial 19 (spring wheat x white lupin, LBI, NL) the mixed lupin/wheat treatments showed a significant higher total yield compared to the pure stand lupin treatments. The high weed pressure probably reduced the yield of the pure stand lupin treatments. So intercropping shows to be a good solution for weed suppression when cultivating lupins. The pure stand wheat treatments showed no difference in total yield compared to the mixed lupin/wheat treatments, indicating the compensation, or 'filling' capacity of the wheat crop, Trial 20 (spring triticale x yellow lupin, IUNG, PL) and 21 (spring oat x yellow lupin, IUNG, PL) indicate that mixtures of lupins with oat performed better (higher yields and lower variability in years) than mixtures with spring triticale, it was also observed that Self-determinate cultivars have a good potential for their wider utilization in organic farming. In addition, a positive relationship was observed for **Main crop/cereal** and **Yield stability** in the Trial 11 (winter triticale x winter pea, CULTIVARI, D) that indicates breeding concerns for not only the adaptation of the crops but their stability

A positive relationship for Main crop/cereal and Overall intercrop yield was observed in the Trial 24 (Maize and beans, IPC-PT) indicating a potential use of the bean's genotypes for co-breeding work. Both the positive relationship, as the positive trend indicate that intercropping is a very important solution for organic, that allow the yield increase in comparison with the crop per se and it is generally an important tool for organic. Nevertheless, important traits for co-breeding and finding the adequate combination has been an important achievement.

However, for Main crop/cereal it was registered a negative trend for the Trials 9 (spring wheat x spring pea, AREI, LV) the adequacy of the plant height is important to allow pea genotypes to grow, in case of Trial14 (s. wheat/s. triticale x faba bean, AREI, LV) there was a decrease of 21% in comparison to pure stand, the choice of cereal species for growing in mixture with field beans had no significant impact on the yield level of the beans., Trial 18 (intercropping with white lupin, FIBL-CH) where the white lupins showed to be more competitive, overgrows companion and undersowing plants, thus also reduction of spread of anthracnose disease was not achieved). Trial 22 (spring wheat/triticale x spring vetch, AREI_LV) the field beans could be the best solution for spring vetch growing in mixture, to provide high seed yield under organic conditions.

Both the positive relationship, as the positive trend indicate that intercropping is a very important solution for organic, that allow the yield increase in comparison with the crop per se and it is generally an important tool for organic. Nevertheless, important traits for co-breeding and finding the adequate combination has been an important achievement.

The **Seedhead density** (indicates the number of spikes per m²) had a positive trend Trial 9 (spring wheat x spring pea, AREI, LV) due to the increase of 20% of spring wheat to the sowing rate of semi leafless pea genotypes, without the reduction of pea sowing rate, could be a viable option for growing of semi leafless varieties under organic conditions. The **Grain weight** had a positive trend for Trial 10 (Pea, GZPK+FiBL-CH) and positive relationship for Trial 11 (winter triticale x winter pea, CULTIVARI, D) (Figure 23 and Figure 24 to address specially peas).

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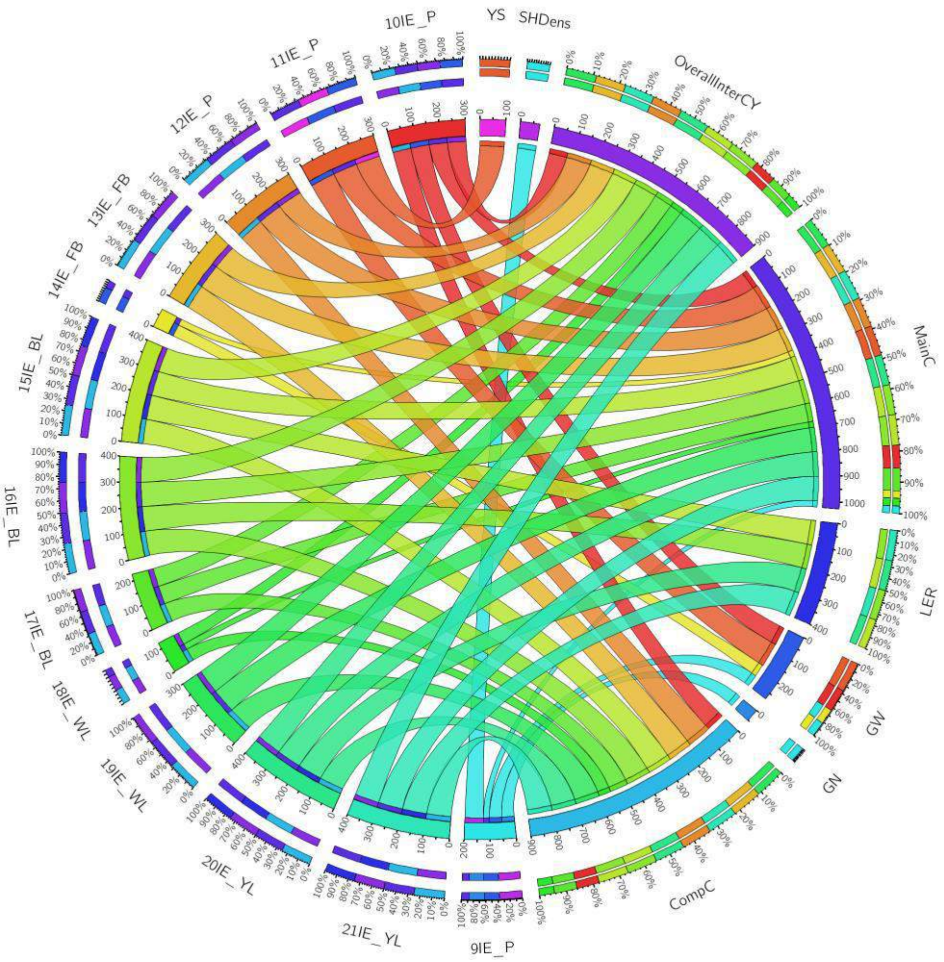

Figure 23. Traits from interspecific trials and the production performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

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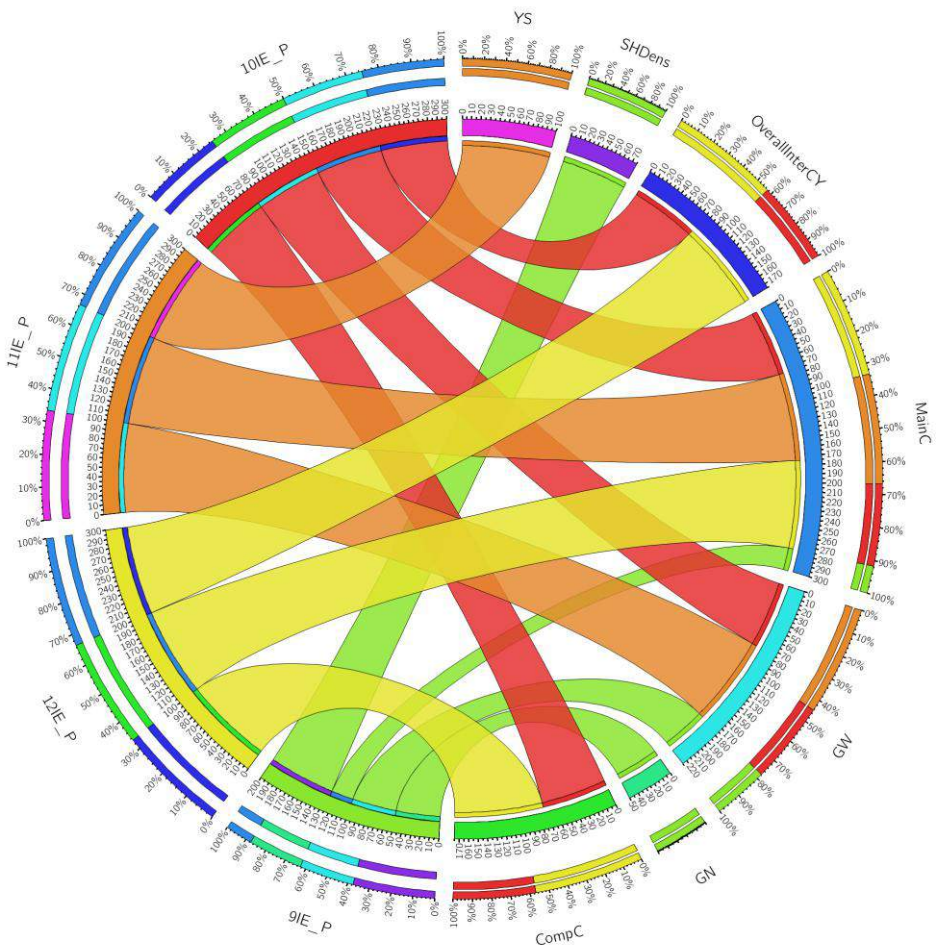

Figure 24. Traits from interspecific trials (Pea) and the production performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

The **Mixing efficiency (Land Equivalent Ratio - LER)** had a positive relationship for lupins (Figure 25). Trial 15 (spring triticale x blue lupin, IUNG, PL), Trial 16 (spring oat x blue lupin, IUNG, PL), Trial 20 (spring triticale x yellow lupin, IUNG, PL), and Trial 21 (spring oat x yellow lupin, IUNG, PL). It was observed that high values of LER noted for mixtures of oat/triticale with lupins demonstrated their good mixing ability, it was also understood that mixtures of lupins with oat performed better (higher yields and lower variability in years) than mixtures with spring triticale, and results showed that yellow lupin in a mixture performed better than narrowleaf lupin.

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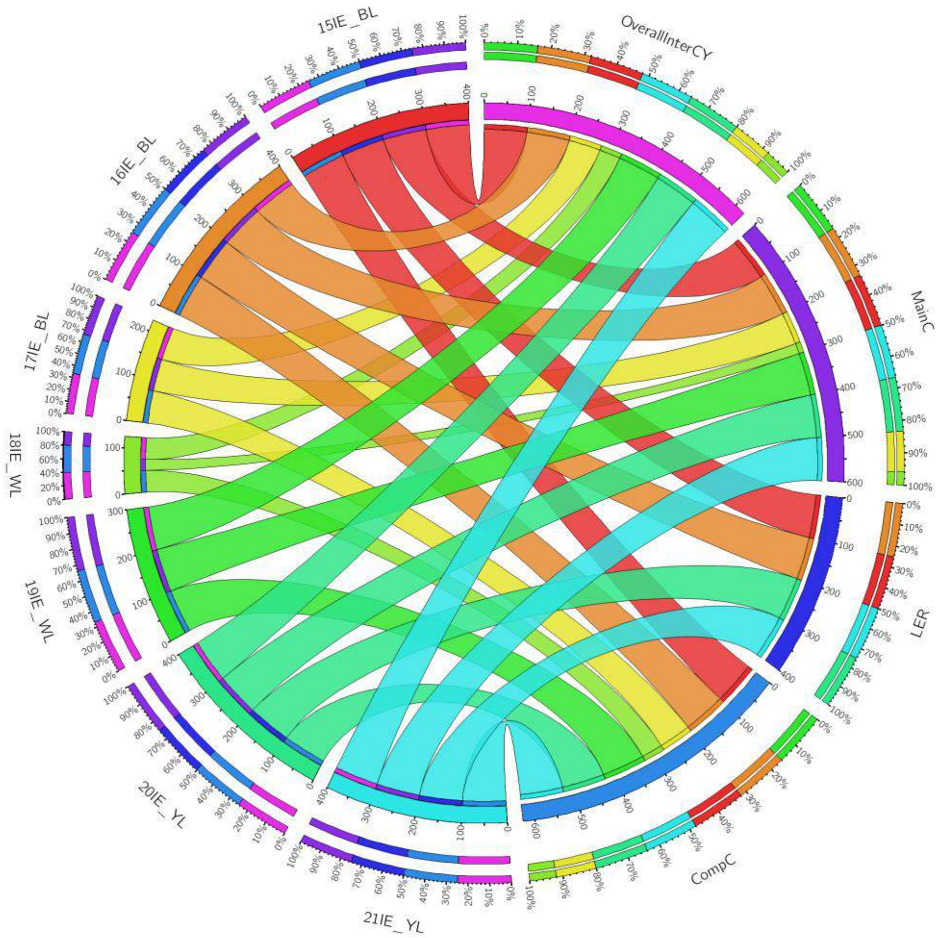

Figure 25. Traits from interspecific trials (Lupins) and the production performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

Quality performance

Crude protein content in grain had a positive trend for Trial 10 (spring triticale/barely – pea, FiBL-CH-GZPK) where the selection allowed to increase protein content, and a positive relationship for Trial 11 (winter triticale x winter pea, CULTIVARI, D) where selection for higher protein is very presents at breeders selection traits and also a trait searched by the farmers, Trial 12 (different cereals x pea, LBI, NL), Trial 13 (spring wheat x faba bean, LBI, NL), Trial 19 (spring wheat x white lupin, LBI, NL) (spring wheat x white lupin, LBI, NL), for LBI trials Wheat in mixed cropping conditions have a significant higher protein content, compared to wheat in pure stand, in addition an effort by The Netherlands to increase protein from plant based origin. In the case of Trial 14 no clear relation was achieved. A positive relationship for **Zeleny wheat** and **Falling number wheat** was present for Trial 13 (spring wheat x faba bean, LBI, NL) providing a better characterization of the wheat. **Thousand grain weight** had a positive relationship with Trial 11 (winter triticale x winter pea, CULTIVARI, D) (Figure 26 – all legumes, Figure 27 – peas only) indicating the effort of selection by the breeder.

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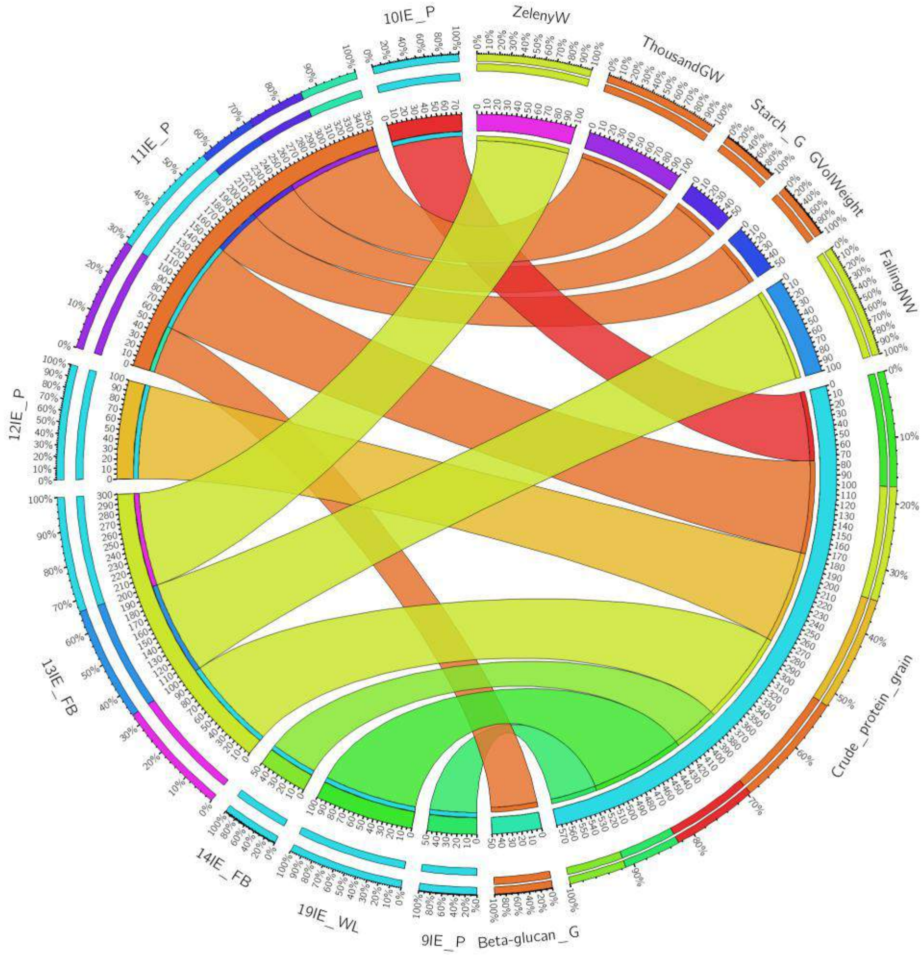

Figure 26. Traits from interspecific trials (legumes) and the quality performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

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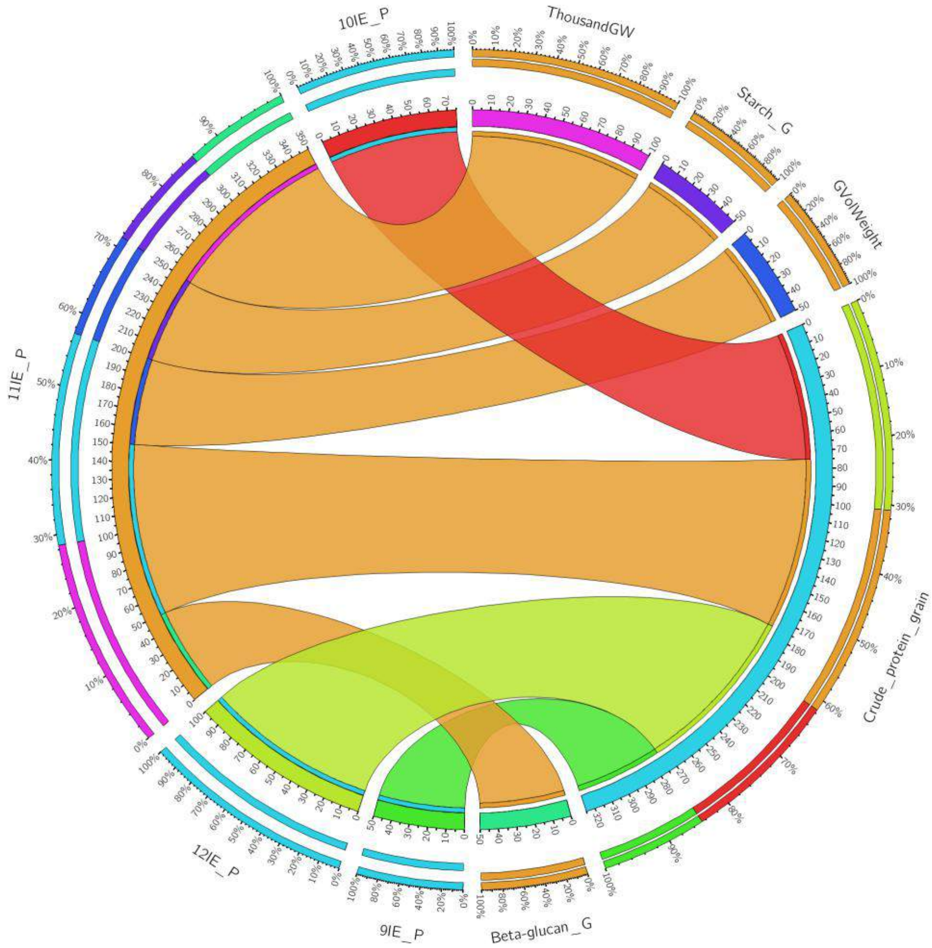

Figure 27. Traits from interspecific trials (peas only) and the quality performance associated, indicating also what were the most relevant traits for the trials. The width of each cord varies from 1 to 100 (1-negative relationship, 25-negative trend, 50- absent or no clear, 75-positive trend and 100-positive relationship) representing the relevance of certain traits for each of the trials.

Interspecific analyses (perennial crops)

For Plant descriptors, an absent on no clear trend was observed for the trait **Height** with grass-legume mixtures Trials 23a (AGROS, CH) and 23b (NARDI, CH) and for **70 Fruit formation** for the Trial 22 (s. wheat/triticale x s. vetch, AREI, LV).

For Agroecosystem performance, an absent on no clear trend was observed for the trait **Vigour** and **Biotic Stress Response** with Trials 23a (AGROS, CH) and 23b (NARDI, CH), **cover** with Trial 22 (s. wheat/triticale x s. vetch, AREI, LV).

In the case of Production performance, the traits **Main crop/Cereal and Overall intercrop yield** had a positive relationship with Trial 24 (maize and beans, IPC, PT) indicate some interesting combinations of maize and beans (Tables 14-21).

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2. Agroforestry

2.1. Methodology

Crops are generally bred with a homogeneous open field in mind as a target environment. An agroforestry system is inherently different from an open field. We have worked to begin quantifying these differences by exploring the range of 'agroforestry environments' that exist and the potential selection pressures that they create. Agroforestry results in a diversified in-field landscape composed of many environments (e.g., close to the tree, far from the trees, different tree species, seminatural elements, etc.). This leads to three questions:

- **Agroforestry as an environment or as a mosaic of micro-environments?** To what extent can a given agroforestry system constitute a target environment for breeding, or does it need to? To what extent, and at what spatial scale, should the micro-environments within that agroforestry system be targeted to breed adapted crops? Which role can participatory breeding and/or Organic Heterogeneous Material have in this?
- **Which breeding stage for which agroforestry challenge?** Are there crop species inherently better suited to coexistence with trees than other? How can different stages of crop breeding contribute to agroforestry-adapted crops, e.g., selection in the target environment (evolutionary breeding, mass selection, etc.) within and between populations, or testing existing varieties / elite breeding material / other genetic resources to better support informed choice?
- **Turning the question inside-out.** Can agroforestry systems offer opportunities for the breeding of sustainable and resilient crops thanks to their inherent spatial diversification? Can agroforestry systems offer new solutions for more effective variety testing for low-input systems?

To address these questions, a matrix consisting of different agroforestry systems on one dimension and different breeding stages or approaches on the other dimension has been built to help understand strengths, weaknesses, opportunities, and threats of each possible agroforestry system by breeding combination.

To simplify the complexity of agroforestry systems, we identified four main landscapes:

- Parkland agroforestry: possibly the simplest and most extensive type of agroforestry, characterised by mature trees dispersed/scattered on the arable surface.
- Alley cropping systems: by far the most common in temperate regions, where trees are distributed in arable land and arranged in rows.
- Succession Agroforestry System (SAF): distinguished by the cultivation, in the same place, with a diversity of species, that co-operate in a synergic and synchronized way, optimizing their productive cycles, creating rational income and positive energetic balance within the system. It requires an equilibrium between crops with ecological and economic function.

Table 25. Matrix of agroforestry systems and breeding approaches (Boffa, 1999)

	Parkland	Alley cropping	Succession Agroforestry System (SAFs)
Selection within populations	e.g., evolutionary breeding and/or mass selection in a field scattered with trees.	e.g., evolutionary breeding/mass selection taking in different sections of a field surrounded by tree rows.	e.g., evolutionary breeding and/or mass selection in different strata for intercropping for herbaceous crops nested in tree rows.

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Selection between populations or lines	Comparing/selecting different populations or pure lines in a field scattered with trees.	Comparing/selecting different populations or pure lines in a field surrounded by tree rows.	Comparing/selecting different populations or pure lines in a field within the intercropping nested in tree rows.
Cultivar testing	Comparing different cultivars in a field scattered with trees.	Comparing different cultivars in a field surrounded by trees.	Comparing different cultivars in the herbaceous intercrop nested in tree rows.
Crop choice	N/A	N/A	Crop selection based on the complementarity among crops (e.g., aerial and root).

2.2. SWOT-analysis of different agroforestry systems

Parkland agroforestry systems

Parkland systems are typical of tropical and subtropical agriculture, especially in semiarid climates (Boffa, 1999), but are also very common in Southern Europe, especially in the Dehesa and Montado landscapes in the Iberian Peninsula. These systems are characterised by trees scattered on the arable surface and are generally very extensive in temperate climate (unlike the highly intensive traditional farming in e.g., sub-Saharan Africa). Evolutionary breeding and selection within population might be the most feasible option, although breeding achievements might be contained into specific areas and not replicable. On the other hand, it might be challenging to effectively deploy plot-scale testing, as would be required for selection between populations or lines and for cultivar testing. However, any field-scale testing could have the same benefits as selection within populations (Table 26).

Table 26. SWOT analysis of different breeding approaches in parkland agroforestry systems

Parkland agroforestry		
Selection within populations	**Strengths**: - evolutionary breeding could effectively pick up spatial variation and generate very adapted populations.	**Weaknesses**: - the punctiform nature of spatial variation is hard to address explicitly, so breeding focus might be limited to the whole landscape.
	Opportunities: - each landscape could effectively, “breed” its own set of adapted populations.	**Threats**: - high specificity might make it difficult to add value to a population in other, albeit similar, landscapes.
Selection between populations / lines and cultivar testing	**Strengths**: - a high-nature-value landscape whose complexity is not limited to the presence of trees, could be a pivotal case of breeding in target environments.	**Weaknesses**: - the punctiform nature of spatial variation is hard to address explicitly.
	Opportunities: - use of spatial designs or adequate statistical tools can facilitate selection - comparison of lines/cultivars at a field scale could be very successful.	**Threats**: - limited amounts of seed and low replicates might make it very difficult to appropriately select. Likewise, increased number of replicates may increase operating costs.

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Alley cropping systems

Across a section of an alley cropping system, there are at least three micro-environments: the mid-field (where there is no direct interaction between trees and crop, besides microclimate, windbreak, etc.), the tree row and the tree-crop interface (where the trees and the crop directly interact with competitive and/or facilitative interactions). A further distinction can be useful between “wide” and “narrow” alleys:

- “Wide alleys” systems are a spatial arrangement where the “mid field” section would be dominant.
- “Narrow alleys” systems are a spatial arrangement where the tree rows are close enough to one another to make the “tree-crop interface section” dominant.

Alley cropping systems bear important opportunities for evolutionary and/or direct selection harnessing complex interactions, that can be added value in environments different than the selection one. Paradoxically, it may be harder to add value to the selection achievements in the alleys themselves. Very suited to plot-scale or single-plant scale testing. Different targets could be explored, such as the most homogeneous performance profile across the alley, the best performance in specific positions, etc.

An example of experimentation in narrow-rows alley cropping system is the one carried out as part of the EU FP7 AGFORWARD project (AGroFORestry that Will Advance Rural Development, 2014-2017) in the narrow alleys (12 m) in the Wakelyns Agroforestry farm in Suffolk, UK. These data were re-analysed during the LIVESEED project yielding the evidence shown in Table 27.

Table 27. SWOT analysis of different breeding approaches in alley cropping agroforestry systems.

Alley cropping		
Selection within populations	**Strengths**: - The distinction of clear sections between of different tree-crop interaction intensity can be harnessed, - temporal variation with trees management cycles can be further addressed.	**Weaknesses**: - would ideally generate selections adapted to different sections of the alley, but it would be unpractical to drill them separately.
	Opportunities: - Pick up very fine microclimatic effects such as daytime variation between east of trees (more even) or west of trees (more extremes) that can be used for selection for different stresses.	**Threats**: - specific sections of the alley (e.g., west of trees, with prolonged morning moisture) could be more prone to seed-borne diseases.
Selection between populations / lines and cultivar testing	**Strengths**: - like for mass selection, could be an exceptional playground to test against different types of interactions.	**Weaknesses**: - deep integration with the systems management is required with an important planning and design effort, - replicates are needed and may increase costs.
	Opportunities: - Results and selections can be easily generalised and made useful in different contexts.	**Threats**: - overlooking the spatial variation can generate very flawed results.

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Successional Agroforestry Systems (SAFs)

The Succession Agroforestry System (SAFs) environment can generate several environments, in addition to alley cropping systems, I.e., mid field, the tree row and the crop row interference, the interaction with several other crops is an additional factor. SAFs provide important opportunities for evolutionary and/or direct selection harnessing complex interactions that become even more complex with intercropping, that can be an added value in environments different than the selection one. Paradoxically, it may be harder to add value to the selection achievements both for the alleys and intercropping themselves. Very suited to plot-scale. Different or targets could be explored, such as the best synchronization in the intercropping, the plant traits best adapted to intercropping both for alleys and interrow traits, and the best performance in specific positions, etc.

An example of experimentation of the SAFs was installed at ESAC-IPC (Portugal) under organic and low input environment in 2019 and can be divided in two steps: 1) Creation of the SAFs itself; 2) To start a maize CCP 'SinPre' and bean cultivars in co-breeding for SAFs and latter only with maize adapted to SAF. During the years of selection, it was possible to obtain seed for next seasons, determination of yield trial will be done this year (Table 28, Table 29). An example for evolutionary selection within population and selection between populations are also presented in Table 30 and Figure 28 below in the Wakelyns Agroforestry (UK).

Table 28. SWOT analysis of different breeding approaches in Successional Agroforestry Systems (SAFs)

Successional Agroforestry systems (SAFs)		
Selection within populations	**Strengths**: -Evolutionary breeding in a system more near its ecological basis. The selection vectors are space and forest interaction, companion crop interaction and synchronization and successional crop interaction.	**Weaknesses**: - Would ideally generate selections adapted to different gradients of the space, forest interaction, companion crop interaction and successional crop interaction vectors, however breeding focus might be limited to the whole complexity of the system.
	Opportunities: - Each "vector of selection" could effectively "breed" its own set of adapted populations.	**Threats**: - High specificity might make it difficult to add value to a population in other, albeit similar, landscapes.
Selection between populations / lines and cultivar testing	**Strengths**: -Possibility of evolutionary breeding in a complex system, near its ecological basis. The selection vectors are focused on forest interaction, companion crop interaction and crop synchronization that can be a pivotal case of breeding in target environments.	**Weaknesses**: - Deep integration with the system management is required with an important planning and design effort, - Replicates are needed and adapted to the complexity of the systems that increase costs.
	Opportunities: - Develop adequate statistical tools that can facilitate selection -Search new traits for adaptation to complex breeding systems, - Comparison of lines/cultivars and their interactions among them and with trees, both at technical and economic.	**Threats**: - Limited amounts of seed and low replicates might make it very difficult to appropriately select. Likewise, increased number of replicates may increase operating costs.

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Table 29. Trial description of the SAFs trial in Portugal

Trial location	Successional Agroforestry System (SAFs), Caldeirão, IPC-ESAC, Portugal.
Breeding case	Evolutionary selection within a maize CCP population "SinPre".
Crop	Maize, *Zea mays.*
Breeding population	Progeny of a Composite Cross Population "SinPre" further selected to adaptation to Successional Agroforestry System.
Agroforestry system	This trial had 2 main phases. 1) implementation of the SAF, during two courses at IPC-ESAC that included 16 perennial species including trees for biomass, fruit trees, shrubs, and aromatic herbs. The horticultural part included 11 species two bean genotypes ('Catarino' and 'Vermelho') initially intercropped with maize. 2) include maize CCP in the SAFs system.
Research Question	Will sub-population evolve divergently in SAFs than in normal field?
Method	**In year 0** (2019) The maize "SinPre" was sown in the prepared SAFs and together with beans. Maize was in temporal isolation, I.e., no crosses with other populations. Due, probably to manure application, the germination was very affected, and few plants germinate adequately. From this plants selection was done and seed was harvested for the next season. **In year 1** (2020), "SinPre" was sown, and 2 kg were obtained for the next season. The same design was adopted in **year 2** (2021).
Statistical analysis	Qualitative analyses were done, but no statistics were used.
Results	**Year 1**. It was possible to obtain more seed to continue the trial selection. **Year 2**. Field trial was installed, and yield will be determined.
Conclusions	The "SinPre" has been selected under SAFs and its seed quantity has been increased. In future it will be important to compare the initial SinPre with last cycle in agroforestry.

Table 30. Evolutionary selection within population AND selection between populations, Wakelyns Agroforestry UK

Trial location	Wakelyns Agroforestry, Fressingfield, Eye, Suffolk. IP21 5SD, United Kingdom.
Breeding case	Evolutionary selection within population AND selection between populations.
Crop	Spring wheat (Triticum aestivum).
Breeding population	Progeny of a Composite Cross Population further selected to adaptation to spring sowing.
Agroforestry system	Narrow alleys in short rotation forestry system surrounded by two mature hazel tree rows in year 1, and by a coppiced willow row on the west and a mature willow row on the east in year 2.
Research Question	Will sub-population evolve divergently after having been grown either east of trees, west of trees and in the centre of the alley? See Figure 28 below.
Method	**In year 0** (2014) A spring wheat composite cross population (CCP) was grown in plots across a willow system agroforestry alley in 2014. Plots of bulk CCP were harvested separately from plots on either side of the alley, adjacent to the tree rows (East of Trees (EOT), West of Trees (WOT)) and the alley centre (Centre of Alley (COA)). **In year 1**, a complete block design with three replicates was adopted. In each block, three replicates of the EOT, WOT and COA were positioned in each respective position. The same design was adopted in **year 2** with the EOT, WOT and COA harvested in year 1 sown in all three positions.
Statistical analysis	For the 2016 dataset, a maximal model assuming block as random term, and selection, position in the alley and their interaction as fixed term was adopted. A deletion approach was used to assess significantly better fit of the model assuming a fixed term against the

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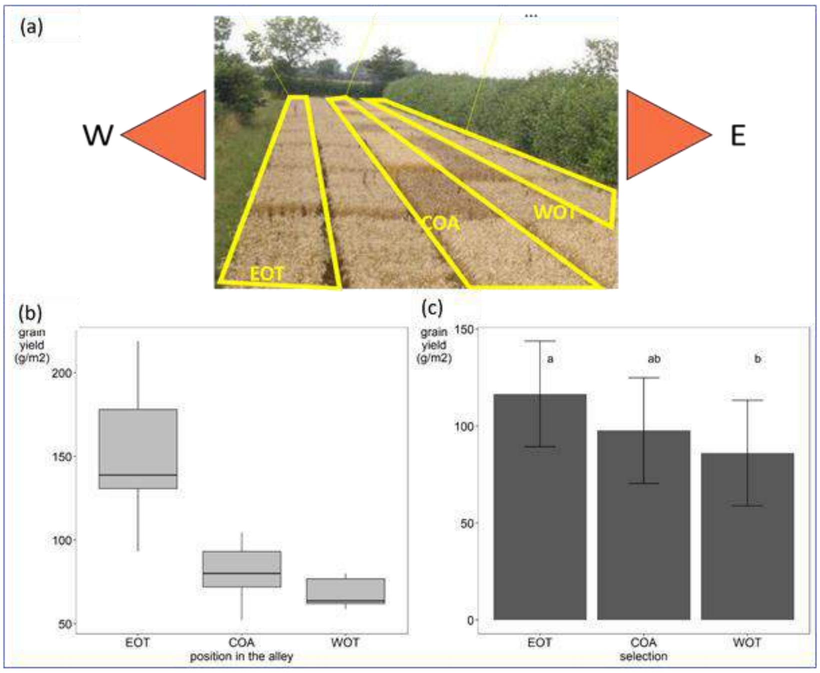
	model not assuming it, by likelihood ratio test. Models with significantly lower AIC were retained. If no interaction was detected, a simplified mixed-effect model was used in assuming ○ The EOT, COA or WOT selection as a fixed term, ○ The block as a random term, ○ The position in the alley as a further random term (with 1 degree of freedom).
Results	**Year 1**. Yield differences were all linked to the position in the alley, with no differentiation between the EOT, WOT and COA populations. Yields and hectolitre weights were significantly higher in the centre of the alley than at either edge. **Year 2**. A different alley with one side coppiced highlighted a much higher yield in the EOT position – alongside the coppiced row, than in either the COA or WOT position. This shows that the alley is dominated by tree-crop interactions. The effect of interaction between selection and position in the alley on grain yield was not significant (p = 0.181). However, the effect of selection on grain yield was highly significant (p 0.006). Offsetting against the position in the alley, the EOT selection was yielding significantly higher than the WOT selection (p < 0.011), see Figure 28 (c)).
Conclusions	The yield potential of a wheat population can be influenced by the position in an alley between two North-south oriented tree rows where it has been multiplied. - However, Whether the EOT selection is better adapted to a silvo-arable context, or the WOT may instead accumulate seed-borne diseases due to higher persistence of humidity on the western side of the tree row, is not clear.
Smith et al. (2017) Lessons learnt: Silvo-arable agroforestry in the UK (Part 1) Report, AGFORWARD Project. URL: https://www.agforward.eu/index.php/en/silvoarable-agroforestry-in-the-uk.html?file=files/agforward/documents/LessonsLearnt/WP4_UK_Silvoarable_1_lessons_learnt.pdf	

Figure 28. A plot trial in an alley in Wakelyns agroforestry, with the East of trees (EOT), Centre of alley (COA) and West of trees (WOT) positions highlighted (a). Grain yield in the three positions (b) and of the EOT, WOT, and COA selected populations (c) in 2016. (c) shows the estimated marginal means generated from linear mixed model assuming, as random terms, the orthogonal effect of block and position in the alley. Bars with different letters are significantly different at a 0.95 confidence interval.



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2.3. Guidelines for selection and cultivar testing for agroforestry systems

The selection and cultivar testing in agroforestry systems increases complexity. This complexity may favour incorrect experimental designs and consequently misinterpretations. Therefore, two cases of the most common errors and three cases of good practices design are presented below (Figure 29-33).

The two cases of the most common errors are presented both as “RBC design with blocks in the wrong direction” (Figure 29) and “Trying to make use of a wrong design: adding a covariate” (Figure 30).

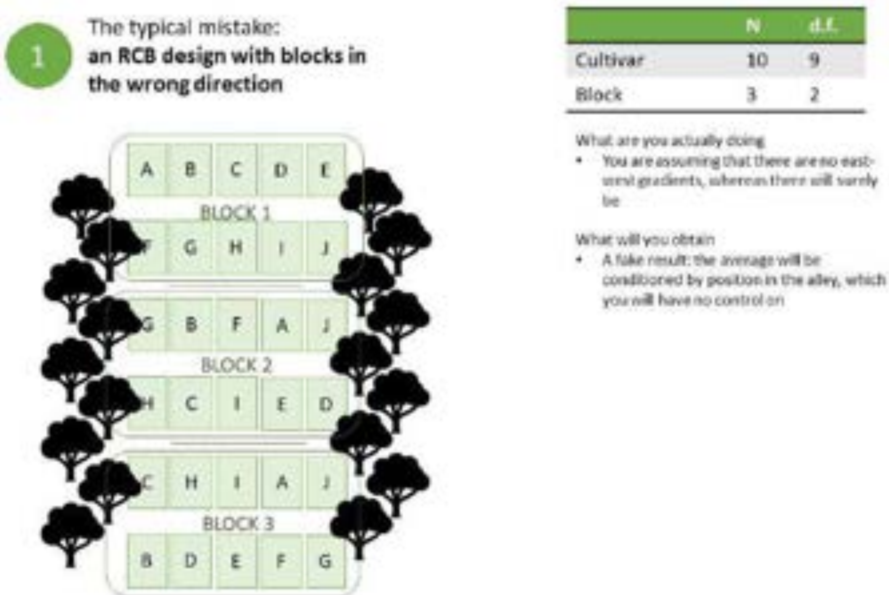

Figure 29. RBC design with blocks in the wrong direction, a typical mistake made in design analyses. In this case, the effect of the East and West gradients provided by the alley are not considered

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Figure 30. Trying to make use of a wrong design: adding a covariate. In this case, trying to not discard the data, a covariate is added. However, what is achieved in an average performance as corrected as possible against varieties position in the row

The three cases of good practices design include the RCB design with blocks in the right direction (Figure 31), an ideal design with fewer varieties in two or more "Latin Squares" (Figure 32), and an example with ten varieties to test with more alleles available, for hierarchical design (Figure 33).

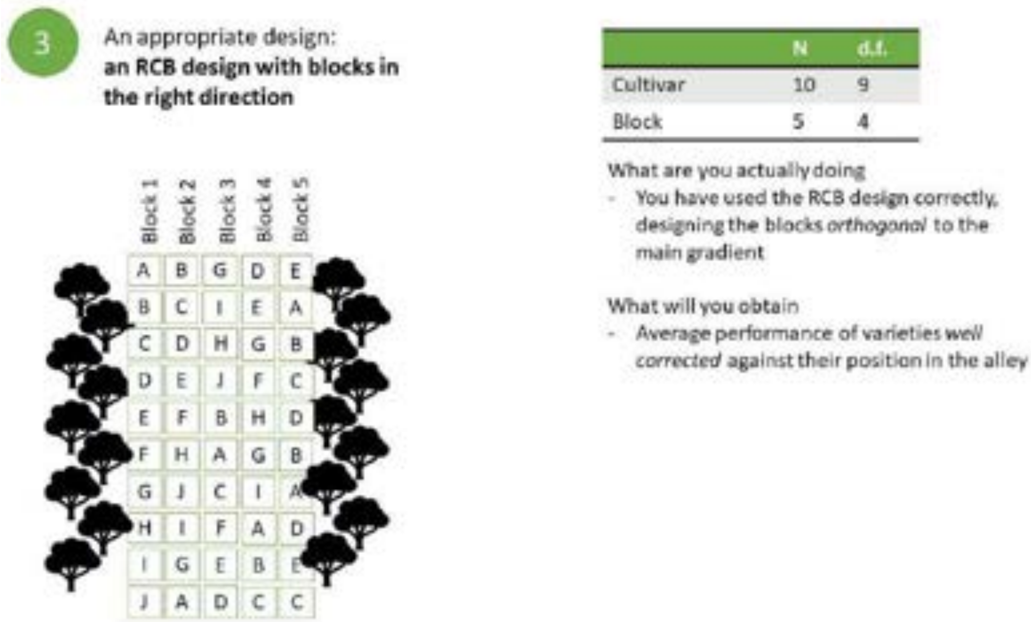

Figure 31. A good practices design includes the RCB design with blocks in the right direction. In this case we use de RCB design correctly, designing the blocks orthogonal to the main gradient, which allows having an average performance of varieties well corrected against their position in the alley.

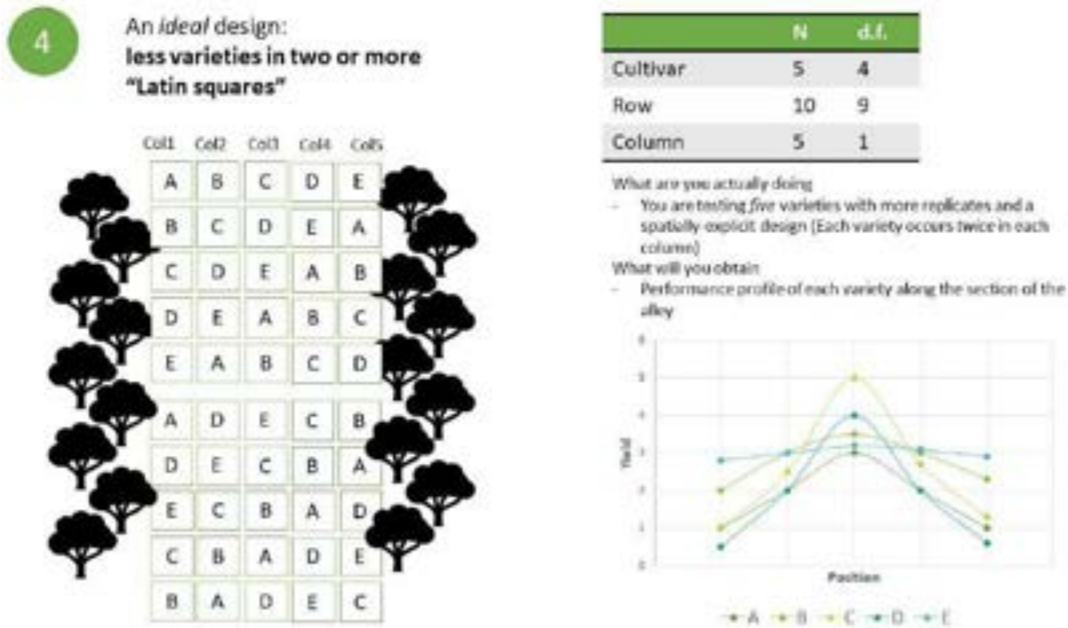

Figure 32. An example of an ideal design with fewer varieties in two or more "Latin Squares. In this example, five varieties testing with more replicates and a spatially explicit design is made to be able to obtain a performance profile of each variety along the section of the alley

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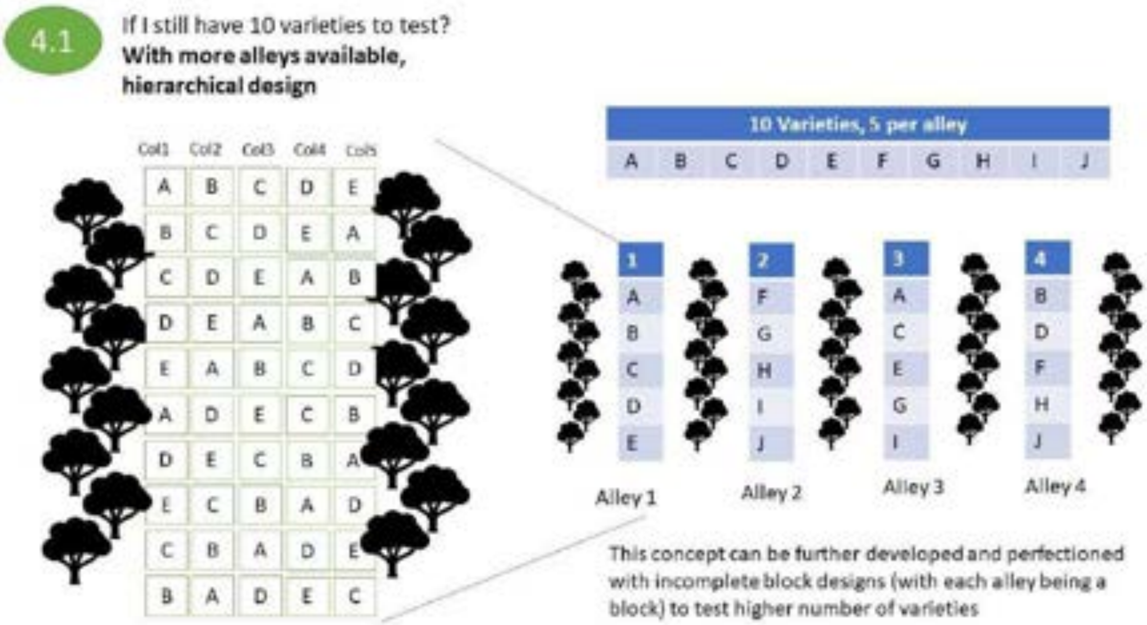

Figure 33. Example with ten varieties to test with more alleys available, for hierarchical design. In this case the alley can be represented by a block, an incomplete block design, that allows to increase the varieties to be tested

3. Implementation and development of multi-actor approaches

3.1. Information gathered through the Annual Reports

Based on self-reporting by partners through the Annual Field Trial Reports, nearly all partners were involved in some form of stakeholder engagement, and several types of activities were applied (Figure 34). In most trials, partners engaged organic farmers, advisors, and other value chain actors in demonstration events with the purpose to widely disseminate the trial information and their results. Another often reported form was to organise regular meetings with farmers, advisors, breeders, processors, or producers to make common decisions on species, variety choice or crossing choices. Workshops, instead of regular meetings, were also preferred when it came to the evaluation of the trials and discussion of outcomes. However, specific cases of participation requiring more in-depth involvement in the process were also reported, such was the case of involving farmers and researchers in the entire methodology design or co-design of farmers' mixtures (INRAE), in the entire process (IPC) or involving farmers in the temporary experiment on Organic Heterogeneous Material (AREI). These require much more commitment and regular effort from stakeholders and will be described in more detail in the following chapter as case studies.

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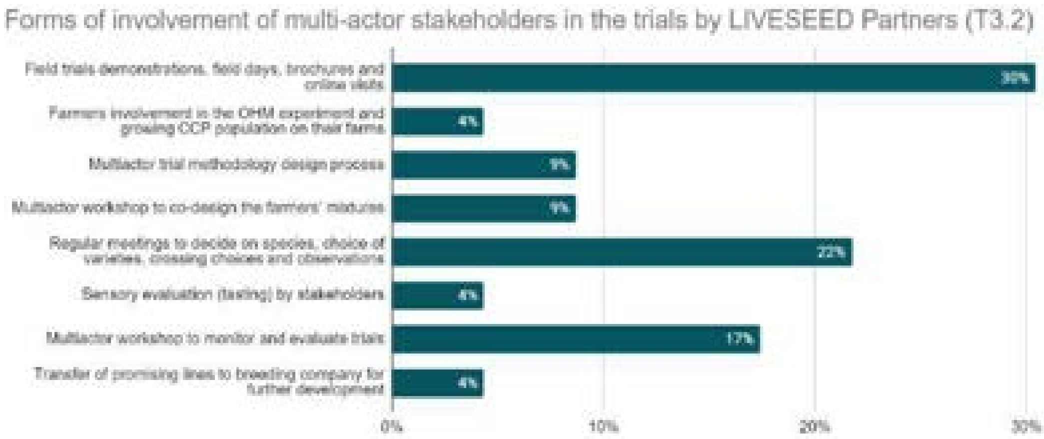

Figure 34. Types of multi-actor stakeholder involvement in the trials by LIVESEED partners (3.2) as reported in the annual reports

When it comes to the types of stakeholders involved (Figure 35), in most cases organic farmers, researchers, agricultural advisors and consultants were engaged, however, other interesting collaborations also formed. For instance, the biggest potato starch producer in the Baltic States ALOJA-STARKELSEN, interested in plant based organic proteins production from pea and field beans, discussed the choice of pea and beans varieties for protein production with AREI, or FiBL-CH's effort to involve mill owners in the evaluation of trial results are interesting examples. On the other hand, it is also clear from these reports that consumers' involvement was mostly possible through organising specific tasting workshops.

Figure 35. Types of stakeholders involved in the trials by LIVESEED partners (3.2) as reported in the annual reports

3.2. Information gained through an in-depth survey on participatory approach at LIVESEED partners

3.2.1. Methodology of auxiliary survey among partners on multi-actor approach

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An evaluation form was developed to collect information on how the partners used participatory and multi-actor approaches in their work in Task 3.2. The survey ran between 08th December 2020 and 15th January 2021, and included four sessions:

1) Background of partners and context: What triggered the setup of the participatory or multi-actor breeding approach involving farmers, processors, bakers, retailers, or consumers? What were the species and major breeding goals?
2) Former experiences: Experience in participatory or multi-actor breeding before LIVESEED (years), type of funding and institutional status; number of persons involved in breeding; geographical scale for distribution of cultivars (e.g., local level, national level, more than one country); selection environment.
3) Organisational model: referring to the number of genotypes tested per year; factors other than genotypes (e.g. different cropping systems, stress environments); what does the testing include (e.g. small trial, strip or pilot field trials, nutritional profiling, what is the task of farmers; if trials are on-farm, what are the incentives for farmers (e.g., they get paid, they get provided seed, completely voluntary, they use the results, etc); approach for breeding for diversity (e.g. breeding of neglected crop species, increase genetic diversity within species or within cultivars, breeding for species mixtures); who is involved in the multi-actor breeding?
4) Expected impact of LIVESEED: capacity building and perspective; Key Exploitable Results and respective description (e.g., methodological tools, screening / selection tools; statistical tools for data analysis; Innovative Cultivars available to farmers after LIVESEED is finalized) and from each of the examples it is requested.

3.2.2. Results of the survey

The results were obtained from 9 institutions and analysed by IPC (in-depth results are presented in Annex II).

Adaptation to organic environments, developing disease resistance, species selection for intercropping, creation, or comparison of different types of populations, and boosting local value chains were the main reasons that triggered the start of a participatory approach. With regards to breeding goals, priorities were given to issues related to disease resistance, weed competition and lodging tolerance, followed by yield stability. In 70% of the cases, the breeding for diversity approach considered intra-specific diversity (i.e., OHM like CCPs or open pollinated populations), and in 60% genetic diversity within species (e.g., selection for local adaptation, special usage, wide crosses).

Projects, like LIVESEED, are boosting such experiences, enabling partners to set up new participatory trials or continue ones that started within 1-4 years prior to the project. Only half of the responding partners had longer experience (5-10) in such approaches showing that there is a need to gain more relevant experience and to share and exchange knowledge with “newcomers”. Moreover, it is clear from the survey that research institutions rely mostly on public funding (66.7%) when it comes to setting up of participatory trials, and the development of new routes of financing are urgently needed to complement this public funding.

Most participatory breeding initiatives are small scale: the number of involved people is only between 1-15, and in 70% of the cases the developed cultivars are disseminated at the local or national levels. In 50% of the cases the focus on genotype selection is of less than 100 genotypes. Other than genotypes, most participatory breeding initiatives addressed the differences in cropping systems (80%) followed by addressing stress environments (40%). Horizontal proliferation and upscaling of such small initiatives remain a challenge and requires enough qualified facilitators or researchers to initiate and coordinate such approaches, as well as diversified funding sources involving the value chain (especially producers and processors) at the local and national levels.

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The role of farmers in the participatory breeding activities of the respondents was mainly to provide an organic field and to take (to some extent) part in the selection and the evaluation process together with the researchers. To a lesser extent, they are involved in defining the breeding goals, select the genotypes themselves or conduct pilot or demo trials of candidate cultivars. According to our results, farmers' main incentive was to collaborate with breeders/researchers to get access to seed of improved cultivars. This result confirms that access to improved cultivars is a priority for organic farmers, and that they are willing to sacrifice time and land area to facilitate this access.

The role of the researchers/facilitators remains crucial to coordinate and guide the process and taking the lead in tasks that the farmers are not equipped to do (knowledge on breeding methods or materials and germplasms), or farmers lack (time, personnel capacity, network, or access to genetic materials). In most cases farmers collaborated with researchers and breeders instead of collaborating with other farmers, which confirms that they are looking for the expertise provided by knowledge institutions. Farmers, though to a less extent, were also motivated to develop own cultivars for short value chains and to get involved in seed multiplication/seed business. It was particularly in France where farmers were highly motivated to develop own cultivars and populations. This may be the result of long working relationships with breeders. It could be that a long-term relationship is needed for farmers to become interested in doing their own breeding.

A more in-dept analysis can be provided when layering the different stakeholders per activity compared to the self-reporting in annual reports by the partners. Based on the heatmap below (Figure 36), the role of the researchers is essential in most steps (except for seed production). It is also clear that field visits and workshops are the main platforms where all stakeholders engage with each other and the trials. Depending on the set-up, public breeders are also included in most of the steps. Millers and bakers are mostly involved in the definition of breeding goals and in processing quality. Interestingly, dissemination and facilitation mostly fall under the responsibility of the researchers. On the other hand, it also shows that farmers could be provided education to close knowledge gaps (e.g., registration, selections, resistance screening) and advisors could take more part in facilitation and dissemination for instance.

	Researcher	Public breeder	Private breeder	Seed company	Farmer	Advisor	Miller	Baker	Other processors	Retailer	Chief / cook	Students/volunteers	NGO	Public authorities
Definition of breeding goals	10	1	1	1	9	3	4	1	0	0	0	0	1	0
Collection of germplasm	9	1	0	0	0	0	0	0	0	0	0	0	0	0
Conduction of crosses	7	1	0	0	1	0	0	0	0	0	0	0	0	0
Selection for agronomic performance of early generations	6	1	0	0	2	0	0	0	0	0	0	0	0	0
Selection of late generations	6	1	1	1	4	1	0	0	0	0	0	0	0	0
Resistance screening to pest and diseases	7	1	1	0	3	0	0	0	0	0	0	1	0	0
Resistance screening to abiotic stress	5	1	0	0	2	0	0	0	0	0	0	1	0	0
Nutritional quality analysis	7	1	0	0	0	0	1	0	0	0	0	0	0	0
Processing quality	4	0	0	0	0	0	4	4	0	0	0	0	0	0

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Organoleptic quality	4	1	0	0	2	1	1	0	0	0	0	2	0	0
Maintenance breeding	5	2	1	0	3	0	0	0	0	0	0	0	0	0
Consumer studies	5	0	0	1	0	0	1	1	0	0	0	0	0	0
Statistical analysis	9	1	0	0	0	0	0	0	0	0	0	2	0	0
Final selection	6	1	1	1	4	0	2	1	0	0	0	0	0	0
Registration	4	2	1	0	1	0	0	0	0	0	0	0	0	0
Cultivar trials	8	1	1	1	6	0	0	0	0	0	0	1	0	0
Seed production	3	1	0	2	5	0	0	0	0	0	0	0	0	0
Field visits	9	3	3	4	8	6	4	2	2	3	1	3	2	3
Workshops	8	2	3	3	8	5	4	3	2	1	1	2	1	2
Dissemination	8	3	2	1	2	3	1	2	0	1	0	2	1	0
Facilitation of multi-actor processes	9	1	0	0	0	2	0	0	0	0	0	0	0	0

Figure 36. Heatmap representing stakeholder involvement in the partners' different PPB activities

In most cases, working with a participatory breeding approach improved the efficiency of the breeding methodology, secured the future of the breeding project after LIVESEED, and improved the knowledge of involved actors. It also provided farmers with innovative cultivars.

3.3. Case Studies

Participatory approaches in France

Bringing together knowledge between actors from different disciplines and perspectives (agronomists, geneticists, eco-physiologists): Participatory ideotyping of mixtures of bread wheat varieties adapted to organic conditions

Variety mixtures represent an interesting diversification approach to stabilise production, especially in organic conditions. However, farmers lack practical recommendations for designing efficient winter wheat mixtures that favour complementarity and synergies between the components based on key traits for plant-plant interactions. To develop variety mixtures adapted to various environmental conditions and practices, a participatory ideotyping approach was implemented.

This approach was first proposed for pure stand varieties, and it was transposed to variety mixtures. With the aim to design wheat variety mixtures specifically adapted to the local farmers' production context (pedo-climatic conditions, objectives, practices), first interviews were conducted with the farmers to describe their farming systems, practices, and end-use requirements for wheat production.

To co-develop variety mixtures adapted to the farmers own various environmental conditions and practices, the next step was to organise workshops with a group of farmers from the Ile-de-France region, and advisors and researchers to identify assembly rules for improving complementarity and synergy between mixture components for mixtures of 3 to 5 bread wheat varieties tailored to their local context. The agreed assembly rules helped to co-design each farmers' mixture based on the assembly rules relevant to their context and based on a multi-criteria assessment tool to help farmers with designing mixtures. On-farm trials (for 4 consecutive years) were then managed by the farmers and facilitated by a partner organisation GAB Ile-de-France.

INRAE carried out the assessments and the statistical analysis based on the comparison of the mixtures and the pure stand components, analysed the mixtures' composition in functional traits and validated the assembly rules. To interpret the performances of the mixtures, workshops were organised, and the participating farmers were also interviewed at the end of the growing seasons to

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record their satisfaction levels. Most of the farmers were satisfied with their mixture and wished to use them for wheat production on their farm.

The participatory approach was particularly relevant for identifying and addressing farmers' and practitioners' needs for optimising mixture design in terms of practical recommendations, experimental results, and a flexible multi-criteria assessment tool.

Participatory selection of wheat CCPs

Genetic diversity incorporated in CCPs can improve crop resilience and be attractive for farmers to work with participatory approach. With the aim of developing wheat CCPs specifically adapted to the local farmers' production context (pedo-climatic conditions, objectives, practices), farmers were involved in the choice of the starting genetic material that were developed into 3 new CCPs (from 6 wheat landraces). Besides adaptability to poor and productive lands, the different CCPs were created to fit the different types of market of the farmers: on-farm baking or selling grain to on-farm bakeries, as well as long chain markets.

The crossings were carried out by INRAE. The selection, multiplication, and observations of these novel CCPs took place with the involvement of farmers: The CCPs were sown in different environments to evolve in the conditions for which they were created and to be observed also in other environments to test their adaptability. Partner organisation UBIOS took the role of facilitator with the farmers, organised regular meetings, and helped farmers with observations and harvest. The morphological characteristics of these CCPs were recorded by the farmers.

The farmers' involvement at the different stages (choice of the CCP parents, observations, and breeding) were really motivating for them. It proved essential to have a facilitator to "coach" the farmers and make the link between farmers and scientists.

Participatory approaches in Portugal

Culinary breeding: Exploring Portuguese maize landraces as fresh maize

The aim of this work was to make a sensory analysis with five maize landraces grown under organic conditions. Colour, texture, and flavour were some of the traits observed and studied to know what the purchase intention would be among those who tried the maize landraces.

The maize landraces ('Milho Vermelho', 2495, 'Amiúdo', 'Pigarro', 'Broa 70') were harvested in August 2020. In total 30 participants contributed to identify the characteristics of the maize that attracted their attention most. Each sample was presented sequentially to the participants and each participant tasted five varieties. The scale used was hedonic with nine points (with 9 as "I liked it very much" and 1 as "I disliked it a lot"). A buy intention test was also carried out, where a five-point scale was used, ranging from certainly not buying (rating 1) to certainly buying (rating 5). It can be concluded that culinary breeding networks can be a way to promote underutilized maize landraces.

Participatory installation of Successional Agroforestry systems (SAFs) where maize CCP was included

Portugal has a long tradition in Agroforestry Systems that could serve as a solution for the recuperation of burned areas in the country and for the recuperation of uncultivated soils. The Successional Agroforestry Systems (SAFs) are practices based on the interaction among species to explore synergies above and below soil, creating a process of regeneration that results in high

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productive agricultural areas with higher independence on inputs and irrigation, and contributions of the ecosystem services (e.g., formation of soil, regulation of microclimate and favouring water cycles).

Aiming to promote SAFs in Portugal, a system was created for the first time in the Certified Organic Campus of IPC, with both yield and pedagogic purposes. The SAF included 16 perennial species including trees for biomass, fruit trees and shrubs. The horticultural part included 11 species plus maize. Workshops were held with a total of 50 participants (workers from representative enterprises, producers, students, professors, and other interested parties). These 50 participants had the opportunity to help in the installation process and to learn by doing. These participants were following this study closely, visiting the site regularly, and disseminating the information on the importance of agroforestry systems. A maize CCP was also included on the SAF, with the intention to multiply the seed and make their adaptation and selection, to be evaluated in the future by the researchers.

4 Improved plant populations selected for mixtures and in mature agroforestry systems under organic farming conditions

4.1. Improved plant populations (CCPs and DOPs)

Table 31 summarizes the populations improved in the scope of LIVESEED with enhanced intraspecific diversity that can be commercialized as organic heterogeneous material (OHM) from 2022 onwards. The exploitation plans to bring these populations to the market have been developed by the different partners involved.

Table 31. Summary of exploitation plans for breeding materials developed within the trials targeting intra-specific diversity

Partner	Planned cultivar release	Planned year of release	Further use of the material
UBIOS & INRAE - Trial 3	Three composite cross populations (CCP) were created to increase genetic diversity and to promote adaptation in different cultivation environments.	From 2022 Possible notification as OHM.	The CCPs will continue to be selected on farm. Adoption by farmers on their farms (farms cultivars). Multiplication and marketing under OHM to other farmers of the cooperatives, and maybe outside the cooperatives.
INRAE & ITAB – Trial 4a-d	Multiplication and distribution of Diversified Oriented Populations (rivet wheat, spelt, oat). Diversified Oriented Populations (DOP) are composed of accessions (around 200) multiplied from gene banks, mixed according to farmers' criteria. Those	From 2022 Identify interesting populations that may be notified as OHM.	Distribution of DOPs and follow-up with involved farmers. Use the same methodology to mobilize diversity of other species from gene banks, chosen out of farmers' needs.

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	populations are adapted to farmers' needs and created on their demand.		
AREI - Trial 6	Several spring barley composite cross populations (CCPs) were developed and tested in various environments at AREI within LIVESEED and national projects.	Already released One of CCPS was already listed in the temporary experiment on Heterogenous Cereal Populations marketing. From 2022 Additionally, one or two CCPs are foreseen to be notified as OHM in 2022. Another population, with AREI reference CCP-4, was tested by a seed producing farmer in 2020 and the farmer is interested to continue multiplication and marketing of this population starting from 2022.	The materials tested in LIVESEED trials are being re-used in further development of new CCPs and improvement of existing CCPs (mass selection, crosses done to and between CCPs) and selection of pure lines out of CCPs.
IPC - Trial 7	Widening maize genetic base for evolutionary breeding in Portugal. Populations selection from a broad source of genetic resources. Continuation of breeding of maize populations not yet registered. Newly developed composite cross population.	From 2022 Notification as OHM of the best performing populations	Continue to select CCPs from the material developed during LIVESEED. Use recurrent selection to improve populations and extract some inbred lines. Further evaluation and selection of maize germplasm for human consumption.
FiBL CH - Trial 8	New CCP is being set-up at FiBL in 2021 from tolerant lines adapted for spring sowing and carrying the same mutation for low-alkaloid content.	From ca. 2028 Potential for notification as OHM after some years of selection and adaptation.	This new CCP will be monitored for yield, anthracnose tolerance and alkaloid content in the following growing seasons starting from 2022.

4.2. Selections for interspecific mixtures and mixtures in mature agroforestry systems under organic farming conditions

Table 32 below summarizes the populations and cultivars selected for interspecific mixtures in the scope of LIVESEED which are available for the official variety registration or as candidate for the temporary experiment on organic varieties suited for organic production with adjusted DUS and VCU testing. The exploitation plans to bring these cultivars to the market have been developed by the different partners involved.

Table 32. Summary of exploitation plans for breeding materials developed within the trials targeting inter-specific diversity

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Partner	Planned cultivar release	Planned year of release	Further use of the material
GZPK - Trial 10	Spring pea lines selected for mixed cropping with barley. During LIVESEED activities barley was selected as best cereal partner for the selection of spring pea lines adapted to intercropping in Switzerland. By screening a wide base of pea germplasm, promising varieties/traits were included in GZPK pea breeding program in mixed cropping with barley.	From 2030 The first crosses within the LIVESEED project were made three years ago. Thus, results from these crosses will be seen in varieties released in 10 or more years.	Continuous update of the germplasm material tested (supported by LIVESEED) to include new materials with positive traits as parental lines in the pea crossings for selection in mixed cropping.
CULTIVARI - Trial 11	Winter-pea lines suitable for mixed cropping with winter triticale.	From 2021 application for VCU trials and registration. From 2035 Possible release of first cultivars.	Three of the tested breeding lines will be used as parents in further breeding to develop winter peas with better winter hardiness and times of ripening suitable for mixed cropping with winter triticale. Additionally, CULTIVARI will test in a new project by different winter triticale varieties and breeding lines for their use as partner for winter peas.
LBI - Trial 19	White lupin lines for mixed cropping. Sweet lupin candle types, suitable for mixed cropping and suitable for calcareous soils under Dutch short season conditions.	From 2025 application of first candidates for official registration trials.	The material is released by LBI to a small breeding company; they aim to develop one or more lupin varieties that are suitable for mixed cropping with maize. The mixed lupin / maize crop is used as whole crop silage for ruminant animal feed.
AGROSCOPE - Trial 23a	During LIVESEED, two experimental populations of alfalfa based on parents that were selected for a good performance in a spaced plant nursery in combination with grasses were developed. This genetic material of alfalfa is adapted to Swiss forage mixtures (local climate, frequent cutting, mixed cropping with drought tolerant grasses).	From 2034 application of first candidates for official registration trials.	The alfalfa material will be used to continue breeding activities to select alfalfa adapted for use in mixture with grasses.

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NARDI - Trial 23b	Development of one dynamic wheat population, one wheat CCP and one barley CCP.	From 2022 Notification of the populations as Organic heterogeneous material (OHM).	All materials from LIVESEED will be used for further breeding.
IPC - Trial 26	Maize CCP developed in Agroforestry.	2030 application of first candidates for official registration trials. Two years were accomplished in mass selection.	The maize CCP will be continued to be under selection and adapted to the agroforestry system. Plan the distribution for PPB for agroforestry from IPC.

5. Common Discussion

5.1. Traits and characteristics for improved mixing ability

5.1.1. Intra-specific diversity trials

An important question was how to predict mixing ability for cultivar mixtures with a focus on cereal crops. Several partners have been comparing cultivar mixtures with the same cultivars in monoculture. Unfortunately, no generalisations can be made based on the trial results. In the case of barley trials in Latvia, the applied strategy of **combining mixtures with components differing for 3 target traits** did not ensure the expected effect in all cases and therefore could not be generalized. **Stable mixtures arose by combining components with different adaptability,** but not in all cases. The same question was dealt with in the Netherlands for spring wheat, but overall, the cultivar mixtures did not show a better performance than the best performing cultivars. One mixture consisting of four commercial spring wheat varieties stood out compared to the pure stand varieties for total yield. However, no general relationship with specific traits could be found. Due to low disease pressure in all the seasons the trials were conducted, it was also not possible to determine whether the cultivar mixtures had improved disease tolerance.

The trials in France confirmed that farmers had much interest in cultivar mixtures. A cultivar mixture ideotyping participatory approach was successfully implemented. This means that assembling good cultivar mixtures is based on integrating the knowledge of farmers, technical experts and researchers through workshops, interviews and a set of assembly rules considering the local context. For now, it seems that this participatory ideotyping approach is the best way to predict the mixing ability of cultivars.

Another question was to better understand the potential benefits of CCPs compared to cultivar mixtures and cultivars in single stands. The barley trials in Latvia showed that **CCPs had a potential for organic and stress environments** (for yield, yield stability, diseases and weed competition) and CCPs might be better suited than homogenous varieties. Similar results were observed for winter wheat in France. The results from Latvia suggest that that for developing populations with improved resilience, the components need to be contrasting genetically, in particular, to traits related to **disease resistance**. In the Netherlands, the spring wheat populations showed slightly lower yields compared to commercial cultivars. These spring wheat populations were developed in Germany, about 400 km

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south in other climatic conditions. Together these results suggest that it is best for CCPs to be developed under the same growing conditions as the target environment.

5.1.2. Mixtures selected in mature agroforestry systems in LIVESEED. Inter-specific diversity trials

An important reason to grow legume-cereal mixtures is their improved competitiveness to weeds compared to pure stand legume crop. Such cropping system fits well within organic farm management. In the trials oriented towards interspecific diversity, different combinations of cereals and leguminous crops were tested based on the local agro-ecological conditions and farming contexts. First some general conclusions are provided followed by specific results for pea, lupin, alfalfa forage grass mixtures and agroforestry systems.

An important question was how to breed for improved mixing ability. Based on the results of the pea and lupin mixtures, it is clear that for each crop mixture, plant type can determine whether breeding for mixing ability is necessary or not. For example, ranking of semi-leafless pea genotypes in a mixture with wheat and in the pure stand was similar. However, this was not the case for full-leaf pea genotypes. In such case, breeding for mixed cropping requires to evaluate both species in mixed stand, as the ranking in performance of pure stand crops can differ from the ranking in mixed stand.

The traits to look at in a crop for good mixing ability can be very broad. For example, in the case of searching for better triticale genotypes for mixed cropping, very different traits require attention, such as: broadness of leaves, date of ear emergence and rapidness of juvenile growth, plant length and tillering ability (e.g., ears per square meter).

The trials on white lupin in the Netherlands and Switzerland showed very contrasting results: e.g., in the Netherlands it seems quite well possible to mix existing genotypes of white lupin and spring wheat whereas this was not possible in Switzerland. Depending on the local context, specific plant types (e.g., dwarf, tall, branching, early, late, etc) are more appropriate and determine whether breeding for mixing ability is necessary or if it is possible to combine the best performing cultivars of two crops with similar phenology, of which a similar ripening time is the most important aspect to look at.

It was also concluded that it is important to define the objectives which performance and ecosystem functions should be improved, because in many cases it is not the total yield of the mixture, but the yield of the two components that will be processed separately, particularly in the case of food. A good example is that in the Netherlands a mixed stand can improve the baking quality of the wheat when combined with a legume crop. However, in the case of food purposes, an important aspect is the quality of the grain separation to avoid the chances of allergenic reactions when some grains of a lupin crop remain in the cereal grains. Hence, the companion crop must also have market potential next to good agronomic potential in mixtures.

Pea

Here a number of country dependent results are listed. In Latvia, full leaf short pea genotypes were more sensitive and the decrease in **yield** components was more pronounced for them in a mixture with spring wheat than the tall pea genotypes. Some pea genotypes were more vigorous after germination than others and a trait such as early **vigour** may be useful for pea genotype selection for mixed cultivation under organic conditions. In Germany, taller winter pea varieties were also recommended in mixtures with triticale for more or less extensive locations. Normal leafed types winter pea due to their enhanced **weed suppression** and their slight yield advantage should be preferred under organic farming. In the Netherlands spring barley was considered a poor companion for pea because both crops are considered to have poor lodging tolerance, whereas in Switzerland the

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experience was that spring barley can improve the standing ability of pea. Switzerland, mixed cropping of pea with cereals highlighted the importance to developing infrastructure to separate harvest (if not used as fodder). Selection for seed size may make the separation easier. For the production for large-scale value chains, infrastructural issues must be solved (acceptance and sorting facilities for mixed product, product purity standards, etc.).

Lupin

In Poland, mixtures of yellow and blue lupines with cereals could be recommended for organic farmers, especially on poor, sandy soils, but specific measures should be developed and implemented to encourage stockless organic farmers to cultivate them. Mixtures of lupin with oat performed better (higher yield and yield stability) than mixtures with spring triticale. Yellow lupin in a mixture performed better than blue lupin. In Switzerland, experiences with mixtures of white and blue lupin and cereals highlighted that there is a difference between the two species in their **weed competitiveness** (e.g., mixed cropping of blue lupin- cereal gave better results than mixed cropping white lupin – cereal) because white lupin is more competitive and can easily overgrow the companion crop due to its vigour and larger leaf area, particularly under drought conditions. However, in the Netherlands late maturing cereal varieties were found to be a good partner for white lupin. In Switzerland, **weed pressure** of white lupin might be solved more efficiently with improving the crop order and diversity in the crop rotation than with diversity of mixed cropping, while for blue lupin mixed cropping with spring triticale is recommended for organic farmers.

Alfalfa-grass mixtures

From the experiences on growing forage legumes and grasses in mixture, many open questions regarding an ideotype of alfalfa as forage legume for use in mixture remain. One recommendation would be that when working within a set of elite germplasm with reduced variation, selection decisions might again depend on the cultivation system (different in mixture to non-mixture) and direct selection in the mixture would be more straightforward. Using a nursery system with undersown lawn type grasses might be a good compromise: it is predictive for the system with forage type grasses but offers easier phenotyping compared to the forage type grasses and easier trial handling (reduced weeding efforts) in comparison to the bare soil system.

Agroforestry

Each of the agro-forestry systems (described in Chapter 4) provides important opportunities for evolutionary and/or direct selection harnessing complex interactions that become even more complex when intercropping is included. Paradoxically, it may be harder to add value to the selection achievements for some of the systems, e.g., the alleys and the intercropping themselves. In these systems, different targets could be explored, such as the best synchronization in the intercropping, the plant traits best adapted to intercropping both for alleys and interrow traits, and the best performance in specific positions, etc.

5.2. Better understanding of the importance of breeding for plant – plant interaction to promote cultivation of highly resilient crops with inter- and intra-species genetic diversity on field specifically fitted for organic farming

As we mentioned in the Introduction, LIVESEED's approach was to evaluate how resilience can be improved at various levels. We looked at crop level through the creation of more diverse cultivars, cultivar mixtures and heterogeneous materials, at the field level through various types of mixtures of annual and perennial crops, and at system level by increasing crop numbers and the complexity of its relationships, such as done in agroforestry systems. Overall, the results suggest that increasing diversity is particularly useful to improve resilience in stress prone conditions.

5.2.1. Improving resilience at the crop level (cultivar mixtures, DOPs, CCPs)

In the five experiments that took place in France, priority was given for farmers to select or co-design a broad range of mixtures and populations which were adapted to their local needs, contributing to increasing resilience at the farm level and at the territory level. Based on a high number of accessions, the dynamic populations created by mixture (diversity-oriented populations, DOPs) were very diversified to answer to the environmental constraints, whilst they were homogeneous for a few common traits. In the case of winter wheat CCPs, genetic diversity enabled resilience for those populations. Both genetically diverse populations (DOPs and CCPs) are included in the definition of organic heterogeneous material (OHM) in the new Organic Regulation (EU 2018/848 and the respective delegated act which can be commercialized after notification from 2022 onwards. Thus, new sources of diversified organic seeds (OHM) will be available for the farmers, which will contribute to resilience at the seed system level.

In the case of on-farm breeding for diverse wheat mixtures, the investigated strategies yielded different results, and although responses were positive, not with the same significance. While the selection within the mixture tended to be more efficient, selection within the components tended to conserve more diversity. Therefore, selection strategies for mixtures should be designed depending on the objectives and priorities of farmers. Over a four-year trial of co-designing and assessing wheat mixtures, the mixtures never performed lower than the worst pure stand yields, showing the ability of mixtures to **limit the risks of yield losses** and **stabilise the protein content** compared to the pure stands whilst the designed diverse mixtures were adapted to the farmers' local environment.

In the barley trials in Latvia, the diversity improved **yield stability**, proving that diversity incorporated in CCPs can improve yield stability over environments. Yield advantage was found especially under stress environments: some (3 out of 8) mixtures were stable yielding over the environments (but not all of them). Barley mixtures also showed a positive effect with larger probability in respect to the reduction of the presence of leaf **diseases**.

In the maize trials, resilience was improved by incorporating diversity and heterogeneity (multiplying the germplasm in a marginal low input organic land for two years, then via the creation of CCPs from 40 populations collected in Azores – BulkAzores1 and BulkAzores2). This diversity **improved yield stability over environments**.

5.2.2. Improving resilience at the field level (crop mixtures)

In general, improving resilience through crop mixtures proves more difficult compared to increasing intraspecific diversity. In the first place, in the case of crop mixtures it is important to attune the crop phenology, ripening time and plant density of the crops. Tillering ability, directly related to plant density, is important for improved resilience at monocrop level, but can hamper crop growth of the companion crop under specific conditions.

In the trials mixing leguminous crops with cereals, contrasting results were observed. In Latvia, mixing spring wheat with spring pea allowed for a better coping with **weed pressure**, and in addition improved the **lodging resistance** under unfavourable conditions. Similar results were observed in the Netherlands, where better resilience to weed pressure was found statistically significant in the wheat–lupin mixed crop and in the wheat–pea mixture. Improving the lodging resistance was also confirmed in the mixed crop treatments, where the plants can grab onto the cereals to prevent lodging,

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concluding that intercropping seems to be a good solution for lodging of pea plants. This is particular the case for low-input conditions. Under more favourable organic conditions in Germany, early results suggest that to study the effect of winter pea-triticale-mixtures on resilience, more winter-pea-varieties needs to be tested for their suitability for mixtures and emphasis must be placed on careful selection of the right variety to reach the desirable effect.

In the Netherlands, the effect of the mixtures on resilience by offering a better **resistance against** several above and or below ground **diseases** could not be confirmed due to very low disease pressure. In the spring wheat-lupin mixed crop, some brown rust in the lower parts of wheat plants in some plots were observed, which was not present in the pure stand crop. This could be due to the higher relative humidity inside the mixed crop. This finding is contrary to the hypothesis and in line with earlier on-farm observations of mixed stands.

In Poland, the results revealed higher resilience of the lupin-cereal mixtures in comparison to the crops sown in pure stands concerning **yield stability** over years. This was in line with what was expected in the hypothesis and what is found in the literature, but it was particularly evident in the experiment as yields of grain legumes in pure stands were very variable and unpredictable during the trial's 3 years.

In the perennial alfalfa-grass mixtures trials and maize-beans intercropping trials no clear improvement of resilience of the system were observed.

5.3. New breeding methods, experimental designs, and selection tools for various mixed cropping systems

Apart from the necessary development of new breeding, experimental designs and selection tools, the transfer of knowledge is also very important. Figure 37 below visualizes the different categories in which results can be transferred to other situations. These will be briefly explained below.

Figure 37. Information transfer categories from our trials' results

5.3.1 New breeding methods and experimental designs

Intraspecific diversity: cultivar mixtures, DOPs and CCPs

To increase intraspecific diversity, various approaches can be used. Cultivar mixtures can be used when a sufficient number of suitable cultivars are available. An advantage of cultivar mixtures is that they can be spread through existing market channels. In order to develop more tailor-made variety mixtures, a participatory ideotyping approach was developed in France. Co-design assembly rules tailored to the farmers' local contexts were developed based on interviews and workshops with these organic farmers.

The advantages of CCPs, improved yield stability and resilience, have been clearly demonstrated. These advantages are particularly useful under suboptimal conditions. Although a relatively cheap breeding method, the development of CCPs is based on making multiple crosses, which requires expert knowledge. A new method to enhance diversity in cereal populations is the so-called dynamic population or Diversified Oriented Population (DOP). The strategy is to request many accessions of a species from gene banks, which are then multiplied and observed (for main phenological and morphological criteria) by the team for 2 to 4 years. Then the accessions were mixed according to the farmer's choices based on various criteria. The DOP can be used very easily for on-farm breeding and adaptation to local conditions.

Interspecific diversity: annual crop mixtures, perennial crop mixtures, crop adaptation in agroforestry systems

The results in this deliverable show that the breeding for crop mixtures needs to be context specific (both at the agro-climatic and value-chain level). It is very important to have clear goals set for the plant type and the companion crop(s), which depend on the local context.

it is clear that for each crop mixture, plant type can determine whether breeding for mixing ability is necessary or not. A first step is to determine whether simultaneous ripening is important or not. This is the case for annual crop mixtures for food, but less so for crop mixtures for feed or for crop adaptation in agroforestry systems. A second step is to see whether the locally available cultivars are suitable for crop mixtures, and if not, if cultivars in neighbouring countries may be more suitable (such as later maturing spring wheat which can ripen almost simultaneously with spring lupin).

If it is decided that some breeding is deemed necessary, the question is whether regular breeding approaches can be applied. For example, ranking of semi-leafless pea genotypes in a mixture with wheat and in the pure stand was similar. However, this was not the case for normal pea genotypes. In such case, breeding for mixed cropping requires to evaluate both species in mixed stand, as the ranking in performance of pure stand crops can differ from the ranking in mixed stand. When designing mixed stand cereal-legume evaluations, one or a few testers of the cereal crop (cultivars) are sufficient (only GMA effects, no SMA).

The type of companion crop also determines the crop phenotype to breed. For example, to breed for improved mixing ability of pea, a tall normal-leafed winter pea will need to be chosen when combining pea with winter triticale. Another aspect is to decide which is the main crop, in other words, of which crop the yield is the most important. In the case of breeding for improved mixing ability of triticale with pea, a large range of traits need to be considered, such as leaf width, time to maturity, juvenile growth and vigour, plant length and tillering ability.

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5.3.2 Participatory Plant Breeding methodologies

Several different participatory methodologies were developed or tested in LIVESEED, including:

- Participatory development of CCPs: breeders developing one base population and then farmers multiplying (and thus adapting) it in diverse environments (which could be interesting to reduce breeding costs);
- Involving farmers in the choice of the starting genetic material for crosses to be made by researchers or breeders, followed by selection and multiplication of novel CCPs together with farmers. This approach is easy to set up but facilitation to enable farmers to train and exchange is necessary.
- When comparing DOP and CCPs, mixtures were easier to handle by farmers than CCPs which might encourage farmers to create their own populations.
- Depending on the objectives and priorities of farmers, farmers can either select within cultivar mixtures to make greater selection advance or select within the cultivar components to maintain more diversity.
- Breeding for mixed cropping may require evaluating both species in mixed stand, as the ranking in performance of pure stand crops can differ from the ranking in mixed stand. When designing mixed stand cereal - legume evaluations, one or a few testers (cultivars) may be sufficient, which could be handled by farmers to allow on-farm evaluation.

5.3.3 The SHiNeMaS (Seeds History and Network Management System)[6]

In France, the history of the seed lots (creation of CCP, breeding and multiplication) used in the experiments were recorded in a database which keeps track of the information and relates the results to the breeding processes (and therefore facilitates transfer to other situations). The database is called SHiNeMaS (Seeds History and Network Management System), and it has a web interface, dedicated to the management of the history of the seed lots and related data (like phenotype, environment, and cultural practices). Its development started in 2005, as a collaboration between INRAE and the French farmers' seed network Réseau Semences Paysannes (RSP) on bread wheat. In this project, researchers and farmers' organizations needed to map the history and life cycle of the populations using the network forms. All this information was centralized and stored in a database. Since then, the platform was further developed and is now freely available and distributed under GNU Affero GPL License.

5.3.4 Statistical tools

In a series of workshops (Figure 1, page 12: 18th September 2020, 10th November 2020, 8th December, and 21st January 2021), partners shared their practices for data analyses. The discussions showed that the statistical methods most used for mixtures were mainly descriptive statistics, frequencies, coefficient of variation, ANOVA, and multiple tests (LSD, Tuckey), regression and Pearson correlation (Figure 2, page 14). For crop mixtures, the LER (Land Equivalent Ratio) was also a commonly used evaluation technique. Other statistical methods exist, but their application is not used because they may not fit the collected data or trial set up, or there is lack of background knowledge for the breeder to use these methods.

5.3.5 Mix targets

For mixtures it was relevant to develop synchronization of the crop cycles to make harvesting easier. As mentioned in Chapter 5.2.2, the effect of pea-triticale-mixtures on resilience needs further testing of winter-pea-varieties for their suitability for mixtures (e.g., Trial 11 winter triticale x winter pea,

[6] SHiNeMaS: https://sourcesup.renater.fr/projects/shinemas

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CULTIVARI). To better cope with weed pressure as it was the mixing spring wheat with spring pea in Latvia and for wheat–pea mixture and wheat–lupin mixed crop) that also improve the lodging resistance, generally associated to the capability of one plant to support the other, under unfavourable conditions (Trial 9, Trial 12, Trial 19). Agronomic aspects such as plant density, seed ratio of the mixed crops, time of sowing, sowing depth, sowing method, soil type, and soil fertility can be important factors that influence plant growth and hence the selection process and breeding progress. Climatic conditions, weed pressure and presence of disease may also influence decisions made regarding the agronomic practices and hence the breeding process. Exchange on all these aspects among breeders may be very useful to improve breeding progress.

5.3.6 Training

Some data treatments for mixtures are not used because they do not fit the data or there is a lack of ability for the breeder to use it, i.e., there are some tools that are particularly used by some researchers but not of interest for all the partners. To fill this gap LIVESEED organized meetings already described in Figure 1 that gave the opportunity for collaborations. The presentations of these meetings were shared with the partners, along with relevant papers and documents. Following those meetings, IPC, with the collaboration of FEUP (University of Oporto), started to apply the methodology of Random Forest, CART and MARS to AREI data from Trial 6 (spring barley mixtures and populations). The Random Forest was particularly selected to respond to the question: what are the variables that better explain a target variable, that for our case was yield (Mendes-Moreira et al. 2014). In addition, data were analysed in subgroups: organic versus conventional crop management system and homogeneous versus heterogeneous material.

Overall analysis considering all types of investigated barley material and in the two crop management systems showed that spring barley traits related to grain yield for the greatest extent are **crop ground cover** (GR.COV_st.el_Perc) and **canopy height** measured at stem elongation stage (CAN_st.el.cm) and **weed suppression ability** measured at flowering (WSA_fl_Perc) and to lesser extent in ripening (WSA_ri_Perc) and **stem elongation stages** (WSA_st.el_Perc). Traits with medium importance were **early vigour scored at tillering** (E.VIG_til) and **tillering capacity** (TILL_coef) (Figure 38).

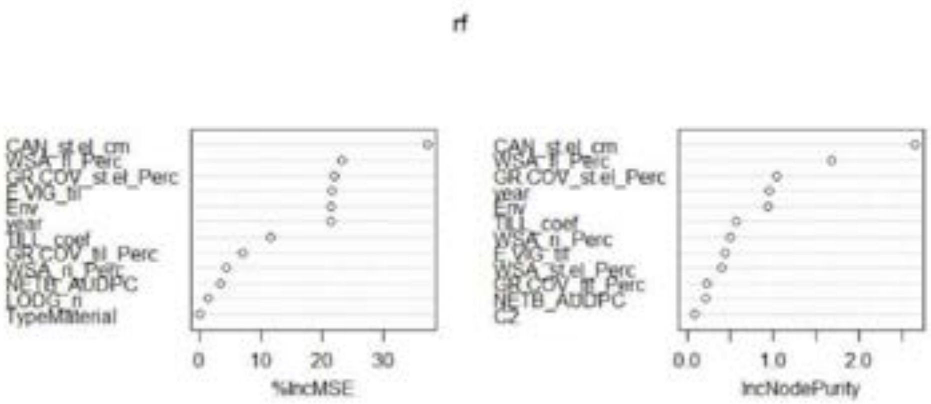

Figure 38. The Random Forest (rf) overall analysis (49 accessions, 14 environments), indicate the explainable variables for the target variable yield in spring barley were crop ground cover (GR.COV_st.el_Perc), canopy height measured at stem elongation stage (CAN_st.el.cm), weed suppression ability measured at flowering (WSA_fl_Perc).

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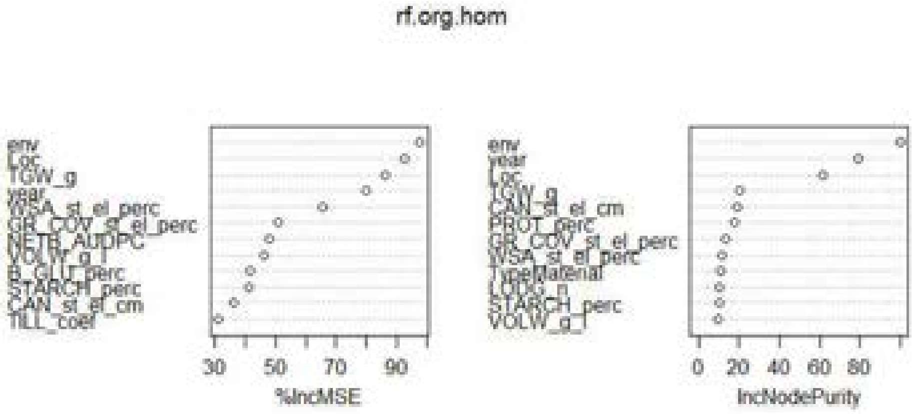

Figure 39. The Random Forest (rf.org.hom - random forest in organic for homogeneous germplasm) analysis, indicate what are the explainable variables for the target variable yield in spring barley for organic crop management system and homogeneous germplasm.

When data for homogenous and heterogenous materials were processed separately, some differences can be found, e.g., weed suppression ability (WSA) appeared to be more related to yield at earlier plant development stage (stem elongation) in case of homogenous material (Figure 39 above) and in later stages (flowering and ripening) for heterogenous material (Figure 40 below), which might be able to better compete weeds in the second part of the crop growth.

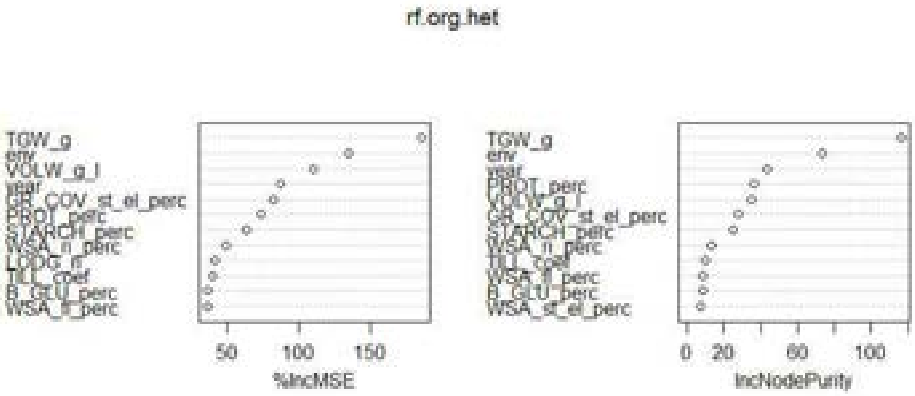

Figure 40. The Random Forest analysis (rf.org.het - random forest in organic for heterogeneous germplasm), indicate what are the explainable variables for the target variable yield in spring barley for organic crop management system and heterogeneous germplasm.

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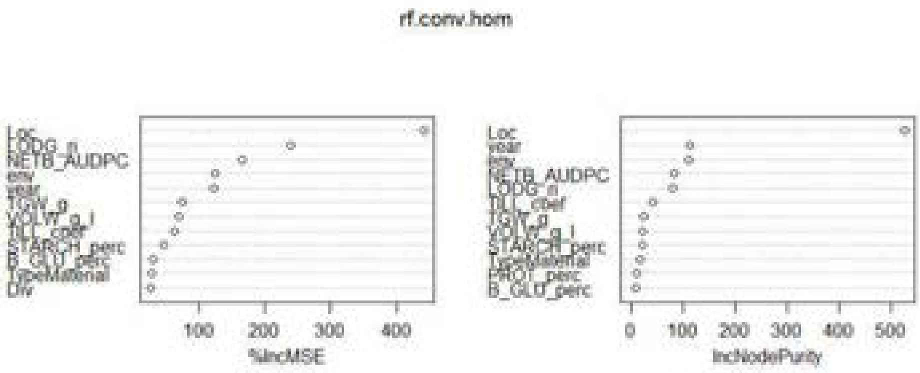

Figure 41. The Random Forest (rf.conv.hom - random forest in conventional for homogeneous germplasm) analysis, indicate what are the explainable variables for the target variable yield in spring barley for conventional crop management system and homogeneous germplasm.

Lodging (LODG_ri) logically was more related to yield under conventional crop management system (Figures 41 above and 42 below), although, for heterogenous material the importance of lodging was notably less than for homogenous material indicating some buffering capacity and possible advantages also for higher input crop management. A similar trend can be seen also for leaf disease net blotch infection severity (NETB_AUDPC).

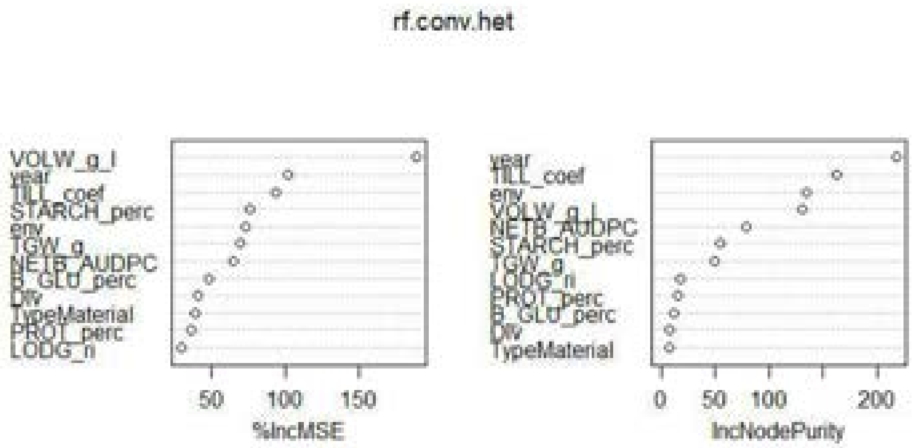

Figure 42. The Random Forest analysis (rf.conv.het - random forest in conventional for heterogeneous germplasm), indicate what are the explainable variables for the target variable yield in spring barley for conventional crop management system and heterogeneous germplasm.

5.3.7 Sources of resistance

For organic farmers, it is very important to maintain a favourable balance between general resilience and yield stability rather than maximum yield potential. To achieve these goals, the resistance to biotic and abiotic stresses can be improved by mixing components with distinctive disease resistances. A general conclusion may be that for developing populations with improved resilience, the components need to be contrasting genetically but also phenotypically, in particular to traits related to disease resistance.

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5.4. Novel approach to co-design with farmers: blends of cultivars or species mixtures

In the LIVESEED project, several co-design approaches (CCPs, DOPs, participatory ideotyping involving farmers in the choice of the starting genetic material, culinary breeding, etc.) were applied, responding to different local pedo-climatic conditions, markets, seed systems, and socio-economic contexts (see Chapter 3 and Chapter 5.2). These differences may be influencing the acceptance of co-designed materials, and the co-creation process, e.g., CCPs may be more accepted in one country than in another. CCPs and DOPs were reported as materials that can be used easily for on-farm breeding, increasing the adaptability to a diversity of pedo-climatic conditions, and reducing the costs of breeding.

Creating and selecting mixtures of populations was a flexible way for farmers to increase diversity on-farm and obtain populations adapted to their objectives and practices. Farmers also often expressed a need for diversification. In France, the results of participatory on-farm breeding (wheat mixtures) showed that the selection within the mixture tended to be more efficient, but selection within the cultivar components tended to conserve more diversity, and therefore strategies of selection for mixtures should be designed depending on the objectives and priorities of farmers. For productivity traits, the range of response to selection depends mostly on farmers' objectives and selection practices, and little on the number and diversity of components, and consideration of farmers' affinity with the selection method is essential.

In the ideotyping trial, the co-designed assembly rules implemented on a multi-criteria assessment tool helped farmers when designing mixtures. This work emphasises the interest to joining up knowledge between actors form different disciplines and perspectives. Farmers were also highly motivated to co-create populations, especially if they were involved in the selection of the CCP parents, in the observations and in the breeding. However, they need facilitators to coordinate and maintain the link with scientists. From a single base CCP various diversified populations can be obtained (different populations created from the different crosses and some inbred lines derived from the crosses), which engages farmers. When comparing CCPs with DOPs, farmers wishing to increase intra-specific diversity by on-farm selection preferred mixtures as they are easy to make and allow the development of diversified populations. CCPs were more interesting for more precise selection. Farmers were also interested in DOPs because they can choose more relevant criteria answering to their conditions and market. However, seed quantities especially of genebank accessions might be too low in some cases which may deter farmers. Also, for facilitators the following up of the work by farmers and the gathering of the responses of every person who received some DOPs takes significant amounts of time and effort. However, it was also clear that good facilitation is important to maintain a collective dynamic among farmers, particularly at the beginning of the process.

In the maize trial in Portugal, it proved also important to keep farmers motivated and interested in germplasm evaluations to adapt their preferred populations to their systems. Culinary breeding networks could promote the use of traditional maize populations and underutilized maize landraces, as they have great grain quality traits that allow a wider range of products to be developed.
In Portugal, the approaches involved farmers in different stages of the breeding process from providing the field, to defining the breeding goals, selecting the genotypes/starting genetic material, taking part in the selection or co-design assembly rules of the farmers' variety mixtures to tailor them to their local context, to multiplication and/or the evaluation process together with the researchers.

From the experiences it can be concluded that researchers/facilitators will be playing a crucial role in the near future in providing capacity building for farmers, to allow them to be able to work with

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such materials on their own. For the time being, farmers require the link with scientists to access materials, for methodology, evaluation and statistical analyses and coordination.

Farm advisory services could build in such programs for on-farm breeding across Europe to facilitate uptake. According to the survey results, farmers are willing to sacrifice land from production and their time to develop improved cultivars suited for their local conditions. The level of motivation by farmers to develop their own cultivars and populations in France was increased by long working relationships with breeders. This also indicates that systematic and long-term collaborations would be advisable to horizontally upscale such on-farm breeding initiatives, which would motivate farmers to develop their own diversified materials and take part in other activities in the seed systems. Furthermore, systematic collaborations with farmers should expand with public breeders, millers, bakers, and other actors in the value chain.

Several CCPs or DOPs of the diverse crops have been developed in the scope of LIVESEED. Due to the new Organic Regulation (EU, 2018/848) about the simple notification of Organic Heterogeneous Material (OHM), these new sources of diversified organic seeds can be made commercially available to a larger number of organic farmers, which will contribute to resilience at the seed system level.

5.5. Proof of concept for breeding for more diverse cropping systems

Based on the results described in this deliverable we can describe a proof of concept for breeding for more diverse cropping systems as follows.

- The results show that for breeding for more diversity considering the local context is very important (more than for homogeneous crops), in particular for mixed cropping and agroforestry systems. Good collaboration with farmers and other value chain actors is important for the new mixture to be well embedded in the food system. Agricultural experts can help understand how agro-climatic conditions can influence the crop growth of crop mixtures.
- For developing cultivar mixtures, Diversity Oriented Populations (DOP) and Composite Cross Populations (CCP), a participatory ideotyping approach is advised to understand which cultivars to combine. Farmer and expert knowledge are integrated through workshops.
- For breeding for mixed cropping of different species, several breeding tools can be helpful. Depending on the set goal and the local context, cultivars bred for monocropping can be used (either from the local region or from outside the region), or specific breeding for mixed cropping needs to be set up.
- Multi-actor approaches are advised to involve farmers and other actors of the local or regional food system. The potential advantages are cleared defined breeding goals (fit for purpose), better adoption of the developed material and a better distribution of the work reducing the financial costs.
- The potential advantages of breeding under agroforestry are not yet fully understood and need to be investigated further.
- A more flexible normative and legal framework is important for upscaling such results and for commercialization of the developed improved cultivars for organic production. This is highlighted in the new Organic Regulation (EU 2018/848) with respect to Organic Heterogeneous Material (OHM) and Organic Varieties suited for organic agriculture.

Concluding remarks and next steps

A general conclusion is that a breeding strategy with a focus on increasing diversity can contribute to yield stability and resilience. In most LIVESEED trials the results were overall positive or neutral (Tables

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9-21) in terms of the importance of the characteristics contributing to breeding for resilience. The results also showed to consider pedo-climatic conditions and socio-economic context. Several populations and mixtures were developed through the project, the uptake of which can be recommended or promoted for use and further development in low-input and organic farming.

The project implemented a set of commonly agreed overall hypotheses as well as individual hypotheses for each trial defined by the project partners. The results show that although some of the common hypotheses could be validated by several partners, it was not possible to have generic hypotheses that could be confirmed by all partners, and that it is important to have context-specific hypotheses.

Multi-actor approaches were used to develop locally adapted populations with more stable performance. These participatory approaches proved to be very important for smaller crops, and for the engagement of farmers with the intention to develop their skills to do on-farm breeding in the future. However, horizontal upscaling across Europe requires more systematic exchange and training by advisors and knowledge institutions, and more systematic and long-term funding.

The project also developed a range of breeding methods and statistical methods that are fit for use in different geographical regions and for different crops by breeders in the future, potentially in collaboration with farmers.

Agroforestry systems may not only provide opportunities to improving resilience but also allow a very different perspective on current farming, breeding, and research practices. However, these systems are complex, and more research is needed to elucidate several aspects: the plant-plant interactions, the competition for resources, and the soil-plant interactions were much more complex than what we originally thought (see INRAE's trial #25 on tomato in agroforestry system, and IPC's maize trial in SAF system, Trial #26, Annex II). These questions to our original research questions, namely whether (i) agroforestry systems are an environment or as a mosaic of micro-environments, (ii) which breeding stage is suited best for which agroforestry, and (iii) if agroforestry systems can offer new solutions for more effective breeding and variety testing for low-input systems could not be fully answered in the timeframe of the project. For now, what we can conclude is that (i) the yield potential of a wheat population can be influenced by the position in an alley (constructed by two North-South oriented tree rows) where it has been multiplied, and (ii) presence of diseases (including seed-borne diseases) in a wheat population can be influenced accordingly due to differences in micro-climate (e.g., humidity and solar radiation). However, new research questions regarding improving resilience have arisen during the research project, e.g., one important question is how to better understand and benefit from plant-soil interactions. The agroforestry trials of LIVESEED ultimately contributed with important input to the study of the microbiome (summarized in LIVESEED document D3.7, Report on importance of holobiont as potential selection target).

Another topic that was identified as an important research topic for the future is the issue of social resilience, if we are to implement the systems-based breeding framework developed by LIVESEED (Lammerts van Bueren et al., 2018). Aspects for future research could look at (i) Genotype x Environment x Social (GxEXS) interactions and how maintaining and enriching cultural diversity helps broaden the perspectives on breeding, (ii) how social resilience is formed in different breeding orientations, and (iii) how a more evolved social resilience influences the efficiency of the breeding process (e.g., definition of breeding goals or selection) in different seed systems. As was described by Lammerts van Bueren et al. (2018), and further elaborated in Deliverable 3.5 by Nuijten et al. (2020), ecological and social resilience are interrelated.

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ANNEX I. Detailed reports of the field trials

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LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

INRAE – Participatory on-farm breeding for diverse wheat mixtures (Trial #1)

Lead: INRAE GQE-Le Moulon
Isabelle Goldringer – (isabelle.goldringer@inra.fr) & Gaelle van Frank (gaelle.van-frank@inrae.fr)

Abstract
INRAE and RSP (Réseau Semences Paysannes) have investigated the best approaches to select for mixtures on farm from 2015-2016 to 2018-2019. The objective was to assess different selection procedures for the on-farm breeding and management of mixtures. In the previous H2020 project DIVERSIFOOD, a protocol was co-designed with the farmers for assessing four different selection strategies for bread wheat mixtures. Eleven to fifteen farmers participated to the experiment. Morphological and grain production traits have been measured on individual plants, as well as yield and protein content at the plot level over the four years. Data have been analysed using Bayesian models for decentralized on farm trials already developed. Significant positive effects of mixtures and farmers' selection were detected. Two methods can be interesting depending on the objectives of the farmers. Part of the analyses and dissemination have been conducted within the LIVESEED project.

Introduction

In France, a Participatory Plant Breeding (PPB) project has been conducted on bread wheat since 2006, involving farmers and facilitators of the farmers' seed network Réseau Semences Paysannes (RSP) and INRAE researchers (Diversity, Evolution and Adaptation of Populations team). The project aims at developing farmers' varieties that are adapted to farmer's objectives and needs based on decentralized selection in farmers' fields. Methods and tools for on-farm breeding were developed to help farmers' reappropriation of on-farm breeding knowledge and seed autonomy. The farmers who are partners in the participatory selection project have been selecting wheat populations adapted to their practices and needs for nearly 20 years. However, a common practice in production is to mix these populations, so many farmers create a mixture that will be re-sown over the years, giving it the possibility to adapt to the soil and climate context and to the farmer's practices. These mixtures can be composed of commercial varieties, but the components can also be populations that have been tested separately by the farmers, selected, and then mixed with other interesting populations on their farms. Farmers' interest in improving their practices of creating and selecting suitable mixtures was identified through discussions during farm visits or meetings. To understand farmers' practices and questions regarding the creation, management and selection of on-farm mixtures, a series of semi-structured interviews was conducted in 2015. The aim of these interviews was to identify questions that could be answered by an experiment.

Hypothesis and research question

The survey revealed a need for information on the impact of selection practices on the behaviour of mixtures, and it is this question that we wanted to answer with this experiment. Several hypotheses based on the literature or on previous results were formulated:
- It has been shown that mass selection can have a positive and significant impact on several traits of interest (e.g., thousand kernel weight, spike weight and spike length), so it is expected that mixing seeds from selections made in the components will perform better than mixing populations without prior mass selection;

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- Knowing that the behaviour of a plant in a mixture cannot be predicted from its behaviour in pure stand, it could be more interesting to select directly in the mixture the plants that behave well in interaction. Indeed, by selecting in the components before mixing, there would be a risk of eliminating plants that might perform better than the others in the mixture.
- Given the results showing positive impacts of mass selection in populations, one would expect to observe an added value to mass selection practices on mixtures compared to the mixture evolving without this selection.

Based on these hypotheses, an on-farm experiment conducted from 2015-2016 to 2018-2019 was designed to investigate the best approaches to select for mixtures on farm. The objective was to assess different selection procedures for the on-farm management of mixtures.

Material and Methods

Location(s)

14 farms in France and one farm in Belgium the first year, 10 farms in France and one in Belgium the following years and six farms the last year:
2015-2016: DAV, CHD, FRC, JUBA, MAV, CHH, HEA, LOD, ROG, ADP, OLR, RAB, JSG, ANB, RDR;
2016-2017 and 2017-2018: CHD, CHH, FRC, HEA, JUBA, MAV, ROG, RAB, JSG, RDR, SOLE;
2018-2019: CHD, CHH, HEA, RAB, JSG, RDR.

List of accessions

Each farmer assembled his own mixture(s), from 1 to 3 mixtures. As a whole 28 mixtures with 2 to 15 components were designed and evaluated (Fig.1).

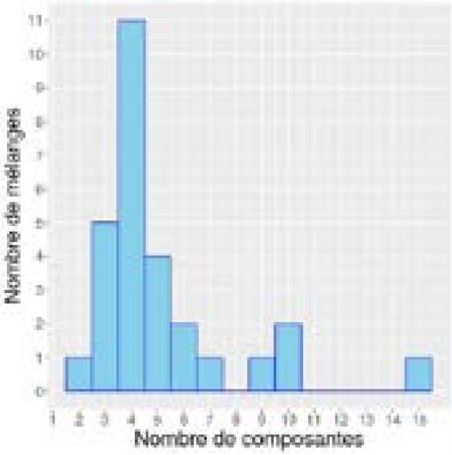

Figure 1: Distribution of the number of components per mixture created in 2015.

Experimental design

The protocol was co-designed in 2015 with farmers and facilitators of the wheat RSP group for assessing the different selection strategies of interest. Four different selection strategies (no selection, selection within the components, selection within the mixture and a combination of them) were assessed in on farm experiments (Fig. 2). Fourteen farmers sowed a trial in autumn 2015. In 2016-2017 and 2017-2018, 11 farmers continued or joined the experiment. In 2018-2019, six farmers evaluated the different mixtures again, without additional selection, but in general in larger plots.

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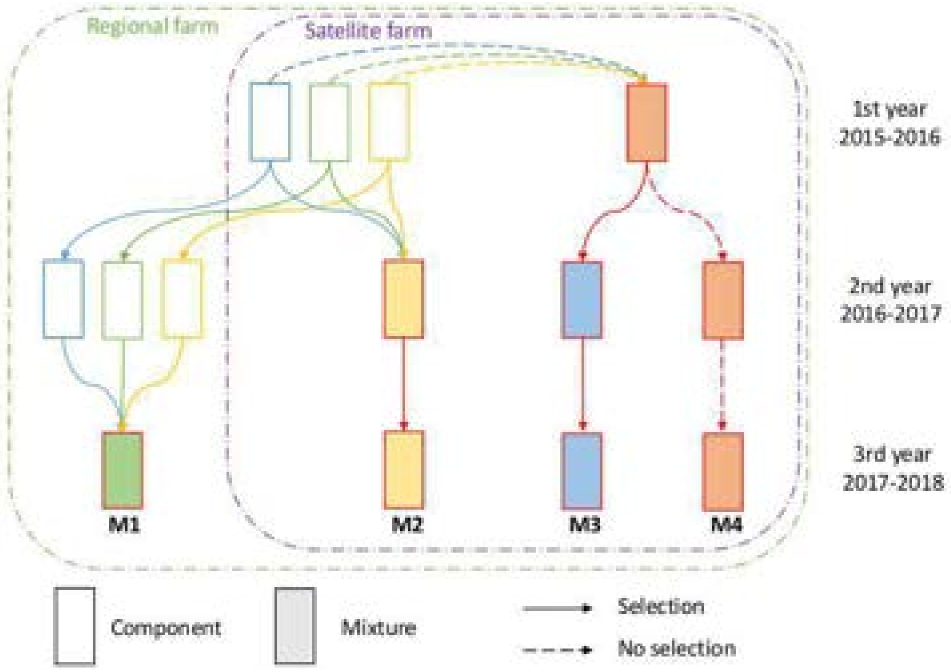

Figure 2 On-farm experimental design over 3 years. The mixtures and their components were sown the first year, then 3 selection practices led to mixtures that have been compared to the non-selected mixture (M4): (i) two years of selection within the components before mixing (M1), (ii) one year of selection within the components, mix those selection then select within this new mixture and (iii) two years of selection within the mixture.

Evaluation

Morphological and grain production traits have been measured on individual plants, as well as yield and protein content at the plot level.
The list of the traits collected in the trials are shown in Table 1-3.

Table 1. List of the traits collected for bread wheat concerning Crop development and agro-ecological performance

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Plant height	Distance from the soil to the top of the spike. Trait measured on around 30 plants / plot.	Individual quantitative data
Distance between last leaf and spike basis	Distance measured on around 30 plants / plot.	Individual quantitative data
Spike length	Distance measured on around 30 plants / plot.	Individual quantitative data
Spike colour, awness, curve	Scoring on around 30 spikes / plot based on reference scales.	Individual qualitative data

Table 2. List of the traits collected for bread wheat concerning crop productive performance

Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**

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Spike weight	Weight measured on around 30 spikes / plot.	Individual quantitative data
Thousand kernel weight	Averaged weight of thousand kernels based on counting the number of kernels of a 20g sample.	Plot quantitative data
Yield	Grain weight harvested at the whole plot level	Plot quantitative data

Table 3. List of the traits collected for bread wheat concerning crop quality performance.

Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		
Trait	**How it has been assessed**	**Type of data available**
Protein content	Assessed on plot samples of kernels after harvest using NIRS	Plot quantitative data

A survey of the farmers' practices with regards to mixtures management has been done in 2016, and in 2018 on their feeling about the behaviour of mixtures and the participatory dimension of the project.

Statistical methods

Mean genetic effects and variances of populations within farms were estimated using Bayesian models developed in DIVERSIFOOD (http://www.diversifood.eu/wp-content/uploads/2018/12/booklet3_decentralized_on_farm_breeding_BAT_web_A4_2.pdf).
Mixtures' overyielding for each trait was computed as the genetic effect of the mixture divided by the weighted mean genetic effect of their components. The overyielding was then compared to zero using a student test in case of a normal distribution or a Wilcoxon non-parametric test otherwise.
Selection differential was calculated as the genetic effect of the selected spikes minus the genetic effect of spikes taken randomly in the population, while response to selection was computed as the genetic effect of the selected population minus the genetic effect of the non-selected population.
The responses to the selection practices were calculated as the genetic effect of the selected mixtures (M1, M2 or M3) minus the genetic effect of the non-selected mixture (M4).
The effect of location or mixture, selection modality and year on response to selection (mean genetic effect and variance of the trait) were tested using an ANOVA.

Results

The impact of growing a mixture compared to pure stand differed between traits, with a significant positive impact of growing mixtures on spike length (+3.4% *), mean number of grains per spike (+4.8% *) and height (+4.8% **). Only for thousand kernel weight did the mixtures behaved overall worse than the mean of their respective components (-2.1% *). The samples of spikes selected by the farmers in components and in mixtures were heavier with more kernels than the mean of the population. Selection response was in general lower than selection differential, although the response to selection practice over all mixtures was significant for spike weight (+13.6% *) and number of grains per spike (+15% *) when selecting within the mixture (M3) (Fig. 3). There were significant effects of farm or mixture on the gains obtained from selection, meaning that selection response depended on farmers' selection

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practices (selection intensity, willingness to conserve diversity within mixtures, …) more than on the selection modality.
Surprisingly, selection response for protein content was always positive although seldom significant meaning there was no dilution effect of spike weight increase in the selected populations.
There was no significant decrease in phenotypic diversity in mixtures associated with the different selection practices for the traits measured at the individual level (Fig. 4). However, there was a tendency to conserve more diversity when selecting within components before mixing for the traits that responded most to selection compared to selecting within mixtures.

Trait	M1 vs M4	M2 vs M4	M3 vs M4
Thousand kernel weight	-3.4 % (n=11)	-1.7% (n=16)	+1.5% (n=16)
Spike weight	+1.7% (n=12)	+13.6% (n=17)	**+13.6% * (n=17)**
Spike length	-2.7% (n=9)	+2.2% (n=13)	+2.3% (n=13)
Plant height	-1.3% (n=9)	-0.5% (n=13)	+1.4% (n=13)
Last leaf to spike distance	+0.02% (n=9)	+1.9% (n=13)	+7.9% (n=13)
Protein content	+4.6% (n=7)	+1.0% (n=9)	+0.7% (n=9)
Number of grains per spike	+5.2% (n=11)	+17.5% (n=14)	**+15.0% * (n=15)**

Figure 3 Mean response to selection (mean genetic effect) the 3rd year.

Trait	M1 vs M4	M2 vs M4	M3 vs M4
Spike weight	+21.5% (n=10)	+6.4% (n=15)	+3.9% (n=14)
Spike length	-12.6% (n=7)	-3.4% (n=11)	+5.2% (n=10)
Plant height	+14.9% (n=7)	+7.5% (n=11)	+0.9% (n=10)
Last leaf to spike distance	-7.9% (n=7)	-7.1% (n=11)	-11.1% (n=10)

Figure 4: Mean response to selection (variance of genetic effect) the 3rd year.

Participatory/multi-actor approach

The results have been discussed with the farmers involved and their feedbacks were that two years of selection and observation of response on the third year was too short to get a real idea of the differences among the mixtures obtained with different selection practices. They also needed to observe mixtures on larger plots. This led us to run a fourth year of experiment with six of the farmers who were highly committed.
They identified some difficulties in selecting within components or mixtures as expressed in their comments: “Components were visually less diverse (...) fewer things were obvious, so maybe a weaker selection”, “I found it very hard to select within mixtures (...) I tried to keep a bit of everything (…). So, I felt less free with my selection practice”.

Discussion

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Main outcomes

This experiment allowed to identify numerous benefits of mixtures:

- A positive overyielding for most productivity and quality traits except thousand kernel weight;
- Farmers take less risk by sowing a mixture compared to a single variety not knowing the year conditions.

From the comparison of selection practices, we found that:

- Mostly farmers selected heavier spikes with more kernels that were larger;
- Positive response for most traits, selection within mixtures allowed to maintain TKW;
- Larger positive responses were observed on productivity traits (spike weight, number of grains per spike) for selection within the mixtures;
- None of the selection practices led to a significant loss of phenotypic diversity;
- Selecting within components tends to better conserve the phenotypic diversity within mixtures.

Main lessons learned

Different selection strategies for the management and breeding of mixtures on-farm have been investigated. Positive although not always significant responses were observed. The selection within the mixture tended to be more efficient but selection within the components tended to conserve more diversity. So strategies of selection for mixtures should be designed depending on the objectives and priorities of farmers.

Publications

- Gaëlle van Frank et al. 2021 “Participatory on-farm breeding for diverse and adapted wheat mixtures”. Eucarpia – LIVESEED conference on Breeding and seed sector innovations for organic food systems, 8th of March 2021.

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INRAE - Participatory ideotyping and assessment of winter wheat variety mixtures (Trial #2)

Lead: INRAE GQE-Le Moulon
Isabelle Goldringer – (isabelle.goldringer@inra.fr), Emma Forst
Winter wheat – participatory ideotyping (CASABio 2018)

Abstract

Variety mixtures represent an interesting diversification lever for stabilising production, especially in organic conditions. However, farmers lack practical recommendations for designing efficient winter wheat mixtures that favour complementarity and synergies between components based on key traits for plant-plant interactions. Using an ideotyping participatory approach based on workshops with a group of organic farmers from the Ile-de-France region, we co-designed assembly rules and farmers' variety mixtures of 3 to 5 bread wheat varieties tailored to their local context. The mixtures were assessed for four years (2016 to 2019) with on-farm strip experiments, allowing for comparison with the varieties in pure stands. They showed a reduction of the risks of yield losses and a stable protein content compared with pure stands. The co-designed assembly rules are implemented on a multi-criteria assessment tool to help farmers when designing mixtures. This work emphasises the interest to joining up knowledge between actors form different disciplines and perspectives.

Introduction

Mixtures may allow to buffer abiotic stresses, stabilise productions, regulate pests and foliar diseases, improve weeds control, and optimise the use of resources, due to compensation and complementarity between varieties.
In the case of winter wheat, mixture components have been assembled primarily based on their yield and protein content, their complementarity for disease resistances and to maintain homogeneity for height and maturity. Moreover, variety mixtures represent a simple lever to finely tune the choice of the varieties to fit with local context, through the opportunity of combining interesting variety traits. However, other criteria of interest for plant interactions such as tillering and earliness should be considered to design mixtures, but very few is known about plant interactions within variety mixtures despite the increasing surfaces sown with variety mixtures in France. We aimed to optimise the design of variety mixtures using relevant criteria and integrating farmers' practices and objectives for production and assessing designed variety mixtures in on-farm trials.

Hypothesis and research question

A participatory ideotyping approach has been developed during a previous project for low input conventional farming systems (Wheatamix project). This approach has been applied to organic farming systems. The aim was to design wheat variety mixtures specifically adapted to the local farmers' production context (pedo-climatic conditions, objectives, practices) (D.3.6). Workshops were carried out with farmers, advisors, and researchers to identify assembly rules for improving complementarity and synergies between mixture components. These assembly rules were mobilised to co-design the farmers' mixtures which have been assessed on on-farm trials. These outputs could be further used for designing system-based breeding strategies for variety mixtures (Deliverable 3.1).

Material and Methods

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

Location(s)

2016

7 on-farm trials in Ile-de-France region (Paris), France. (Farm names: DAB, EMV, FRC, JOS, OLR, RIV, THL).

2017

3 on-farm trials in Ile-de-France region (Paris), France. (Farm names: DAB, OLR, RIV).

2018

4 on-farm trials in Ile-de-France region (Paris), France. (Farm names: GER, NIR, REG, RES).

2019

3 on-farm trials in Ile-de-France region (Paris), France. (Farm names: GER, NIR, RES).

Genetic resources

List of accessions

2016-2017

Commercial varieties available for organic production in France used for the 2016 and 2017 codesigned mixtures: Attlass, Capo, Element, Energo, Ghayta, Hendrix, Renan, Skerzzo, Titlis, Venezio.
Codesigned mixtures (with some adaptations the second year):
- DAB 2016 and 2017: Attlass, Element, Renan, Titlis
- EMV 2016: Attlass, Element, Renan, Titlis. 2017: Energo, Element, Renan, Titlis.
- FRC 2016: Attlass, Element, Renan, Titlis.
- JOS 2016: Attlass, Hendrix, Renan, Skerzzo. And 67% Renan + 33% Attlass.
- OLR 2016 and 2017: Capo, Element, Energo, Titlis.
- RIV 2016 and 2017: Ghayta, Renan, Venezio.
- THL 2016: 50% Renan + 25% Attlass + 25 % Skerzzo. 2017: 50% Renan + 25% Attlass + 25 % Hendrix.

Each mixture has been assessed on the corresponding farm, along with the 3-4 components in pure stand.

2018

Commercial varieties available for organic production in France used for the 2018 codesigned mixtures: Adesso, Ehogold, Energo, Fructidor, Lukullus, Renan, Rubisko, Skerzzo, Venezio.
Codesigned mixtures:
- GER: Energo, Renan, Rubisko
- NIR: Energo, Renan, Rubisko, Skerzzo
- REG: Energo, Fructidor, Renan, Venezio
- RES: Adesso, Ehogold, Energo, Lukullus

Each mixture has been assessed in the corresponding farm, along with the 3-4 components in pure stand.

2019

Commercial varieties available for organic production in France used for the 2019 co-designed mixtures: Annie, Apache, Ehogold, Energo, Lukullus, Renan, Rubisko, Tengri.
Co-designed mixtures:
- GER: 40% Energo, 40% Renan, 20% Tengri
- NIR: Annie, Energo, Renan, Rubisko
- RES: 20% Apache, 20% Ehogold, 20% Energo, 40% Lukullus

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Each mixture has been assessed on the corresponding farm, along with the 3-4 components in pure stand.

Co-design of assembly rules and wheat variety mixtures

To develop variety mixtures adapted to various environmental conditions and practices, an ideotyping participatory approach was implemented. This approach was first proposed on pure stand varieties, and it was transposed to variety mixtures as part of the Wheatamix project. Here, we have adapted this approach to the context of organic farming with eleven organic farmers from the Ile-de-France region (GAB IdF).

i) Interviews were conducted with the farmers to describe their farming systems, practices, and end-use requirements for wheat production.

ii) Workshops were carried out with farmers, technical experts, and researchers to define assembly rules describing the traits to combine in a mixture (based on morphological, phenological, physiological characteristics and disease resistances) to buffer specific stresses or optimise resource use.

iii) Then, each farmer's wheat mixture was co-designed based on the assembly rules relevant to his context, the characteristics of available organic varieties and his practices.

Experimental design

The on-farm trials have been managed by the farmers, they benefited from the help of the partner facilitator (GAB Ile-de-France) for sowing and harvesting. The assessment was mainly carried out by the INRA GQE-Le Moulon.

Trials were composed of strips of 10-12m width minimum, and ~100m long. Each variety and the mixture have been sown in one strip each, and the mixture was replicated in one additional strip. Each trial was divided in three blocks, measures were performed on each block (except for the harvest measures).

The statistical analysis was based on the comparison of the mixtures and the pure stands components. Overyielding (OY) were calculated.

Evaluation

List of the traits collected in the trial are shown in Table 1-3.

Table 1. List of the traits collected for bread wheat concerning Crop development and agro-ecological performance

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
2016-2017-2018-2019		
Trait	**How it has been assessed**	**Type of data available**
Disease development	Visual assessment using QuantiPest scoring scales for the main diseases	Scoring
Crop ground cover	Visual assessment using ITAB scoring scale	% of covered soil
Wheat biomass	Sampling of 50cm x 50cm surfaces **In 2019:** Sampling of 50cm x 50cm surfaces, drying at 80°C during 48h.	Weight (ears and vegetative parts separately), nitrogen content **In 2019:** Weight of dry matter (ears and vegetative

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		parts separately), nitrogen content
Weed biomass	Sampling at the same time as wheat **In 2019:** Sampling and drying at the same time as wheat	Weight, species identification **In 2019:** Weight of dry matter, species identification

Table 2. List of the traits collected for bread wheat concerning crop productive performance

Crop productive performance (yield, yield components)	
2016-2017-2018-2019	
Trait	**How it has been assessed**
Yield	Farmers harvesting machine, and measure of the harvested surface
Number of plants per m²	Counting on 1m on two rows, during winter
Number of spikes per m²	Ear counting for the 50cmx50cm wheat samples
TKW	Samples at harvest

Table 3. List of the traits collected for bread wheat concerning crop quality performance.

Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		
2016-2017-2018-2019		
Trait	**How it has been assessed**	**Type of data available**
Protein content	Samples at harvest	
Bread making test (in 2018 only)	Samples at harvest (4kg) “bipea” tests	Scoring on 300 (and detailed scores)

Multi-criteria assessment tool

The assembly rules designed during the workshops and validated on the data are currently implemented in a multi-criteria assessment tool (OptiMix, with an R/Shiny web interface available at http://moulon.inra.fr/optimix/), previously developed on rules to reduce disease development in mixture. This tool aims at helping farmers designing and evaluating variety mixtures tailored to their needs. A specific workshop was dedicated to users’ tests and feedbacks to further improve the tool functionalities and user interface.

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Results

1 Co-design of assembly rules and variety mixtures

The knowledge shared during the workshops has led to the identification of various strategies and assembly rules for designing mixtures for two major objectives:

- to increase weed control by limiting the weed density (early vigour) and limiting the weed development through improving wheat competitive ability (diversified heights and growth habits),
- to face nitrogen stress by tolerating an early deficiency, complementary nitrogen demand in time (heterogeneous earliness) or complementary nitrogen use efficiency (but with the same quality type).

2 On-farm assessment

The on-farm agronomical assessment of the **20 co-designed mixtures over four years showed a mean over-yielding of 3.07%**. Mixtures never performed lower than the worst pure stand variety within trials, showing their ability to limit risks of important yield losses. The worst under-yielding over the four years was obtained in the GER 2019 trial (-27.3%). Further analysis of the trial is needed to understand this counter-performance of the mixture (e.g., important differences in weed biomass between blocks were observed, lower plant density, lower TKW than expected based on the mean of the pure stand).

The mixtures stabilised the protein content (compared to the pure stands) even in the GER 2019 trial, and improved other evaluation criteria for farmers such as disease reduction and weed control. They obtained high scores on baking tests (compared to pure stands).

3 Multi-criteria assessment tool

The tool provides farmers with a detailed feedback on the assessment of their mixtures (for their local context) and the rules underlying this assessment, helping them in adjusting the choice of the varieties composing their mixtures.

Statistical methods

The statistical analyses were based on the comparison of the mixtures and the pure stands components. Overyielding (OY) were calculated.

How resilience was improved

Over the four years, mixtures never performed lower than the worst pure stand yields, showing the ability of mixtures to limit the risks of yield losses. Mixtures also stabilised the protein content (compared to the pure stands), and no dilution of the protein content by the higher yields in mixture was observed.

Moreover, farmers designed a diversity of mixtures adapted to their local needs, which also contributes to increasing resilience at the territory level.

Participatory/multi-actor approach

The participatory approach was particularly relevant for identifying and addressing farmers' and practitioners' needs for optimising mixture design in terms of practical recommendations, experimental results, and flexible multi-criteria assessment tool.

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How information was transferred to the other situations

The participatory ideotyping approach was suggested to other farmers for designing new mixtures and they could use the multi-criteria assessment tool Optimix (http://moulon.inra.fr/optimix/).

Discussion

The results of the on-farm assessment of the variety mixtures tend to confirm the interest of mixtures in organic systems due to their ability to improve several criteria (quantity and quality) together. The high results on baking tests should be validated on further experiments.

Farmers designed a diversity of mixtures adapted to their local needs. However, the organic wheat variety offer remains limited, it should be enriched by varieties specifically bred for improved mixing ability (Forst et al. 2019) and using mixture ideotyping.

This work emphasises the interest to joining up knowledge between actors form different disciplines (agronomists, geneticists, ecophysiologists) and perspectives (theoretical, applied, and practical) for co-designing ideotypes and for decentralised evaluation of the mixtures.

This ideotyping method based on shared knowledge between actors and on-farm trials might further benefit from exchanges with crop modellers for further understanding plant-plant interactions within wheat variety mixtures using simulated data.

The farmers' satisfaction level was recorded during interviews at the end of the growing seasons. Most of the farmers were satisfied with their mixture and wished to use them for wheat production on their farm. The farmers' observations generally converged with the on-field measures.

Outcomes and conclusions (regarding the efficiency and the limits of the approach (if possible))

A participatory ideotyping approach has been carried out for determining assembly rules and for codesigning wheat variety mixtures, thus improving the adequacy of mixtures to the local production context. The three-year on-farm agronomical assessment of these co-designed mixtures tends to confirm the interest of mixtures, especially in stressing conditions such as in organic systems.

Publications

- Poster at Diversifood Congress 2018: Emma Forst, Jérôme Enjalbert, Julie Borg, Arnaud Gauffreteau, Bastien Paix & Isabelle Goldringer. Participatory ideotyping for organic and locally adapted wheat variety mixtures. Congrès Diversifood, 10-12 December 2018, Rennes, France. (Poster + oral flash presentation).
- Poster at the 10th IDEEV Day 2019: Emma Forst, Jérôme Enjalbert, Julie Borg, Arnaud Gauffreteau, Bastien Paix et Isabelle Goldringer. Participatory ideotyping for organic and locally adapted wheat variety mixtures. 10th journal of l'IDEEV, 24 January 2019, Gif-sur-Yvette, France (Poster).

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

UBIOS and INRAE – Winter wheat CCPs (Trial #3)

Lead: INRAE, partner UBIOS
Estelle Serpolay (estelle.serpolay@protonmail.com) and Isabelle Goldringer

Abstract

UBIOS Is a Seed Station belonging to the cooperatives BIOCER and COCEBI (Normandy and Burgundy). To enlarge their seed proposition to their members, and increase seed autonomy for the farmers, some farmers of the cooperatives, who are in touch with INRAE (Le Moulon), decided, together with Isabelle Goldringer, to create specific CCPs and test their adaptability in different production context. 2 CCPs (CCP1 and CCP2) were created in 2018, and a third one (CCP3) was created in 2019. They were multiplied at the farmers in 2019 and 2020 and testing started in 2021.

Introduction

Hypothesis and research question

Genetic diversity incorporated in CCPs can improve crop resilience and be attractive for farmers to work with participatory approach.

Objectives

The different CCPs were created to fit the different types of market of the farmers of the cooperatives: on-farm baking or selling grain to on-farm bakeries, as well as long chain markets, and poor and productive lands.

Material and Methods

Locations

The CCPs were sown in different environments in order to evolve in the conditions for which they were created and to be observed also in other environments to test their adaptability: they were sown in Burgundy Region (low potential soils) at a farmer (St Fargeau – VL) and on a varietal platform (Auxerre), some were sown in the Centre Region (Orveau-Bellesauve (farm) – JPB) where there are high potential soils, and in Normandy Region on a varietal platform (Ste Marie d'Attez - PlatBiocer) where there are also high potential soils. See table 1.

Genetic resources

11 "old varieties" and 4 modern varieties of winter wheat were selected to create 3 different CCPs.
"old varieties" : Rouge de Bordeaux, Rouge d'Alsace, Gros bleu, Ilde de France, Roux des Ardennes, Essex Glad Chaff, Champagne barbu, Automne Rouge barbu, Rouge d'hiver de Lozère, Blanc des Flandres and Redon Guer 332.
Modern varieties: Energo, Titlis, Geny and Butaro.

Creation of experimental population

3 basic CCPs were created (each one from 6 different varieties), and other populations were also created with the remaining seeds of the different crosses.

The 3 basic CCPs:

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CCP1 was created from 6 landraces with the objective to be adapted to productive lands and for on-farm bakery. CCP1 parents are : Rouge de Bordeaux, Rouge d'Alsace, Gros bleu, Ilde de France, Roux des Ardennes and Essex Glad Chaff.
CCP2 was created from 6 landraces with the objective to be adapted to poor lands and for on-farm bakery. CCP2 parents are : Rouge de Bordeaux, Champagne barbu, Automne Rouge barbu, Rouge d'hiver de Lozère, Blanc des Flandres and Redon Guer 332.
CCP3 was created from 4 modern varieties and 2 landraces, with the objective to be adapted to productive lands, and for long chain market. CCP3 parents are: Energo, Titlis, Geny, Butaro, Gros bleu and Rouge de Bordeaux.

The basic CCPs were created by bulking the same quantity of seed from each cross (CCPx – balanced). When some crosses have produced a biggest quantity of seeds, the "surplus" seed was bulked to create another CCP (CCPx – surplus).
5 CCP were finally created:

- CCP1
- CCP2 - balanced
- CCP2 – surplus
- CCP3 – balanced
- CCP3 – surplus

In addition, about 47 individual "lines" (descents of crosses) from each CCP1, CCP2 and CCP3 are observed individually.

	St Fargeau (farm) - VL	Orveau-Bellesauve (farm) - JPB	PlateBiocer	PlateCocebi
CCP1		X	X	X
CCP2 balanced	X		X	X
CCP 2 Surplus	X			
CCP3 balanced		X	X	
CCP3 surplus	X		X	X
F2 lines of CCP3		X		
F3 lines of CCP1 & 2		X		
CCP2 surplus selection	X			

Table 1. Cultivation place of each population in 2020 /2021 season

Evaluation

By 2020, CCP1 and CCP2 had been observed (crosses in 2018, multiplication of crosses lines harvested in 2019, mixture of the crosses and multiplication of the CCPs in 2020). Observations were done by INRAE-le Moulon consisting of heading date, lodging, and some measurements on spikes.

For the season 2020-2021, all the CCPs were observed by farmers, technicians, and researchers. The traits observed are presented in table 2.

Table 2. Observations planned to be performed in 2020/2021 season.

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	Crop development and agro-ecological performance (phenology, weeds, diseases, …)	
2020-2021		
Period	**Trait**	**How it has been assessed**
Full tillering	Tillering	Note from 1 (very weak) to 5 (very strong)
	Diversity of tillering	Note from 1 (very weak) to 5 (very strong)
	Plant growth habit	Note from 1 (very laid out) to 5 (very erected)
	Diversity of plant growth habit	Note from 1 (very weak) to 5 (very strong)
	Weeds - poaceae	Note from 1 (very weak) to 5 (very strong)
	Weeds - perrennial	Note from 1 (very weak) to 5 (very strong)
	Weeds - dicotyledonous	Note from 1 (very weak) to 5 (very strong)
	Vigour	Note from 1 (very weak) to 5 (very strong)
	Diversity of vigour	Note from 1 (very weak) to 5 (very strong)
Stem extension	Soil covering (by wheat)	Note from 1 (very weak) to 5 (very strong)
	Weeds - poaceae	Note from 1 (very weak) to 5 (very strong)
	Weeds - perrennial	Note from 1 (very weak) to 5 (very strong)
	Weeds - dicotyledonous	Note from 1 (very weak) to 5 (very strong)
	Leaves width	Note from 1 (very thin) to 5 (very large)
	Diversity of leaves width	Note from 1 (very weak) to 5 (very strong)
	Leaves colour	Note from 1 (very light) to 5 (very dark)
	Diversity of leaves colour	Note from 1 (very weak) to 5 (very strong)
	Global height	cm
	Stem extension speed	Plant height at different dates
Spring	Disease 1	Note from 1 (very weak) to 5 (very strong)
	Disease 2	Note from 1 (very laid out) to 5 (very erected)
	Disease 3	Note from 1 (very weak) to 5 (very strong)
	Disease 4	Note from 1 (very weak) to 5 (very strong)
	Global sanitary aspect	Note from 1 (very weak) to 5 (very strong)
	heading	Date and percentage of heading at the date
	Precocity	Note from 1 (very early) to 5 (very late), according to dates of heading
	Earing duration	Note from 1 (very short) to 5 (very long) according to dates of heading
	Weeds - poaceae	Note from 1 (very weak) to 5 (very strong)
	Weeds - perrennial	Note from 1 (very weak) to 5 (very strong)
	Weeds - dicotyledonous	Note from 1 (very weak) to 5 (very strong)

Other crop development and agronomic traits were not evaluated by the time of the writing of this deliverable but will be at the end of the growing season, such as plant height, lodging, spike colour, presence of awns etc. Productive performance traits will be chosen by the partners. A priori yield and some yield components on ears will be observed (number of kernels per spike, grain weight per spike, TKW).

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Results and discussion

The data will be analysed during the summer 2021 to be disseminated before the end of the project (so that farmers can choose if they continue or not the experiment).

How had the resilience improved?

Not yet evaluated at the agronomic level, but we suppose that genetic diversity will enable resilience for those populations. In addition, as new sources of diversified organic seeds will be available for the cooperatives' farmers, it will contribute to resilience at the seed system level.

Outcomes and conclusions (regarding the efficiency and the limits of the approach)

Main outcomes

- 3 different CCP created and under multiplication for the third or second year.
- From each CCP created (6 parents crossed), 2 populations were developed in a second step: a "balanced" CCP (mix of the same quantity of each cross) and a "surplus" CCP (mix of the crosses that have produced more seed).
- One farmer started a selection of different inbred lines derived from the crosses.

Main lessons learned

- Farmers' involvement at the different stages (choice of the CCP parents, observations, and breeding) is really motivating for them.
- Need of a facilitator to "coach" the farmers and make the link between farmers and scientists.
- One CCP created = more than one diversified population => many breeding possibilities (different populations created from the different crosses and some inbred lines derived from the crosses)

How the information can be transfer to the other situations?

When we have agronomic results, we will see if the genetic diversity of each population is a good starting point for "low tech breeding": creating one population and then multiplying (and thus adapting) it in diverse environments could be interesting to reduce breeding costs.
The whole history of the seed lots (creation of CCP, breeding and multiplication) has been recorded in a database system (SHiNeMaS: https://sourcesup.renater.fr/projects/shinemas) which allows to keep track of the information and relate the results to the processes in order to facilitate transfer to other situations.

Participatory/multi-actor approach

The participatory approach is the basis of this experiment. Farmers, researcher, and facilitator work together. The different choices and decisions (what to cross with what, which observations to do, etc.) are done in collaboration during regular meetings. Observations are done by farmers and facilitator, and eventually by researcher. The trials are set up by the farmers. Selection if any, is done by the farmers. The facilitator is organising meetings, helping with observations and harvest.

Publications

No references yet

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ITAB – Comparing bread wheat CCPs and DOPs (COBRA populations (Trial #4a)

Lead: Emma FLIPON (ITAB), partner: INRAE

Abstract

The objective of the COBRA experiment was to compare two selection strategies: CCPs and Dynamic populations for a self-pollinating species, bead wheat, in terms of diversity level and adaptation.
The 10 COBRA populations were cultivated in 2017-2018 and in 2018-2019 in the same locations (Saudrais and Bouchemaine).
The hypothesis was that: (1) diversity will evolve differently according to the initial "mixing" methods (simple mixture or crosses) and (2) the new populations will have different potentialities to adapt to farmer or processor's needs and to the environment (see deliverable 3.1).
The results have shown in 2019 that human selection mainly determines crop traits (phenotypic characteristics and yield components). The other determinant factor is the location. The farmer's and bakers' selection have conserved the overall diversity even if they have fostered some chosen traits. There are no significant differences between the two selection strategies, after 5 generations, which invalidates our hypothesis.
For the period 2019-2020, it was not possible to saw the populations at the location "Saudrais" because of the climatic conditions (heavy rainfalls), but they were sawn in Bouchemaine. Then, the populations will only be observed at Bouchemaine place before harvest time, by the end of July.

Introduction

For several years, some farmers in France have developed farm varieties for organic farming with high diversity, by mass selection, mixtures (that continue to evolve by open pollination – they are called dynamic populations) and/or by crosses. Those different strategies are questioning some farmers and researchers about the breeding potential of the different strategies (are they different, if yes for what type of characteristics). To explore this question, INRA-ITAB team has created 2 experimental bread wheat populations in 2013 from the same 6 parents (landraces): one dynamic population by mixing seeds of the 6 parents, and a Composite Cross Population (CCP) by crossing all the parents together (2x2, each parent being once male and once female). The 2 populations are cultivated in 2 places since 2015 and are evaluated each year for phenotypical characteristics and level of diversity.
The populations have been created in the framework of a European project called COBRA. Therefore, they were named as" COBRA populations" to recall their origin.

Hypothesis and research question

In 2018, a farmer (in one location) and 2 bakers (in the other location) have realized a mass selection. In 2019, we evaluated phenotypical diversity of the different populations ("original populations" in both places and selected populations in both places too).

Hypothesis in 2019:
We wanted to explore the impact of 3 factors on the behaviour (deriving from the evolution) of the different versions of the COBRA populations:

- Impact of location (Pays de la Loire vs Brittany);

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- impact of breeding strategy (CCP versus dynamic population);
- impact of human breeding (selection by bakers and farmer).

We will look at the behaviour and at the level of diversity.
Our hypotheses are that:
- level of diversity of CCPs is higher than the one the dynamic populations;
- location influences the behaviour and level of diversity of the populations;
- human selection homogenises the populations.

Material and Methods

Location

COBRA trials. The 10 populations cultivated in 2017-2018 were sown in the same locations (Saudrais and Bouchemaine) in 2018-2019, West of France, 2 locations: Britany (Rennes) and Pays de la Loire (Angers), and in 2020, West of France, 1 location: and Pays de la Loire (Angers).

Genetic resources

The 6 parents chosen are landraces from a INRA gene bank, multiplied in a farmer's field for 4 years at least before 2013 and judged interesting by the farmer and Véronique Chable:
- Bladette de Provence
- Blé de Redon Guer 335
- Blé de Redon Guer B7
- Blé de Redon Sixt dur Aff 346
- Blé de Redon Roux pâle 1.13
- St Priest et le Vernois Rouge

In 2019 were assessed the 2 COBRA populations (PopDyn and CCP) in different versions:
cultivated for 4 years in Pays de la Loire
cultivated for 4 years in Britany
selected in 2018 by baker 1 in Britany
selected in 2018 by baker 2 in Britany
selected in 2018 by farmer in Pays de la Loire
There were in total 10 modalities.

the basic populations were created from 6 landraces from INRA gene bank and multiplied in a farmer's field for 4 years at least before 2013 and judged interesting by him and Véronique Chable (INRAE):
- Bladette de Provence
- Blé de Redon Guer335
- Blé de Redon Guer B7
- Blé de Redon SixtdurAff 346
- Blé de Redon Roux pâle 1.13
- St Priest et le Vernois Rouge

In 2020, were assessed the 2 COBRA populations (PopDyn and CCP) in different versions:
-cultivated for 4 years in Pays de la Loire
-selected in 2018 by a farmer located in Anjou
4 modalities were then observed.
The populations were not observed in Brittany, contrary to the other years.

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The initial populations were created from 6 landraces coming from the INRA gene bank and multiplied in a farmer's field for 4 years (before 2013) and were chosen by this farmer and an INRAE researcher, Véronique Chable:
- Bladette de Provence
- Blé de Redon Guer335
- Blé de Redon Guer B7
- Blé de Redon Sixt sur Aff 346
- Blé de Redon Roux pâle 1.13
- St Priest le Vernois Rouge

Experimental design

2019 trials were sown with farm material for basic populations (in natural evolution) in plots of about 200 square meters. There were no replicates, but 3 small plots identified within the 200 square meter for some measurements. For human selections (done in 2018), plots sown were smaller because the small quantity of seeds available (grain from 60 spikes). The plots were about 10 square meters and sown with experimental sowing machine of hand sowing machine. The scheme after presents the "map" of the trials. In green, the basic populations, in yellow the selections. "La Saudrais" = Brittany and "Bouchemaine" = Pays de la Loire (Figure 1).

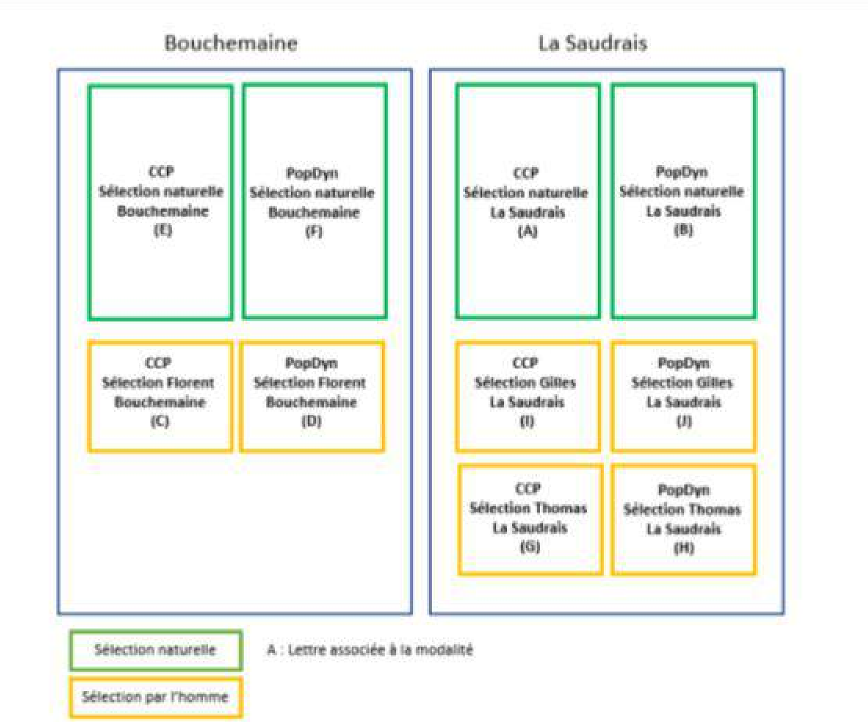

Figure 1. Plot design of the COBRA trials

Evaluation

The evaluated traits are presented in Table 1-3 below.

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Table 1. Crop development and agro-ecological performance traits

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
2019		
Trait	**How it has been assessed**	**Type of data available**
weeds	**Score** from 1 (very few weeds) to 5 (too many weeds), in small plots inside the experimental fields (3 small plots per field at Rennes Location)	Score This measurement was not done in 2019
diseases/**health**	**Score** from 1 (a lot of diseases) to 5 (very good health), in 3 small plots per experimental plot (excepted for selection plots)	Score
Vigor	Subjective observation… **Score** from 1 (very weak vigor) to 5 (very vigorous), in 3 small plots per experimental plot (excepted for selection plots)	Score
2020		
diseases/**health**	**Score** from 1 (a lot of diseases) to 5 (very good health), in 3 small plots per experimental plot (excepted for selection plots)	Score
Vigor	Subjective observation… **Score** from 1 (very weak vigor) to 5 (very vigorous), in 3 small plots per experimental plot (excepted for selection plots)	Score
morphological traits: **-** awns - spike color* only in 2019 - spike shape	observation of spikes and attribution of a score to each spike for each trait for all versions of COBRA populations	Scores

Table 2. *Crop development and agro-ecological performance traits*

Crop productive performance (yield, yield components)		
2019 and 2020		
Trait	**How it has been assessed**	**Type of data available**
Plant measurements: - last leaf-spike base distance - total plant height	Measuring 60 stems per experimental plot at maturity, in cm	Distances in cm
Spikes measurements (yield components):	Counting on 60 spikes per experimental plot (the 60 spikes corresponding to the 60 stems so that the data can be linked)	numbers

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- number of spikelet (sterile and fertile ones) - number and weight of grains per spike		
yields: - straw - grain	weight of the straw and grain collected on the 3 small plots for populations in natural evolution	quintal/ha

Statistical methods

The results were treated by R software and presented differently for qualitative and quantitative data. Qualitative data were as presented as frequencies and Nei indexes for diversity. Quantitative data were presented with boxplots (showing median, third and fourth quantiles, min and max) and with PCA for diversity.

Participatory/multi-actor approach

The idea of the experimentation comes from a discussion between farmers and researchers. Véronique Chable started the experimentation with a farmer. The scheme below (Figure 2) sums up the story and the management of the process of this experimentation.

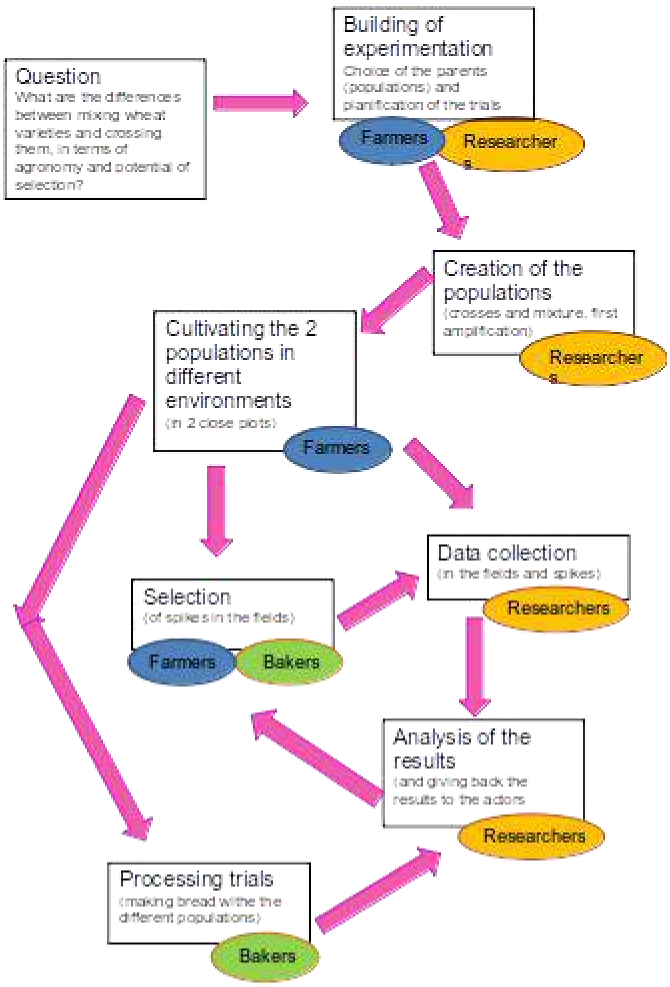

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Figure 2. Participatory approach in the COBRA trials

Results and discussion

The results are summarized in Table 4 below, providing aa synthesis, answering the hypothesis.

		Effect of breeding strategy	Effect of human selection
Qualitative data	Spike's colour	-less diversity in the selected Dynamic Population -CCP and PopDyn selected naturally have the same diversity	
	Spike shape		-more diversity in the CCP population that evolved without selection
	Awns		-less diversity in the CCP population that evolved without selection
	Husks	-May include a measurement bias	-May include a measurement bias -more diversity in the PopDyn that was not selected by the farmer
	Overall diversity		Diversity level is equivalent between populations selected by the farmer and naturally selected populations.
Quantitative data	Diversity and agronomical characteristics	More important diversity in PopDyn than in CCP	-increased performances in populations selected by the farmer (CCP and PopDyn)

Table 4 Summary of the results of the COBRA trials

In the following, we include graphics to illustrate some of the results synthesised in the table (only examples because a lot of graphics are available).

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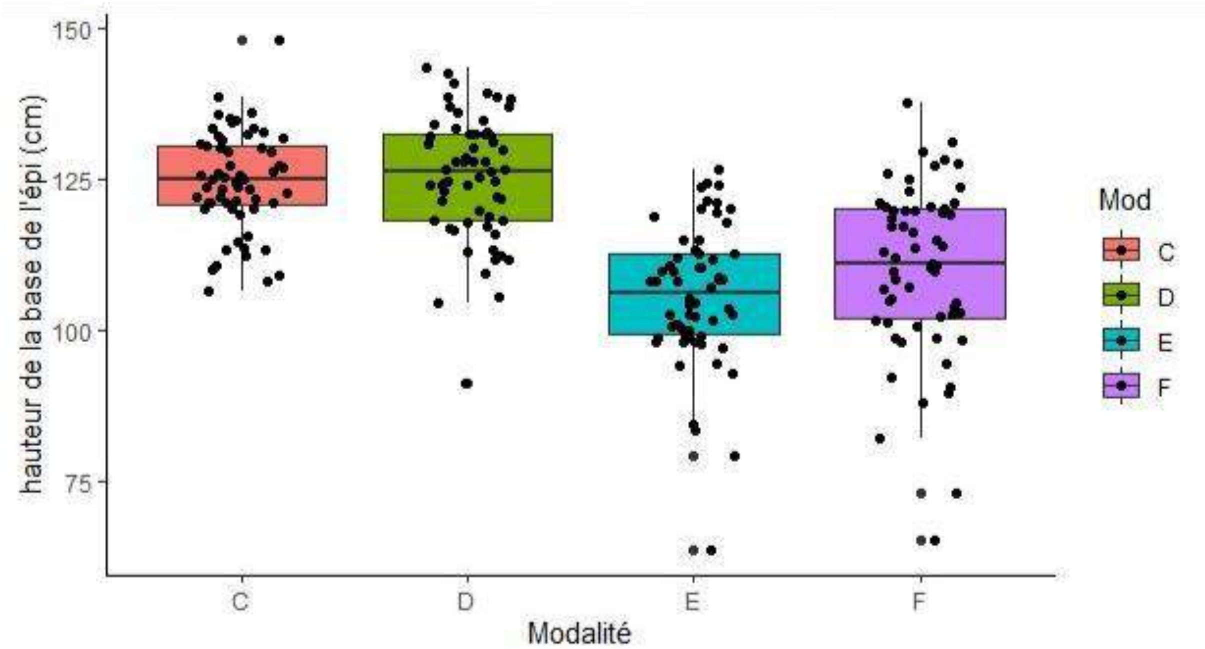

Figure 3. Height of the spike's base for each modality

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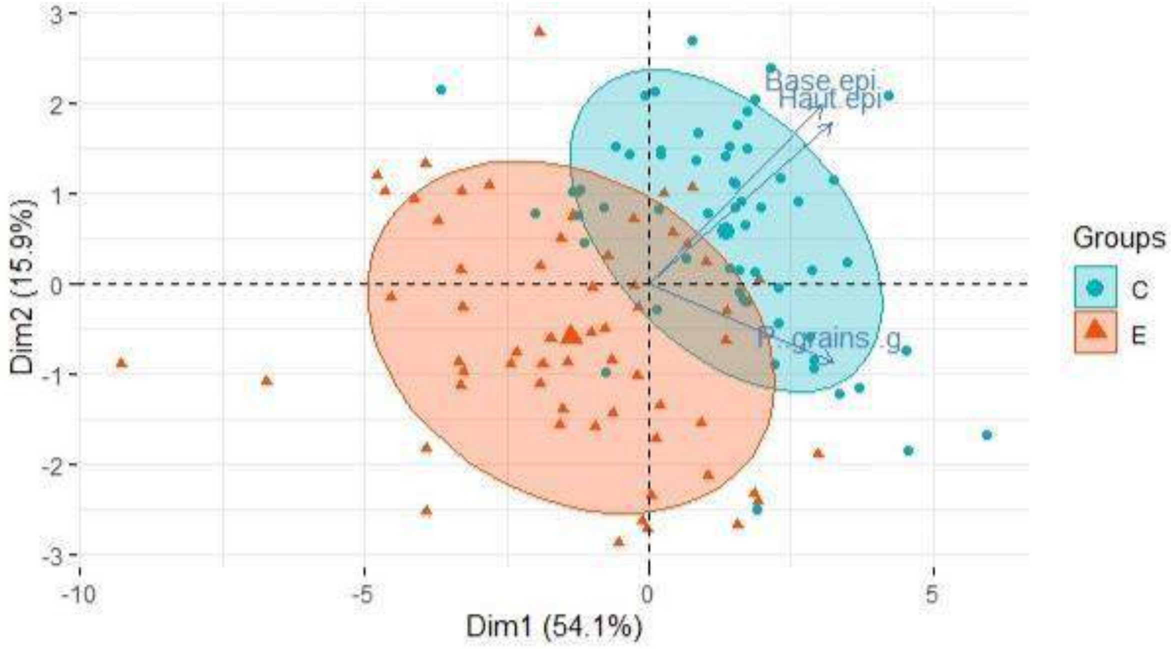

Figure 4. PCA of the CCP populations (E=natural selection, C=Farmer's selection)

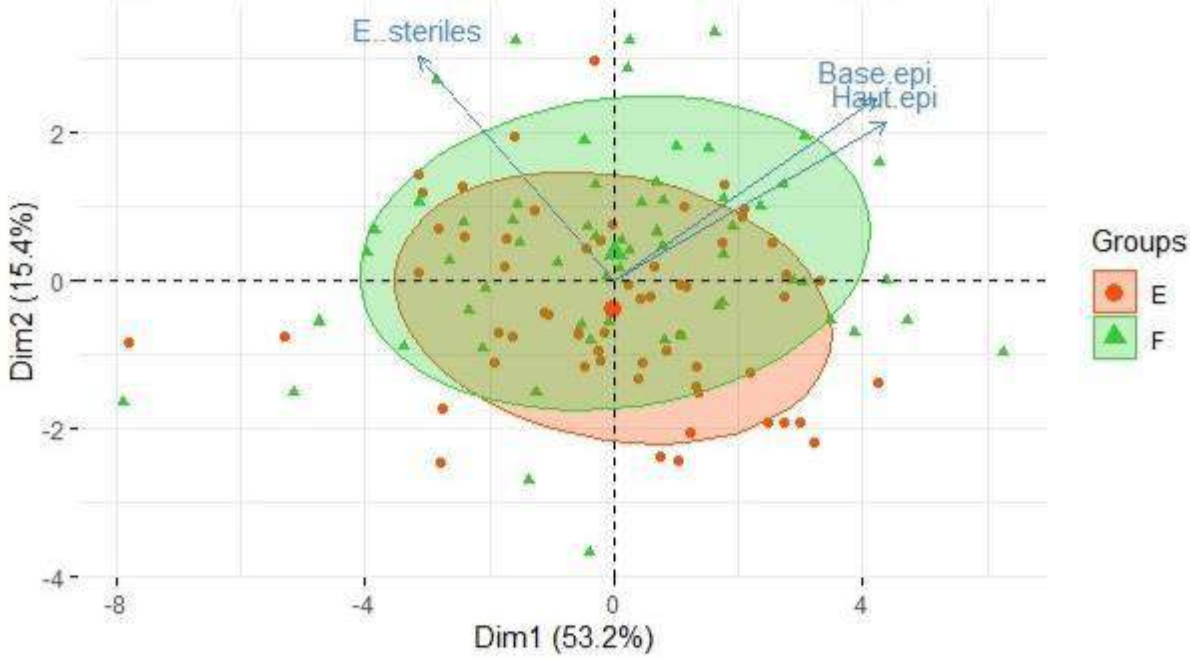

Figure 5. PCA of the Natural selection (E=CCP selection, D=PopDyn)

Genetical analyses were also conducted on each parent population. The main outcomes are individuals are diversified among the populations (Fis<0):

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Fis	Indice de Fixation intra-population					
Bladette	St Priest	Sixt sur Aff	Guer B335	Redon Roux Pale	Guer B7	Tot
-0.9887	-0.6874	-0.8587	-0.7066	-0.7805	-0.8292	-0.8176

Table 5. Genetical analyses of the COBRA population

The results have shown in 2019 that human selection mainly determines crop traits (phenotypic characteristics and yield components). The other determinant factor is the location. The farmer's and bakers' selection have conserved the overall diversity even if they have fostered some chosen traits. There are no significant differences between the two selection strategies, after 5 generations, which invalidates our hypothesis.

For the period 2019-2020, it was not possible to sow the populations at the location "Saudrais" because of the climatic conditions (heavy rainfalls), but they were sawn in Bouchemaine. Then, the populations will only be observed at Bouchemaine place before harvest time, by the end of July.

Like in 2019, human selection seems to be the most impacting factor on the performances and diversity of the populations. The breeding strategy seems to have a lesser impact.
During 2019/2020 season we preferred to keep the same protocol as followed during the previous years so that it could be comparable and so that the conclusions of the 2018/2019 season could be validated.

The results of 2020 are aligned with those of the previous year. To go further with this experiment, it would be necessary to observe more than 60 spikes per modality, and to engage bakers to test the transformation and hedonic aspects of the populations.

Outcomes and conclusions

From these results, we can see that the human selection is the factor that impacts more the populations, as it was observed in 2019. Then, in 2019 came the influence of the location, which we could not assess this year due to the climatic conditions. The breeding strategy has a smaller impact on the diversity and agronomical characteristics of the populations.

Looking at Nei indexes, human selection doesn't homogenise the populations but seems to preserve the diversity. Again, this result confirms the results from 2019.

Therefore, we conclude:

-Human selection has a greater impact on agronomical characteristics than the breeding methods used to create the new populations
-Human selection has a positive impact on some agronomical characteristics of the populations
-Human selection on some traits does not seem to reduce the global intra-varietal diversity
-No difference observed between CCP and PopDyn in terms of their internal diversity

Next steps:
- farmer's and bakers' selection evaluation (from fields trials)
- proposing the populations to other farmers and bakery informal tests

How resilience was improved

CCPs and mixtures improve resilience by increasing diversity in the farms and the fields.

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Main lessons learned

Farmers starting on farm selection and wishing to increase intra varietal diversity may prefer mixtures as they are easy to make and are produce diversified populations. CCPs are interesting for more precise selection.

How the information can be transferred to the other situations?

Mixtures are easiest to handle than CCP and this result will encourage farmers to create their own populations.

Publications

-Internship Master thesis: Comparison of two strategies of creation of soft wheat diversified population adapted to organic farming
-Eucarpia presentation: "Comparing two selection strategies of bread wheat diversified populations adapted to organic farming", 2021

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ITAB – INRAE – DOP Populations (Trial #4b-d)

Lead: INRAE, Partner: ITAB
Véronique Chable, Emma Flipon

Abstract

The new methodology is an additional tool to spread and use diversity for organic agriculture. It was implemented during DIVERSIFOOD project and continued in the framework of LIVESEED. Three species were used to set up the method: rivet wheat, spelt and oat.

A Diversified Oriented Population (DOP) is a mixture of several accessions of a same species, which have one or several common phenotypic traits requested by a farmer. Once on farm, these diversified populations are supposed to have a great potential for adaptation. Farmers involved have been satisfied with the method and we wish to develop it for more species.

Introduction

Hypothesis and research question

INRAE and ITAB collaborated to propose a new method to enhance intra-specific diversity within farmer's populations of (underutilized) cereals. Important diversity available in gene banks has not been mobilized yet. Hundreds of accessions of a species are requested in gene banks, multiplied, and observed (for main phenological and morphological criteria) by the team for 2 to 4 years.

Then the accessions are mixed according to the farmer's choice among these criteria.
Hypothesis:
(1) those populations have a great adaptation capacity due to their important diversity
(2) the populations' morphological characteristics described and chosen by the farmers are common to all components of the DOP and may be easily conserved from one generation to the next ones.

This work has been started during the Diversifood project for rivet wheat (4 years), spelt (2 years) and oats (1 year). In LIVESEED, 2018, 157 varieties of rivet wheat were used to create populations that were sent to 24 farmers. 29 personalized populations were created. In 2019, new populations were sent to 10 farmers and at least 6 farmers are resewing the populations they received in 2018.
These populations will be observed by the farmers: some qualitative feedbacks are expected before harvest from them, mainly about the criteria they have chosen.

Material and Methods

Location(s)

The populations were multiplied in 2017 in France, at La Saudrais, Rennes for Rivet Wheat, and at Domaine de la Motte, INRA, Le Rheu for spelt and oats. Then, in 2018 and 2019, rivet wheat populations were sent to respectively 24 and 10 farmers all over France and Belgium.

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In 2020 and 2021 the spelt and oats accessions were not sown, for whether and time reasons. For rivet wheat, some mixtures were sown for multiplication in autumn 2020: INIA, mealy and vitreous, because they were the populations that were asked in majority. They were sown in Le Rheu, Brittany, France.

Genetic resources

Rivet wheat accessions evaluated

Gene bank	Species	Group	Accession name
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	Ou 1 OR BGE 013721
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE000125
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE000168
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE002866
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE002870
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE002873
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE002874
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE002887
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	dreischianum	BGE012400
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	rubroalbum	BGE012488
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	rubroalbum	BGE012489
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012534
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Lusitanicum	BGE012535
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012536
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012537 (rec 2017)
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012538
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	dreischianum	BGE012539
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	speciosum	BGE012540

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CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	speciosum	BGE012541
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	speciosum	BGE012542
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	speciosum	BGE012543
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Lusitanicum	BGE012544
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012545
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar martensii	BGE012546
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE012547*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Nigrobarbatum	BGE012548
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar martensii	BGE012549
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar martensii	BGE012550
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar martensii	BGE012551
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar martensii	BGE012552
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	dreischianum	BGE012554
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012555
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012556
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012557
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012558
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar turgidum	BGE012559
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar turgidum	BGE012560
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE012561
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	var rubroatrum	BGE012562
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar turgidum	BGE012563

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CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	var rubroatrum	BGE012564
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE012565
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	pseudocervicum	BGE012567
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	plinianum	BGE012568
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	pseudocervicum	BGE012569
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	plinianum	BGE012570
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	plinianum	BGE012571
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE012572
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	ramososalomoni s	BGE012573
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013074
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Lusitanicum	BGE013076
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013078
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013081
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013083
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013084
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013085
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013086
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013087
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013088
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	speciosum	BGE013089
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013090
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013091

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CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013092
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013094
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013095
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013097
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013098
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013099*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013101
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013102
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013104
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar turgidum	BGE013711
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar turgidum	BGE013712
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	dreischianum	BGE013717
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013718
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013719
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	OR BGE013720
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013721
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013722
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013723*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013724
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013725
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013726
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE013727

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CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Bucale	BGE013728
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Lusitanicum	OR BGE013729
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013730
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013731*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	megalopolitanum	BGE013732
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013733*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	convar turgidum	BGE013734
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013735*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013736*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013737*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013738*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013739*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE013740*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE013741
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE015009
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE015014
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE015016
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE015395
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018619
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	gentile	BGE018621
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	gentile	BGE018622
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018623

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CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Herrerae	BGE018625
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018626
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018627
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018628
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018641
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018642
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018643
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018644
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018647
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Lusitanicum	BGE018653
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Lusitanicum	BGE018654
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE018655*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Herrerae	BGE018656
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	speciosum	*OR BGE018657*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	var rubroatrum	BGE018659
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018675
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018676
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018677
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018678
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE018680
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE019248
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE019256

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CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	*OR BGE019289*
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE019291
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	var rubroatrum	BGE019293
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE019301
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE019308
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	var rubroatrum	BGE020904
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	var rubroatrum	BGE020905
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	var rubroatrum	BGE020906
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020907
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020939
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020940
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Nigrobarbatum	BGE020941
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	speciosissimum	BGE020942
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020943
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020944
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020945
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Salomonis	BGE020946
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020947
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020948
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020949
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020950
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE020951

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CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	candiens	BGE020952
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	plinianum	BGE020954
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021772
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021773
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Salomonis	BGE021774
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021816
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021818
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021819
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021820
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021823
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021824
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021825
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021826
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Turgidum	BGE021828
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE030923
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE030924
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE030925
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE030926
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE030927
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	Dinurum	BGE030928
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	iodurum	BGE040242
CENTRO NACIONAL DE RECURSOS FITOGENÉTICOS	Triticum turgidum	var martensii	BGE040245

Spelt accessions evaluated (2018-2019)

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Gene bank	Accession number	Accession name
Agroscope	TS0018	Sommerkorn 3L
Agroscope	TS0023	Sommerkorn 3Q
Agroscope	TS0072	VOR 6F
Agroscope	TS0073	Osch 1A
Agroscope	TS0094	Osch 20A
Agroscope	TS0095	Osch 20B
Agroscope	TS0961	Gugg 2G
Agroscope	TS0974	Gugg 6B
Agroscope	TS1029	Napf 3A
Agroscope	TS1038	Napf 3P
Agroscope	TS1056	Napf 9E
Agroscope	TS1060	Mit. 1B
Agroscope	TS1084	Toess 1C
Agroscope	TS1087	Toess 2B
Agroscope	TS1103	Toess 4A
Agroscope	TS1112	Toess 5D
Agroscope	TS1113	Toess 6A
Agroscope	TS1114	Toess 6B
Agroscope	TS1116	Toess 6E
Agroscope	TS1120	Toess 8B
Agroscope	TS1122	Toess 8E
Agroscope	TS1123	Toess 8F
Agroscope	TS1170	Bld. 13D
Agroscope	TS1185	Bld. 18E
Agroscope	TS1264	Bld. 48A
Agroscope	TS1460	ARD. 18B
Agroscope	TS1462	ARD. 18D
Agroscope	TS2124	Winiger-Egg Weiss
Agroscope	TS2278	Zuzgen Zg 15A
Agroscope	TS2308	Salez
CRB Céréales à paille	2768	EPEAUTRE BLANC 1
CRB Céréales à paille	2772	EPEAUTRE BLOND OU DORE
CRB Céréales à paille	2774	EPEAUTRE DE L'AVEYRON
CRB Céréales à paille	2775	EPEAUTRE NAIN
CRB Céréales à paille	6329	ROUQUIN
CRB Céréales à paille	7857	T. SPELTA ALBUM

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Institute of plant genetic resources "K. Malkov"	1980-TRT-SP1	bauig-niederwill
Institute of plant genetic resources "K. Malkov"	1980-TRT-SP2	Blauer winter-kolbendinkel
Institute of plant genetic resources "K. Malkov"	1980-TRT-SP3	Brauner winter-granendinkel
Institute of plant genetic resources "K. Malkov"	1982-TRT-SP1	roter grannenspelz
Institute of plant genetic resources "K. Malkov"	1982-TRT-SP7	renval
Institute of plant genetic resources "K. Malkov"	1987-TRT-SP10	Oberkulm 3
Institute of plant genetic resources "K. Malkov"	1987-TRT-SP17	aus kruzung
Institute of plant genetic resources "K. Malkov"	1987-TRT-SP21	ast.118
Institute of plant genetic resources "K. Malkov"	1987-TRT-SP24	Asturien
Institute of plant genetic resources "K. Malkov"	1988-TRT-SP1	oberkulmer winterdinkel
Nodik	RCAT 002731	
Nodik	RCAT 002732	
Nodik	RCAT 002733	
Nodik	RCAT 002735	
Nodik	RCAT 002736	
Nodik	RCAT 002737	
Nodik	RCAT 002739	
Nodik	RCAT 002740	
Nodik	RCAT 002741	
Nodik	RCAT 002743	
Nodik	RCAT 002745	
Nodik	RCAT 002746	
Nodik	RCAT 002747	
Nodik	RCAT 002748	
Nodik	RCAT 002752	
Nodik	RCAT 002755	
Nodik	RCAT 002756	
Nodik	RCAT 002758	

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Nodik	RCAT 003694	
Nodik	RCAT 0053495	
Nodik	RCAT 003588	
	TRI 11523	
	TRI 263	Steiners Roter Tiroler Dinkel
	TRI 1484	
	TRI 2256	
	TRI 2258	
	TRI 3086	Red winter spelt
	TRI 3309	
	TRI 3665	
	TRI 16774	
	TRI 3666	
	TRI 3770	
	TRI 3772	
	TRI 4273	
	TRI 4298	
	TRI 4318	
	TRI 4394	
	TRI 4439	
	TRI 4473	
	TRI 4609	
	TRI 4610	
	TRI 4611	
	TRI 4612	
	TRI 4613	
	TRI 4629	
	TRI 4656	Baetting-Niederwill
	TRI 474	speltz aus tzaribord
	TRI 685	
	TRI 982	Emmer aus Tharib
	TRI 4684	Brauner spelz aus schefflenz
	TRI 4685	Brengenzer roter spelz
	TRI 4770	Farnsburg 6
	TRI 5309	
	TRI 5648	
	TRI 14165	

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Oats accessions evaluated (2018-2019)

NATIONAL_INVENTORY_NAME	INSTITUTE_CODE	ACCESSION_NUMBER	ACCESSION_NAMES
United Kingdom	GBR016	ABY-Bs 4006	Alsovadaszi
United Kingdom	GBR016	ABY-Bs 4007	ANARCSI
United Kingdom	GBR016	ABY-Bs 4008	Balatonfokajari
United Kingdom	GBR016	ABY-Bs 4009	Baltonmogyorodi
United Kingdom	GBR016	ABY-Bs 4013	BUKI
United Kingdom	GBR016	ABY-Bs 4015	DREGELYPALANKI I
United Kingdom	GBR016	ABY-Bs 4016	DREGELYPALANKI II
United Kingdom	GBR016	ABY-Bs 4017	Gamasi
United Kingdom	GBR016	ABY-Bs 4022	Kecskemeti
United Kingdom	GBR016	ABY-Bs 4023	Kompolti
United Kingdom	GBR016	ABY-Bs 4028	MOCSAI
United Kingdom	GBR016	ABY-Bs 4029	Nagyatadi
United Kingdom	GBR016	ABY-Bs 4030	Pocspetri
United Kingdom	GBR016	ABY-Bs 4031	PUSPOKI
United Kingdom	GBR016	ABY-Bs 4033	Taplanszentkereszti
United Kingdom	GBR016	ABY-Bs 4034	TUNYOGI
United Kingdom	GBR016	ABY-Bs 4036	ZALAHASHAGYI
United Kingdom	GBR016	ABY-Bs 4037	ZALALOVOI
United Kingdom	GBR165	SASACO001	Black Winter
United Kingdom	GBR165	SASACO002	Grey Winter
United Kingdom	GBR165	SASACO003	BLACK TARTARIAN
United Kingdom	GBR165	SASACO004	Black Mogul
United Kingdom	GBR165	SASACO005	BLAINSLIE
United Kingdom	GBR165	SASACO006	Murkle Oats 1
United Kingdom	GBR165	SASACO007	Murkle Oats 2
United Kingdom	GBR165	SASACO008	Radnorshire spring
United Kingdom	GBR165	SASACO009	Mukrle Black Oat 009
United Kingdom	GBR165	SASACO010	Mukrle Black Oat 010
United Kingdom	GBR165	SASACO011	Mukrle Black Oat 010
United Kingdom	GBR247	O0019	Hungarian Winter Oat
United Kingdom	GBR247	O0034	Y 1-4
United Kingdom	GBR247	O0035	Y 1-11
United Kingdom	GBR247	O0196	Chinese No 9
United Kingdom	GBR247	O0197	Japan
United Kingdom	GBR247	O0204	West Australian
United Kingdom	GBR247	O0230	North Finnish

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United Kingdom	GBR247	O0236	West Finnish
United Kingdom	GBR247	O0241	Bohus 306
United Kingdom	GBR247	O0249	Gothlandshavre
United Kingdom	GBR247	O0253	Oland 0691
United Kingdom	GBR247	O0260	Smalands 464
United Kingdom	GBR247	O0419	Hungarian A
United Kingdom	GBR247	O0420	Hungarian B
United Kingdom	GBR247	O0421	Hungarian C
United Kingdom	GBR247	O0422	F Fehar Tavasi Zab
United Kingdom	GBR247	O0446	Sovetskij 0339
United Kingdom	GBR247	O0449	Ozimil Oves 276
United Kingdom	GBR247	O0450	Ozimil Oves 277
United Kingdom	GBR247	O0498	Cenad 88
United Kingdom	GBR247	O0499	Cenad 2T
United Kingdom	GBR247	O0500	6443 S Franciska
United Kingdom	GBR247	O0508	6686 S Jose
United Kingdom	GBR247	O0509	Ankara 3-1252
United Kingdom	GBR247	O0510	Marokko 238
United Kingdom	GBR247	O0515	Krimszkij 90
United Kingdom	GBR247	O0518	Hatvani 187
United Kingdom	GBR247	O0519	Hatvani 181
United Kingdom	GBR247	O0520	Hatvani 117
United Kingdom	GBR247	O0525	Moskovszkij A 315
United Kingdom	GBR247	O0526	Harkovszkij 596
United Kingdom	GBR247	O0601	Tshan-Bei-e-Shen-Tshin
United Kingdom	GBR247	O0608	Balkanischer Nackter
United Kingdom	GBR247	O0636	Nurenberg
United Kingdom	GBR247	O0858	Poland
United Kingdom	GBR247	O0943	Tulunskii Golezernvi
United Kingdom	GBR247	O0947	Sovetskij
United Kingdom	GBR247	O0948	Skajstunes
United Kingdom	GBR247	O0949	Stendskij Zeltyj
United Kingdom	GBR247	O0950	L'Govskij 1026
United Kingdom	GBR247	O0951	Krasnodarskij 73
United Kingdom	GBR247	O0952	Naked
United Kingdom	GBR247	O0969	Bohus 558
United Kingdom	GBR247	O0972	Naked
United Kingdom	GBR247	O0994	Balkanischer Nackter
United Kingdom	GBR247	O1004	Nu 1 aus Frankreich

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United Kingdom	GBR247	O1005	Chinese Hulless
United Kingdom	GBR247	O1006	Nu 4 aus Frankreich
United Kingdom	GBR247	O1051	Hulless
United Kingdom	GBR247	O1055	Naakte Haver uit Overasselt
United Kingdom	GBR247	O1060	Yenmesh
United Kingdom	GBR247	O1097	Golezernvi Osipeva
United Kingdom	GBR247	O1111	Hulless
United Kingdom	GBR247	O1112	Hulless
United Kingdom	GBR247	O1113	Hulless
United Kingdom	GBR247	O1117	Hulless
United Kingdom	GBR247	O1118	Mongolia
United Kingdom	GBR247	O1119	Alaskan Hulless
United Kingdom	GBR247	O1131	L'Govskij 78
United Kingdom	GBR247	O1167	Ethiopia 12/7
United Kingdom	GBR247	O1168	Ethiopia 8/5
United Kingdom	GBR247	O1178	Ethiopia 8
United Kingdom	GBR247	O1184	Ethiopia 13B
United Kingdom	GBR247	O1185	Ethiopia 14
United Kingdom	GBR247	O1203	Ethiopia 33
United Kingdom	GBR247	O1219	Turkey 64
United Kingdom	GBR247	O1398	Dotnuvos 489/1 Is Pasvalio
United Kingdom	GBR247	O1399	Dotnuvos Baltosios
United Kingdom	GBR247	O1400	Stipruoles
United Kingdom	GBR247	O1401	Dotnuvos 301/1-308/2
United Kingdom	GBR247	O1419	Haleto Amaiella
United Kingdom	GBR247	O1475	Ardena Amorilla
United Kingdom	GBR247	O1476	Blanca Alemana
United Kingdom	GBR247	O1477	Blev d'Or Aurilla
United Kingdom	GBR247	O1479	Nassengrund
United Kingdom	GBR247	O1480	Port Adelaide
United Kingdom	GBR247	O1484	Ethiopian X
United Kingdom	GBR247	O1503	6290 Avena di Sao Francesco
United Kingdom	GBR247	O1504	Portuguese Regional Oat
United Kingdom	GBR247	O1507	Mlochowski
United Kingdom	GBR247	O1519	Turkish
United Kingdom	GBR247	O1520	Turkish
United Kingdom	GBR247	O1521	Turkish
United Kingdom	GBR247	O1630	Alaskan Hulless
United Kingdom	GBR247	O1631	Balkanischer

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United Kingdom	GBR247	O1740	Nurnberg 1832
United Kingdom	GBR247	O2074	6370 Avena 5106
United Kingdom	GBR247	O2075	6365 Avena Castanha
United Kingdom	GBR247	O2100	6368 Avena 8608
United Kingdom	GBR247	O2101	6374 Avena Vulgar
United Kingdom	GBR247	O2102	6372 Avena Peluda
United Kingdom	GBR247	O2103	6372 Avena Peluda
United Kingdom	GBR247	O2104	6357 Rosseau
United Kingdom	GBR247	O2105	6352 Avena de Russus
United Kingdom	GBR247	O2106	6358 Rinkau
United Kingdom	GBR247	O2108	Nuda chinensis 1930
United Kingdom	GBR247	O2112	Sativa aristata Kazakhstan
United Kingdom	GBR247	O2126	Sativa mutica Hindu Kush
United Kingdom	GBR247	O2127	Sativa aurea Hindu Kush
United Kingdom	GBR247	O2128	Buddha
United Kingdom	GBR247	O2129	Sativa krausei Hindi Kush
United Kingdom	GBR247	O2132	Sativa montana Hindu Kush
United Kingdom	GBR247	O2154	Sativa genuina mirtica
United Kingdom	GBR247	O2196	Sativa mutica Hindu Kush
United Kingdom	GBR247	O2197	Sativa krausei Hundu Kush
United Kingdom	GBR247	O2198	Sativa krausei
United Kingdom	GBR247	O2207	Ibiza
United Kingdom	GBR247	O2210	Sativa Abyssinica
United Kingdom	GBR247	O2216	Sativa
United Kingdom	GBR247	O2217	Sativa nudae chinensis
United Kingdom	GBR247	O2219	Sativa aristata
United Kingdom	GBR247	O2225	Sativa 4649
United Kingdom	GBR247	O2247	6366 Sativa genuina
United Kingdom	GBR247	O2251	Sativa purpurea
United Kingdom	GBR247	O2252	Sativa
United Kingdom	GBR247	O2301	Afghan R 1185/1
United Kingdom	GBR247	O2311	Ethiopia R 55/1
United Kingdom	GBR247	O2312	Ethiopia R 69/2
United Kingdom	GBR247	O2315	Ethiopia R 180
United Kingdom	GBR247	O2318	Ethiopia R 195/1
United Kingdom	GBR247	O2320	Ethiopia R 196/1
United Kingdom	GBR247	O2321	Ethiopia R 76/2
United Kingdom	GBR247	O2323	Ethiopia R 213/2
United Kingdom	GBR247	O2324	Ethiopia R 218/1

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United Kingdom	GBR247	O2342	Turkey R 2
United Kingdom	GBR247	O2345	Ethiopia R 58/1
United Kingdom	GBR247	O2351	Ethiopia R 83
United Kingdom	GBR247	O2352 A	Ethiopia R 88
United Kingdom	GBR247	O2352 B	Ethiopia R 88
United Kingdom	GBR247	O2358	Ethiopia R 204/1
United Kingdom	GBR247	O2360	Ethiopia R 220
United Kingdom	GBR247	O2363	Ethiopia R 235
United Kingdom	GBR247	O2365	Ethiopia R 400
United Kingdom	GBR247	O2369	Ethiopia R 418
United Kingdom	GBR247	O2370	Ethiopia R 425
United Kingdom	GBR247	O2372	Ethiopia R 439
United Kingdom	GBR247	O2379	Sovetskij
United Kingdom	GBR247	O2380	L'Govskij 1026
United Kingdom	GBR247	O2381	Skajstunes
United Kingdom	GBR247	O2382	Cernigovskij 83
United Kingdom	GBR247	O2383	Stendskij Zeltyj
United Kingdom	GBR247	O2384	Krasnodarskij 73
United Kingdom	GBR247	O2752	PI 266281

Used accessions to create rivet wheat mixtures in 2020:

PROVIN	ESTREG	PAIORI	NOMLOC	var.	Group	Variety
Almeria	Andalucia	ESP	Cañivano		Turgidum	Ou 1 OR BGE 013721
		ESP	Asolacambre		Turgidum	BGE000125
		EGY	Milagro		Turgidum	BGE000168
		ESP	Almendral		Turgidum	BGE002866
		ESP	Cascalbo		Turgidum	BGE002870
		ESP	Farto blanco		Turgidum	BGE002873
		ESP	Farto rubio		Turgidum	BGE002874
		ESP	Rubial de Liebana	var. dinurum	Dinurum	BGE002887
			Nodak	var. dreischianum	dreischianum	BGE012400
			R.a. 29	var. rubroalbum	rubroalbum	BGE012488
				var. rubroalbum	rubroalbum	BGE012489
Navarra	Navarra	ESP	Blanco de Corella		Turgidum	BGE012534
		PRT		var. lusitanicum	Lusitanicum	BGE012535
			T. abysinicum Vav. Rabat		Turgidum	BGE012536

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		PRT			Turgidum	BGE012537 (rec 2017)
		PRT	Novo		Turgidum	BGE012538
				var. dreischianum	dreischianum	BGE012539
				var. speciosum	speciosum	BGE012540
				var. speciosum	speciosum	BGE012541
				var. speciosum	speciosum	BGE012542
				var. speciosum	speciosum	BGE012543
				var. lusitanicum	Lusitanicum	BGE012544
		PRT	Rubiao		Turgidum	BGE012545
				var. martensii	convar martensii	BGE012546
					Turgidum	*OR BGE012547*
				var. nigrobarbatum	Nigrobarbatum	BGE012548
				var. martensii	convar martensii	BGE012549
				var. martensii	convar martensii	BGE012550
				var. martensii	convar martensii	BGE012551
				var. martensii	convar martensii	BGE012552
				var. dreischianum	dreischianum	BGE012554
Albacete	Castilla-La Mancha	ESP	Rubion de Higueruela		Turgidum	BGE012555
		PRT	Barba de lobo		Turgidum	BGE012556
Badajoz	Extremadura	ESP	Almendral		Turgidum	BGE012557
					Turgidum	BGE012558
			3334 D.m. Alonso Peña	var. rubroalbum	convar turgidum	BGE012559
			Elisabeth 201	var. rubroalbum	convar turgidum	BGE012560
				var. dinurum	Dinurum	BGE012561
				var. rubroatrum	var rubroatrum	BGE012562
				var. rubroalbum	convar turgidum	BGE012563
				var. rubroatrum	var rubroatrum	BGE012564
			D.m. 29	var. dinurum	Dinurum	BGE012565
				var. pseudocervinum	pseudocervicum	BGE012567
				var. plinianum	plinianum	BGE012568
				var. pseudocervinum	pseudocervicum	BGE012569
				var. plinianum	plinianum	BGE012570
				var. plinianum	plinianum	BGE012571
					Turgidum	BGE012572
				var. ramososalomonis	ramososalomonis	BGE012573
Navarra	Navarra	ESP	Blanco		Turgidum	BGE013074
Palencia	Castilla y Leon	ESP	Chile	var. lusitanicum	Lusitanicum	BGE013076
Baleares	Baleares	ESP	Argelino		Turgidum	BGE013078

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Lerida	Cataluña	ESP	Pisana de Vilanova		Turgidum	BGE013081
Navarra	Navarra	ESP	Cabezon de Estella		Turgidum	BGE013083
Asturias	Asturias	ESP	Blanco de Vegadeo		Turgidum	BGE013084
Murcia	Murcia	ESP	Caravaca 3		Turgidum	BGE013085
Badajoz	Extremadura	ESP	Entrelargo de Montijo		Turgidum	BGE013086
			Carleton		Turgidum	BGE013087
Gerona	Cataluña	ESP	Forment lampiño		Turgidum	BGE013088
La Rioja	La Rioja	ESP	Gigante lampiño de Najera	var. speciosum	speciosum	BGE013089
Almeria	Andalucia	ESP	Cañivano		Turgidum	BGE013090
Badajoz	Extremadura	ESP	Entrelargo de Montijo		Turgidum	BGE013091
Baleares	Baleares	ESP	Valencia blanco		Turgidum	BGE013092
Asturias	Asturias	ESP	Blanco velloso de Vegadeo		Turgidum	BGE013094
Murcia	Murcia	ESP	Pauleño de Mula		Turgidum	BGE013095
Badajoz	Extremadura	ESP	Almendral		Turgidum	BGE013097
Ciudad Real	Castilla-La Mancha	ESP	Torralba de Calatrava		Turgidum	BGE013098
Toledo	Castilla-La Mancha	ESP	Carriches		Turgidum	*OR BGE013099*
La Rioja	La Rioja	ESP	Gigante velloso de Najera		Turgidum	BGE013101
Asturias	Asturias	ESP			Turgidum	BGE013102
Gerona	Cataluña	ESP	Radondell encarnado de Llosas		Turgidum	BGE013104
				var. rubroalbum	convar turgidum	BGE013711
				var. rubroalbum	convar turgidum	BGE013712
Castellon	Valencia	ESP	Blancal de Nules	var. dreischianum	dreischianum	BGE013717
Asturias	Asturias	ESP	Boroñon de Salas		Turgidum	BGE013718
Badajoz	Extremadura	ESP	Rojo		Turgidum	BGE013719
		PRT	Rubiao de barba preta		Turgidum	OR BGE013720
Almeria	Andalucia	ESP	Cañivano		Turgidum	BGE013721
Malaga	Andalucia	ESP	Abena		Turgidum	BGE013722
		PRT	Alentejo		Turgidum	*OR BGE013723*
Asturias	Asturias	ESP	Serrentin de Salas		Turgidum	BGE013724
Murcia	Murcia	ESP	Jeja vellosa de Caravaca		Turgidum	BGE013725
Murcia	Murcia	ESP	Jeja de Moratalla		Turgidum	BGE013726
				var. dinurum	Dinurum	BGE013727
		PRT	Anafil	var. bucale	Bucale	BGE013728
Huelva	Andalucia	ESP	Blanquillo	var. lusitanicum	Lusitanicum	OR BGE013729

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Badajoz	Extremadura	ESP	Barcarrota		Turgidum	BGE013730
Huelva	Andalucia	ESP	Blanquillo raspinegro		Turgidum	*OR BGE013731*
				var. megalopolitanum	megalopolitanum	BGE013732
Caceres	Extremadura	ESP	Moro		Turgidum	*OR BGE013733*
				var. rubroalbum	convar turgidum	BGE013734
Asturias	Asturias	ESP	Puente Fierros		Turgidum	*OR BGE013735*
			Vermelho fino		Turgidum	*OR BGE013736*
		PRT	Caxudo de sete espigas		Turgidum	*OR BGE013737*
					Turgidum	*OR BGE013738*
					Turgidum	*OR BGE013739*
			Ran. de Bulgaria T.97-19		Turgidum	*OR BGE013740*
					Turgidum	BGE013741
		ESP	Almendral		Turgidum	BGE015009
		ESP	Farto rubio		Turgidum	BGE015014
		ESP	Molla		Turgidum	BGE015016
Baleares	Baleares	ESP	Blat barba blanc		Turgidum	BGE015395
Baleares	Baleares	ESP	Griego de Baleares		Turgidum	BGE018619
Tarragona	Cataluña	ESP	Pisana cañihueca	var. gentile	gentile	BGE018621
Tarragona	Cataluña	ESP	Pisana cañihueca	var. gentile	gentile	BGE018622
Cantabria	Cantabria	ESP	Mocho blanco		Turgidum	BGE018623
Albacete	Castilla-La Mancha	ESP	Mazachon de Balazote	var. herrerae	Herrerae	BGE018625
Gerona	Cataluña	ESP	Gros de Cerdaña		Turgidum	BGE018626
Barcelona	Cataluña	ESP	Forment		Turgidum	BGE018627
Barcelona	Cataluña	ESP	Forment		Turgidum	BGE018628
Asturias	Asturias	ESP	Asturias H2		Turgidum	BGE018641
Asturias	Asturias	ESP	Asturias H4		Turgidum	BGE018642
Asturias	Asturias	ESP	Asturias L1		Turgidum	BGE018643
Asturias	Asturias	ESP	Asturias L2		Turgidum	BGE018644
Asturias	Asturias	ESP	Asturias 4d		Turgidum	BGE018647
Zamora	Castilla y Leon	ESP	Redondillo de Fuentesauco	var. lusitanicum	Lusitanicum	BGE018653
Navarra	Navarra	ESP	Redondillo	var. lusitanicum	Lusitanicum	BGE018654
Cantabria	Cantabria	ESP	Rubial		Turgidum	*OR BGE018655*
Murcia	Murcia	ESP	Caravaca	var. herrerae	Herrerae	BGE018656
Cantabria	Cantabria	ESP	Rojo	var. speciosum	speciosum	*OR BGE018657*
Barcelona	Cataluña	ESP	Forment	var. rubroatrum	var rubroatrum	BGE018659
Granada	Andalucia	ESP	Aragones		Turgidum	BGE018675
Granada	Andalucia	ESP			Turgidum	BGE018676
Granada	Andalucia	ESP	Valenciano		Turgidum	BGE018677

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Granada	Andalucia	ESP	Valenciano		Turgidum	BGE018678
Granada	Andalucia	ESP			Turgidum	BGE018680
		ESP	Blanco de Corella		Turgidum	BGE019248
		ESP	Mamento argelino		Turgidum	BGE019256
Gerona	Cataluña	ESP	Gros de Cerdaña		Turgidum	*OR BGE019289*
Baleares	Baleares	ESP	Mamento de Mallorca		Turgidum	BGE019291
		ESP	Duro mocho	var. rubroatrum	var rubroatrum	BGE019293
Asturias	Asturias	ESP	Asturias B4		Turgidum	BGE019301
		ESP	Asturias L6		Turgidum	BGE019308
		ESP	H-93-11	var. rubroatrum	var rubroatrum	BGE020904
		ESP	H-93-7	var. rubroatrum	var rubroatrum	BGE020905
		ESP	H-93-3	var. rubroatrum	var rubroatrum	BGE020906
		ESP	H-93-36		Turgidum	BGE020907
		ESP			Turgidum	BGE020939
		ESP			Turgidum	BGE020940
		ESP		var. nigrobarbatum	Nigrobarbatum	BGE020941
		ESP		var. speciosissimum	speciosissimum	BGE020942
		ESP			Turgidum	BGE020943
		ESP			Turgidum	BGE020944
		ESP			Turgidum	BGE020945
Huelva	Andalucia	ESP	Trigo	var. salomonis	Salomonis	BGE020946
		ESP			Turgidum	BGE020947
Badajoz	Extremadura	ESP			Turgidum	BGE020948
		ESP			Turgidum	BGE020949
		ESP			Turgidum	BGE020950
		ESP			Turgidum	BGE020951
		ESP		var. candiens	candiens	BGE020952
		ESP		var. plinianum	plinianum	BGE020954
			Griego		Turgidum	BGE021772
Baleares	Baleares	ESP	Mamento argelino		Turgidum	BGE021773
Badajoz	Extremadura	ESP	Fino	var. salomonis	Salomonis	BGE021774
					Turgidum	BGE021816
					Turgidum	BGE021818
					Turgidum	BGE021819
					Turgidum	BGE021820
					Turgidum	BGE021823
					Turgidum	BGE021824
					Turgidum	BGE021825
					Turgidum	BGE021826

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					Turgidum	BGE021828
Asturias	Asturias	ESP	Asturias H1	var. dinurum	Dinurum	BGE030923
Asturias	Asturias	ESP	Asturias H7	var. dinurum	Dinurum	BGE030924
Asturias	Asturias	ESP	Asturias H8	var. dinurum	Dinurum	BGE030925
Asturias	Asturias	ESP	Asturias H9	var. dinurum	Dinurum	BGE030926
Asturias	Asturias	ESP	Asturias 4c	var. dinurum	Dinurum	BGE030927
Asturias	Asturias	ESP	Asturias Hd	var. dinurum	Dinurum	BGE030928
Navarra	Navarra	ESP	Negro de Beunza	var. iodurum	iodurum	BGE040242
Cuenca	Castilla-La Mancha	ESP	Albendea	var. martensii	var martensii	BGE040245

Creation of experimental population

Dynamic mixtures

Experimental design

The method consists in four different steps:
-choosing a species according to farmers' needs and doing bibliography research so that the varieties ordered in the gene banks respect the organic farming principles (in terms of selection processes).
-ordering the varieties in gene banks (around 200 per chosen species), to multiply and observe them during 2 to 4 years.
-creating the Diversified Oriented Populations based on one or several common phenotypic traits previously observed, requested by the farmers.
-following up the population with the farmers.

Evaluation

First evaluation phase of the methodology:
We present here the evaluation of the accessions when they are multiplied and described individually. The choice of evaluated traits differs from one species to another, so the main observed traits are summarised here. For each species, a bibliography is made to identify which traits are most relevant to observe.

Table 1. Summary of the traits for the observation of the spelt and oats accessions

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Pre harvest indicators		
Germination	Evaluating the percentage of germination on the plot	Score
Tillering bearing	Evaluation with visual examples	Score

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Tillering	Counting number of tillers on random plants	Number of tillers
Number of knots	Counting manually number of knots on random plants	Number of knots
Diseases	Overall presence of diseases observed, and percentage of sick plants on the plot appreciated by observer	Scale or percentage
Spiking time	Spiking date	Date
Flowering time	Flowering date	Date
Development stage	Using Zadocks growth scale	Zadocks stage
Post-harvest indicators		
Straw height	From base of the plant to base of the spike	Cm
Grain characteristics	Colour, heterogeneity	Qualitative indicators
Threshing difficulty	Qualitative assessment during threshing	Score

Table 2. Assessed crop productive performance

Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**
Thousand **kernel weight**	On the overall sample	g
Size of the grains	Based on a visual scale	Score

Second evaluation phase of the methodology:
For the second phase, the observation of the DOPs is made by the farmers, on less precise criteria.

Table 3. Assessed crop development and agro-ecological performance traits in the second phase

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Response of the farmer	Some farmers received populations but did not provide observations	Number of farmers providing observations
Sowing date		Date
Sowing conditions		Qualitative information
Germination	0:no germination; 5: very good germination	Score
Tillering	0: no tillering, 5: strong tillering	Score
Soil cover	0: not covering; 5: very covering	Score
Diseases	0: no diseases; 5: completely diseased	Score
Vigour	0: not vigorous, 5: very vigorous	Score
Lodging	0: no lodging, 5: completely lodged	Score
Correspondence of the population with the requested criteria	General appreciation of the farmers	Qualitative information
Diversity level	General appreciation of the farmers	Qualitative information

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Statistical methods

No specific statistical analyses were relevant to use in this methodology experimentation: the idea of this experimentation was to test the feasibility of the DOP methodology for diversity mobilization.
For the first part of the methodology, the purpose of the observations was to be able to create populations responding to farmers' criteria.
For the second part of the methodology (distribution and population follow up), the observations were mostly qualitative (discussions with the farmers).
The rivet wheat distribution was the first step of the implementation of the methodology, that will continue after the LIVESEED project, for spelt and oat. Statistical analyses will be conducted for these two species.

How resilience was improved

From a high number of accessions, the populations created by mixtures are very diversified to answer to the environmental constraints, whilst they are creating focusing on few common traits shared by diversified accessions.

Participatory/multi-actor approach

The method is based on a participatory approach as it was designed to respond to a problematic raised both by farmers and researchers, in several different groups: the need to create populations with a more important internal diversity. Moreover, the choice of the species is also done according to farmers' needs, collected during the different meetings. The farmers also chose the criteria to design their own populations, adapted to their terroir, practices and needs. Finally, the people receiving the populations are reporting information and observations on the populations. Every person receiving populations is also asked to give their opinion on the method so that it can be iteratively adapted.

Results – Rivet Wheat

Over the period of the project, around 500 accessions of rivet wheat, spelt and oats were observed and described, to be distributed to farmers, and bring this diversity back to the fields.
Over the two years of distribution of rivet wheat populations, 35 farmers participated to the experimentation:

During the first rivet wheat distribution period, 24 farmers received 44 samples. 29 different populations were created (some farmers received the same populations). 157 different accessions were assembled to create those DOPs, out of the 172 multiplied accessions, which means that only 15 accessions were not used following the farmers' requests. There were on average 30 varieties per population. Finally, farmers used 24 different criteria to compose their mixes, which shows a great diversity of needs.
During the second distribution period, 11 farmers received populations, based on the same principles as the first year.

Out of the 24 people who received the DOPs in 2018, 14 returned observations to us. For them, there were only two populations that did not fit completely with the criteria they requested.
They were globally satisfied with the method, but some (3 persons) pointed out that the given number of seeds was too small and that there was a risk of losing the populations.

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After the first year of distribution, the questionnaire was improved to get better returns from the farmers: most of the information were provided by phone, following an interview guide.

Discussion

Those results show that there is an important interest from the farmers for this method, as they were a total of 35 farmers on two years to respond to our proposition.
It also appears that the populations that were sent correspond to the criteria the farmers requested.

In 2020, the diversity level of the populations was assessed qualitatively by asking the farmers to evaluate it according to their own scale. It is too early to quantify the adaptability of the populations, but the farmers note a high level of diversity in the populations as the chosen criteria do not reduce diversity for other phenotypical traits.

It could be interesting, at least on some populations, to be able to quantify this diversity level, and to follow it on the long term. Indeed, it would be interesting to see if the populations are diversified enough to be selected by the farmers on specific criteria, while maintaining a high level of diversity on other characteristics.

The main bottleneck that we had while implementing this method for the first two years was to gather the responses of every person who received some DOPs. Therefore, facilitators and animators are needed, to stay in contact with the farmers, and to maintain a collective dynamic among them.

Outcomes and conclusions

Outcomes:
-35 farmers tested populations. Around 30 different populations were created from mixtures of several accessions from a pool of nearly 200 accessions.
-Farmers are interested in this type of populations because they may choose relevant criteria answering to their conditions and market.
-According to farmers, in their fields the main traits of the populations correspond to the criteria they requested among the genetic resources.

The method needs to be carried on assessing its interest among farmers and to see if the hypothesis is valid or not.
The aim of the method is to mobilize as much diversity as possible, so it is needed to assess this diversity level.
There are over thousands of accessions in the gene banks that could be used to build DOP populations. Mobilizing them to make them available ex-situ is then an important work which takes time and energy. It would be interesting to quantify how much time was spent for this work to build a sustainable economic system to continue this action on a larger scale.

We also need to find ways to get more response from the people who receive the populations and to continue the multiplication for spelt and oats. Next year, spelt will be available to distribution. We also want to start alfalfa multiplication.

Main lessons learned

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-Need to keep on improving the populations follow up: farmers' implication requires time.
-Send quantities were too low for some farmers.
-Farmers express a need for diversification.

How the information can be transferred to the other situations?

This mixing methodology can be used very easily for on-farm breeding.

Publications

Eucarpia presentation on DOP populations: "Mobilizing diversity for minor cereals in western France", 2021

- Serpolay, E.; Chable, V. (2020) Shaping Diversity for On-farm Organic Plant Breeding of Wheat (and Other Cereals) in France. In Hubbard, K. (editor). 2020. Organic Seed Growers Conference Proceedings. February 12 - 15, 2020, Corvallis, OR. Organic Seed Alliance, Port Townsend, WA: 39 - 44.

-Article (in French): http://itab.asso.fr/publications/aa-biodiv-oubliee-poulard.php

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LBI - Mixtures of spring wheat varieties and mixtures and populations **(Trial #5)**

Lead: LBI - The Netherlands.
Floor van Malland - f.vanMalland@louisbolk.nl, Abco de Buck - a.debuck@louisbolk.nl

Abstract

Mixtures of several spring wheat varieties and populations were tested and compared with pure varieties in an organic field (marine clay soil) trial in 2018, 2019 and 2020. The measurements consisted of germination, yield, protein content, Zeleny, falling number, development of diseases, plant height, ripening and plant development. The yield of the varieties sown in mixed conditions was comparable with the yield of the pure stand of these varieties. Wheat populations show a slightly lower yield compared to commercial wheat varieties. The disease pressure was very low in these years, hence a higher resilience to diseased of mixing wheat varieties was not confirmed in this research.

Introduction

Objectives

The 'host' farmer for the trial grows a spring wheat population that evolves/adapts over generations to her farm. Together with this example, the trial shows various options to farmers and advisors for more diversity on the field and in the product of baking wheat, e.g., at the annual field days and winter workshops.

The main aim of the trial is to identify traits important for mixed stands of different spring wheat cultivars ('conventional' varieties and populations). Is it possible to predict which (type of) cultivars are suitable for use in mixed stands? Two categories of cultivars were compared: varieties that were developed by different (conventional) breeders, and varieties and populations that were developed by one (organic) breeder.

Material and Methods

Location

The trial fields (2018-2020) were located within a commercial field with wheat at an organic farm in the central polder area (Kraggenburg, The Netherlands) on a limey, light marine clay soil.

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Figure 1: Harvest of wheat trial 2019 (photo taken by Floor van Malland 22-08-2019)

Genetic resources

Wheat varieties, wheat populations and mixtures:

- *Lennox (Agrifirm, The Netherlands)*
- *Lavett (Agrifirm, The Netherlands)*
- *Arabella (Danko, Poland)*
- *Harenda (Limagrain)*
- *Heliaro (Dottenfelderhof, Germany)*
- *Saludo (HSWS 66-08, Dottenfelderhof)*
- *Zino (HSWS 126-11, Dottenfelderhof)*
- *HSWS 2013-616-20 (Dottenfelderhof)*
- *WPB Skye (Wiersum)*
- *WPB Scotch (Wiersum)*
- *population HS-2-08 (original)*
- *population Convinto C (original)*
- *population WSER I-2015*
- *population WSER III-2015*
- *population WSER IV-2015*
- *population WS ER II-08 D*
- *population WS ER II-08 NL*
- *Mixture Lavett & Lennox*
- *Mixture Lavett & Arabella*
- *Mixture Lavett & Harenda*
- *Mixture Lavett & Saludo*
- *Mixture Lennox & Arabella*
- *Mixture Lennox & Harenda*
- *Mixture Lennox & Saludo*
- *Mixture Arabella & Harenda*

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- *Mixture Arabella & Saludo*
- *Mixture Harenda & Saludo*
- *Mixture Heliaro & Saludo*
- *Mixture Heliaro & Zino*
- *Mixture Heliaro & HSWS 2013-616-20*
- *Mixture Saludo & Zino*
- *Mixture Saludo & HSWS 2013-616-20*
- *Mixture ZIno & HSWS 2013-616-20*
- *Mixture Heliaro, Saludo, ZIno & HSWS 2013-616-20*
- *Mixture Lavett, Lennox, Arabella & Harenda*

Creation of experimental population

Mixtures of varieties, populations

Experimental design

Year	Sowing	Harvest
2018	26-03-2018	30-07-2018
2019	02-04-2019	22-08-2019
2020	27-03-2020	19-08-2020

The trials were sown within a production field with a wheat population in 3 repetitions (plot size gross 15m2, net 12.75m2).

Sowing density wheat 300 seeds/m2.

Evaluation

List of the traits collected for the wheat trials concerning crop development and agro-ecological performance, crop productive performance and crop quality performance.

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Germination	Counting plants	%
Disease's wheat	Yellow rust (*Puccinia striiformis*), Brown rust (*Puccinia recondita*) (1 = dead heavy infected to 9 = not infected) *Fusarium ssp.* and *Septoria tritici* (counting infected plants)	Scoring
Plant development wheat	Development of spikes (1 = stretching of flag leaf, 2 = spike developing in flag leaf, 3 = spike in top of flag leaf, 4 = spike emerging, 5 = spike totally emerged)	Scoring
Plant height	Measuring height	cm
Ripening plants	Ripening, state of yellowing (Yellowing of total plant 1=green, 5=yellow)	Scoring
Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**

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Yield	Harvest per plot, dried (mixtures with lupin are separated) and cleaned	t/ha
Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		
Trait	**How it has been assessed**	**Type of data available**
Protein content	Laboratory Gent University	%
Falling number wheat	Laboratory Gent University	Sec
Zeleny wheat	Laboratory Gent University	Ml

Table 1. Crop development and agro-ecological performance, crop productive performance and crop quality performance traits

Statistical methods

Statistical analyses: ANOVA randomised block design, linear regression

Participatory/multi-actor approach

Organisation of workshops and field days to inform and discuss results with growers, researchers, breeders, government officials etc.

Results and discussion

Main results

Table 2 Shows the results of the total yield of the different wheat treatments per year and the average over the three years.

Table 2: Total yield of the different wheat treatments in 2018, 2019 and 2020 and the significant differences between the treatments (a-i). The treatments coloured in orange are wheat populations, in yellow are pure stand varieties, white are mixtures of two varieties and purple are mixtures of four wheat varieties.

	Average yield (t/ha)				
Treatment	**2018**	**2019**	**2020**	**mean**	**Significance**
Population Convinto C/x/x/x	3,76	5,89	3,21	4,28	a
population WS ER II-08 NL/x/x/x	4,08	5,56	3,32	4,31	ab
Lavett/x/x/x	4,00	5,62	3,44	4,35	abc
Population WSER III-2015/x/x/x	3,76	6,10	3,30	4,38	abcd
Population HS-2-08/x/x/x	4,12	5,66	3,39	4,38	abcd
Heliaro/Saludo/x/x	3,80	6,10	3,29	4,39	abcd
Population WSER IV-2015/x/x/x	3,96	6,00	3,46	4,46	abcde
Heliaro/HSWS 2013-616-20/x/x	3,99	6,01	3,43	4,47	abcdef
Saludo/x/x/x	4,07	5,87	3,50	4,47	abcdef
population WS ER II-08 D/x/x/x	4,05	5,84	3,56	4,47	abcdef
Saludo/HSWS 2013-616-20/x/x	3,91	6,14	3,41	4,48	abcdef
Lavett/Harenda/x/x	4,00	6,09	3,44	4,50	abcdef
Heliaro/Zino/x/x	4,04	6,09	3,42	4,50	abcdefg
Arabella/Saludo/x/x	3,82	6,06	3,70	4,52	abcdefg
Heliaro/x/x/x	4,07	6,14	3,37	4,52	abcdefg
Saludo/Zino/x/x	4,16	6,04	3,40	4,52	abcdefg
Lavett/Arabella/x/x	4,09	5,92	3,62	4,53	abcdefg

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Heliaro/Saludo/Zino/HSWS 2013-616-20	3,86	6,25	3,64	4,57	abcdefgh
Lennox/Saludo/x/x	3,99	6,28	3,50	4,58	abcdefgh
Zino/HSWS 2013-616-20/x/x	3,93	6,15	3,72	4,59	abcdefgh
Zino/x/x/x	4,12	6,17	3,52	4,59	abcdefgh
Lavett/Saludo/x/x	4,16	6,03	3,67	4,61	abcdefgh
population WSER I-2015/x/x/x	3,85	6,28	3,77	4,62	abcdefgh
Harenda/Saludo/x/x	4,45	5,95	3,59	4,65	bcdefgh
Lennox/Harenda/x/x	4,36	6,10	3,62	4,68	bcdefgh
Lennox/Arabella/x/x	4,08	6,65	3,42	4,70	cdefgh
Lennox/x/x/x	4,48	6,30	3,41	4,72	defgh
Arabella/Harenda/x/x	4,42	6,26	3,55	4,73	defgh
Lavett/Lennox/x/x	4,18	6,58	3,48	4,74	defgh
HSWS 2013-616-20/x/x/x	4,32	6,62	3,31	4,74	defgh
Arabella/x/x/x	4,24	6,56	3,58	4,78	efgh
Harenda/x/x/x	4,82	6,18	3,42	4,80	efgh
Scotch/x/x/x	4,08	6,68	3,80	4,84	fgh
Quintus/x/x/x	4,42	6,45	3,78	4,88	gh
Lavett/Lennox/Arabella/Harenda	4,34	6,59	3,86	4,92	h
Skye/x/x/x	4,65	7,26	4,00	5,29	I

Figure 2 shows the protein content of the different wheat treatments in 2020. The protein analyses in 2018-2019 were partially done at another laboratory, using another protocol, resulting in different outcomes. No significant differences were found between the different treatments in 2020 (*P=0,882*). Also, no significant differences were found when the pure stands were analysed separately.

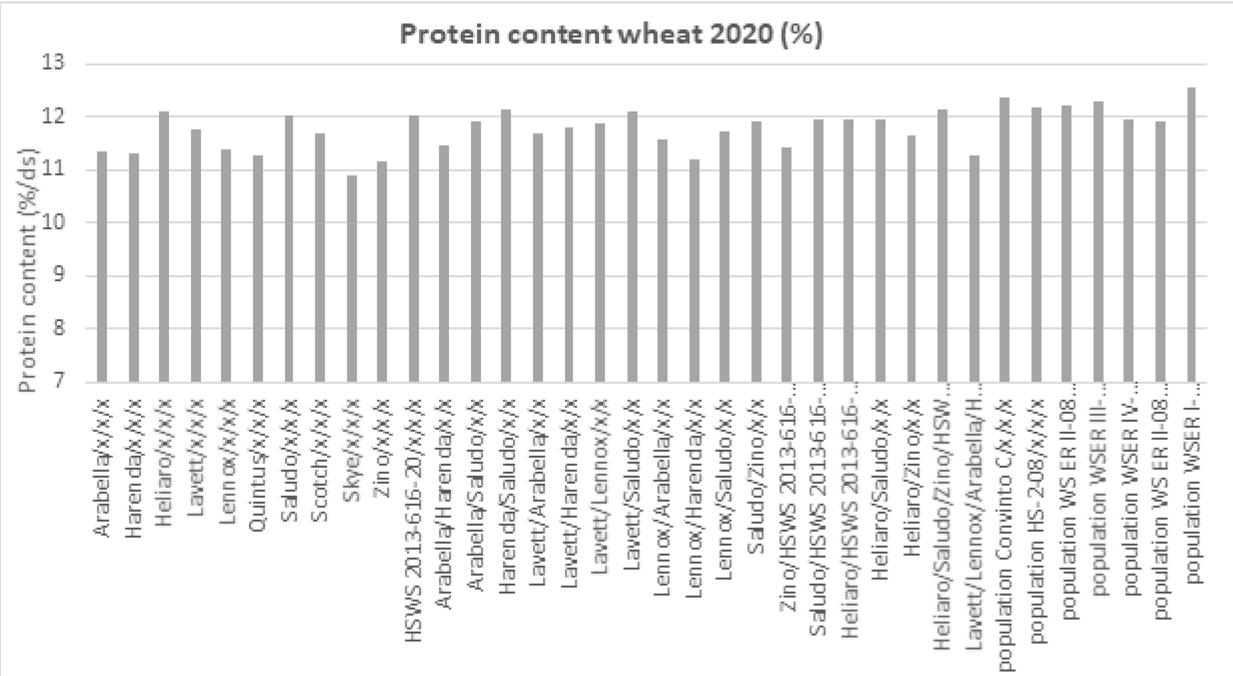

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Figure 2. Protein content of the different wheat treatments in 2020. No significant differences were found between the treatments. Notification:./x/x/x = pure cultivar stand, ././x/x = mix of two cultivars, ./././x mix of three and ././/. mix of four cultivars.

Figure 3 shows the correlation between yield (t/ha) and protein content of the wheat (P<0,001) in 2020.

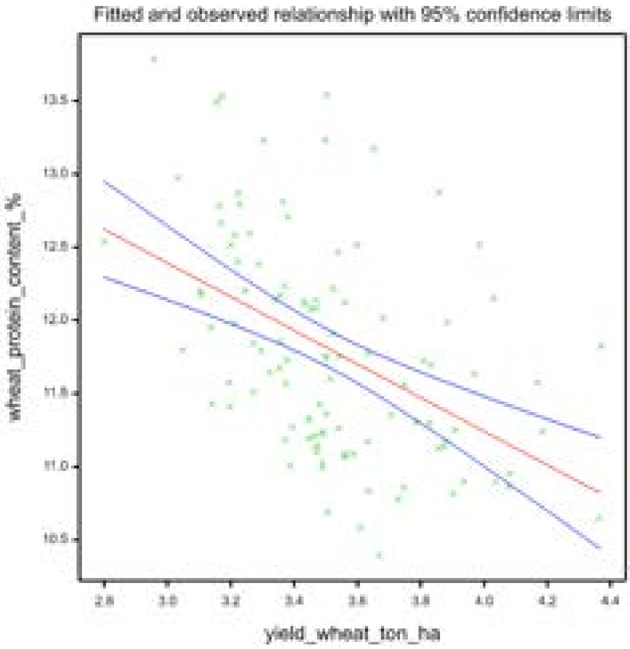

Figure 3: Correlation between yield and protein content determined of the wheat trial 2020 (P<0,001, linear regression)

Conclusions, discussion, and next steps

- Most of the wheat populations showed a lower yield compared to the commercial varieties in pure stand.
- Especially the mixture with 4 commercial varieties (Lavett, Lennox, Arabella and Harenda) stands out compared to the pure stand varieties for total yield. But the yield of this mixed treatment is only significant higher compared to the variety 'Lavett' in pure stand.
- The variety 'Skye' showed the significant highest yield.
- Mixtures of 2 varieties showed in almost all cases no significant differences with the varieties in pure stand for yield. Only for 'Lavett' some differences were found. This variety has already been phased out in the Netherlands because of the low performance.
- Varieties developed by one (organic) breeder (Dottenfelderhof, Germany) showed no differences in yield when comparing to each other; commercial varieties of different breeders showed more variance in yield.
- No significant differences in protein content were found between the treatments in 2020. Although a correlation was found between yield and protein content.
- Disease pressure was too low during the three years to draw conclusion about differences between the treatments.

Spring wheat is an important crop in a crop rotation on organic farms in the Netherlands. Nowadays there are only 1 or 2 varieties on the market, which is by far insufficient to make a good variety choice

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for the specific farm conditions. Especially, to choose the right cultivar for baking a tasteful, artisan, organic bread is a challenge. At the annual field days and winter workshops farmers, artisan bakers and other representatives of (local) value chains could experience options for a more diverse spring wheat production.

Mixed cultivation of wheat varieties has no effect on yield. Whether cultivar mixtures lead to a higher resilience is not confirmed in this trial, due to the very low disease pressure. The disease pressure was too low during the years of the trial to draw conclusion if mixing varieties offers a better resistance against several above and or below ground diseases.
Wheat populations show slightly lower yields compared to commercial varieties. Also, here the question, if populations have a higher resilience, remains unanswered.

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AREI: Spring barley mixtures and populations (Trial #6)

Lead: AREI
Linda Legzdiņa, Indra Ločmele

Abstract

Spring barley mixtures, three types of populations and doubled haploid (DH) lines were evaluated in field trials during 2017-2018 (continued from 2015-2016, national project) in two locations under organic (7 environments) and conventional (7 environments) management. The objectives were: 1. Compare mixtures and components in pure stand, to conclude the effect of specific component traits on mixture performance; 2. Compare performance of populations with different diversity level and compare populations to check varieties to recommend to farmers; 3. Compare DH lines to their initial advanced lines, if there was a difference in yield stability.

Mixtures can provide advantages over growing in pure stand in respect to incidence of leaf diseases and to lesser extent in respect to yield and weed suppression ability. It can be possible to obtain stable and good yielding mixtures by mixing components with distinctive disease resistance, yield potential and adaptability but it is not true for all cases. Stable yielding mixtures can be produced by mixing components with broad adaptability and adaptability to lower yielding environments.

CCPs were stable yielding over various environments can provide comparable yield results to the check varieties under organic farming and stress conditions. Diversity of plant traits of organically cultivated sub-populations was slightly larger than that of conventionally cultivated ones in case of complex and CCP population, but not for simple population cultivated under the respective farming system for longer period. No effect of complete homogeneity of DH lines on adaptability was found, however, they yielded on average lower than the initial advanced lines.

Introduction

Hypothesis and research question

The trial started in a national project during 2013-2016 and was continued in LIVESEED, partially as a work of a PhD work for student (I. Ločmele) who was the main responsible person for data obtaining and statistical analysis.

The results are expected to show the advantages of mixtures of genotypes with particular traits in comparison to pure lines; demonstrate if populations including CCPs can be comparable to check varieties; and find if reduced diversity in DH lines affects yield stability.

Barley traits providing good combination results in mixtures were expected to be listed and CCPs perspective for growing in farms will be described.

Objectives

The objectives were the following:

Crops	Before	In LIVESEED	Locations	Objectives
Spring barley mixtures	2015-16	2017-18	7 Organic vs 7 conv.	1. Compare mixtures and components in pure stand, to conclude the effect of specific

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				component traits on mixture performance;
Three types of spring barley populations	2015-17	2017-19	7 Organic vs 7 conv.	2. Compare performance of populations with different diversity level and compare populations to check varieties to recommend to farmers;
Spring barley DH	2016-18	2017-20	6 Organic vs 5 conv.	3. Compare DH lines to their initial advanced lines, if there was a difference in yield stability

Material and Methods

Location

Priekuli organic (PR_O), Priekuli conventional (PR_C), Stende organic (ST_O), Stende conventional (ST_C). In these 4 locations for 4 years, data of 14 environments (location x year combinations) in total available.

Genetic resources

49 entrees:
• 8 genotype mixtures (2-5 components)
• 16 mixture components in pure stand
• 15 populations (4 simple cross, 6 complex cross, 5 CCP's (2 newly created CCPs in 11 and 7 environments, respectively))
• 4 doubled haploid lines + 2 initial lines
• 4 check varieties

List of accessions in data file

Experimental design

Lattice design, 4 replicates, 12.3 m2 plots (location PR) and 5 m2 plots (location ST). Seed from previous year trial in location PR organic and conventional sites was used; 400 germinable seeds per m2 (450 for hulless barley entrees); mixtures mixed every year in equal proportions considering germination and TGW.

Evaluation

List of the traits collected in the Table 1-3.

Table 1. List of the traits collected for spring barley concerning crop development and agro-ecological performance, crop productive performance and crop quality performance

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Early vigor at tillering (E.VIG_til)	average early vigour estimated on plot level according to crop biomass produced at GS 20-25 rated from 1=low to 9=very high	scoring

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Plant type at tillering (PL. T_til)	plant type estimated on plot level at GS 25-29, 1=very erect, 9=very prostrate	scoring
Canopy height at beginning of stem elongation (CAN_st. el_cm)	average canopy height on plot level estimated based on 3 measures per plot from soil till top of plants in cm at growth stage beginning of stem elongation (GS 31-32)	cm
Days from sowing to start of heading (HEAD_days)	date when 50% spikes are starting to emerge (GS 51-53) estimated on plot bases in 1 replication and number of days from sowing to heading calculated	days
Days from sowing to ripening (RIP_days)	date when grain of 50% spikes is hard (GS 91-92) estimated on plot bases in 1 replication and number of days from sowing to ripening calculated	days
Crop ground cover at tillering (GR.COV_til_%)	average crop ground cover estimated in % on plot level at GS 25-29	%
Crop ground cover at stem elongation (GR.COV_st.el_%)	average crop ground cover estimated in % on plot level at GS 29-31	%
Weed suppression ability at stem elongation (WSA_st.el_%)	average weed ground cover estimated in % on plot level at crop GS 31-39. Weed suppression ability for each crop genotype (S_{var}) was calculated as the difference between weed growth in plots for each genotype (W_{var}) and the maximum weed growth (W_{max}) from adjacent uncropped plot and expressed as a percentage: $S_{var} = ((W_{max}-W_{var})/W_{max}) * 100$ [Hoad et al., 2008]	%
Weed suppression ability at flowering (WSA_fl_%)	average weed ground cover estimated in % on plot level at crop GS 59-65.	%
Weed suppression ability at ripening (WSA_ri_%)	average weed ground cover estimated in % on plot level at crop GS 87-92.	%
Productive tillering coefficient (TILL_coef)	Number of productive tillers and number of plants counted in 0.1 m^2 area in the middle of each plot after harvest (productive tillers are considered the ones with round circle clearly visible from the top; the number of plants is determined after pulling them out of the ground; productive tillering coefficient calculated by dividing number of tillers with number of plants.	coefficient
Loose smut infected plants (SMUT)	number of plants infected with loose smut counted in the plot of one replication after flowering	number
Net blotch estimation (NETB)	average severity of net blotch estimated on plot level rated from 0 = no symptoms till 9= leaves are completely infected = dead tissue according to [Handbook Cereal variety testing for organic and low input agriculture, 2006] assessed 1-5 times, starting when first disease symptoms were visible and after 7-10 days	score
Netblotch AUDPC value (NETB_AUDPC)	AUDPC value calculated from all NETB_scores in case of more than 1 scoring [Tratwal et al., 2007]	value
Powdery mildew estimation (MILD)	average severity of powdery mildew estimated on plot level rated from 0 = no symptoms till 9= leaves are completely infected = dead tissue according to [Handbook Cereal variety testing for organic and	score

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	low input agriculture, 2006]; assessed 1-3 times, starting when first disease symptoms were visible and after 7-10 days	
Powdery mildew AUDPC value (MILD_AUDPC)	AUDPC value calculated from all MILD_scores in case of more than 1 scoring [Tratwal et al., 2007]	value
Lodging at ripening (LODG_ri)	average lodging on the plot bases at the same time when determining ripening date rated from 1= all plants fully lodged till 9=no lodging visible	score

Table 2. Assessed crop productive performance traits

Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**
Grain yield (YIELD_t/ha)	grain yield on the plot bases harvested, dried, and weighed in kg with 3 decimals after storage in the same room; 1 kg sample cleaned by sample cleaner MLN using 1.8 mm sieve and weighed after cleaning; clean yield per plot calculated; yield in t/ha by 14% moisture content calculated using average moisture measurement from 10 randomly taken samples	t/ha

Table 3. Assessed crop quality performance traits

Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		
Trait	**How it has been assessed**	**Type of data available**
Crude protein content in grain (PROT_%)	determined by INFRATEC 1241 grain analyser in each replicate (for populations and check varieties) or in pooled samples from the 4 replicates (for mixtures, DHs)	% in DM
Starch content in grain (STARCH_%)	determined by INFRATEC 1241 grain analyser in each replicate (for populations and check varieties) or in pooled samples from the 4 replicates (for mixtures, DHs)	% in DM
Beta-glucan content in grain (B-GLU_%)	determined by INFRATEC 1241 grain analyser in each replicate (for populations and check varieties) or in pooled samples from the 4 replicates (for mixtures, DHs)	% in DM
Grain volume weight (VOLW_g/l)	determined by INFRATEC 1241 grain analyser in each replicate (for populations and check varieties) or in pooled samples from the 4 replicates (for mixtures, DHs)	g/l
Thousand grain weight (TGW_g)	determined by counting 2 samples x 500 grains (and repeating counting if the difference is larger then 0.5 g) and weighing in grams with 2 decimals in each replicate (for populations and check varieties) or in	g

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	pooled samples from the 4 replicates (for mixtures, DHs)	

Statistical methods

ANOVA was applied to estimate effects of year, genotype and interaction, yield stability calculated according to regression analysis and ranking using data from 4 growing seasons (14 environments), mixing effect for mixtures according to Kiær et al., (2012), the Shannon-Weaver diversity index H' for selected populations according to Bertholdsson et al. (2016). Quantitative traits of populations were compared using t-test. GME and SME is foreseen to be calculated for mixtures in cooperation with Emma Forst and some other specific analysis in cooperation with IPC (Pedro Mendes Moreira).

Results

Average yield and yield stability indicators of hulled spring barley mixtures (M) and populations

Mixtures. *Yield stability* over 14 environments (7 organic, 7 conventional) for 4 years: all mixtures had average yield above the mean yield of the trial; broad adaptability was found for 3 mixtures (one of them significantly surpassed the mean); 4 mixtures showed adaptability to favourable environments and 1 mixture – adaptability to unfavourable environments.

Mixtures M1 and M2 were created by combining components with different adaptability to environments (unfavourable/favourable/broad); they resulted in mixtures with similar yield level (to each other) but different adaptability. M1 provided adaptability to favourable environments (ME_{aver}=-1.8% under O conditions, ME_{aver}=4.8% under C conditions) and M2 - broad adaptability (ME_{aver}=3.2% and -1.7% for O and C, respectively) (Table 1; Figure 2 supplementary). Comparatively best yield results were obtained for M7, which was created for control of leaf diseases. It had significantly higher average yield than the average of all genotypes over the whole experiment, broad adaptability, and the highest mixing effect (ME_{aver}=10.5). The components of this mixture were characterized with different adaptability and yield level – one of them had yield above the average, while second - significantly below the average. The yield gain in comparison to the average of component pure stands for M7 was highest of all tested mixtures under both O and C conditions, respectively 6% and 9% (Table 4).

Table 4. Average yield and yield stability indicators of hulled spring barley mixtures (M) and populations (SP-simple, CP- complex cross, CCP-composite cross populations) over 14 environments, 2015-2018

Genotype	Average yield (t ha^{-1})	Coefficient of regression (b)	Number of rankings					
			Organic (n=7)			Conventional (n=7)		
			I	II	III	I	II	III
M7	4.76*	1.00	6	1		5	2	
M3	4.58	1.12**	2	4	1	4	2	1
M8	4.58	1.01	5	1	1	2	4	1
CCP1	4.52	0.93	5	2	-	2	5	–
Rubiola	4.51	1.22**	3	2	2	4	1	2
M1	4.50	1.16**		5	2	4	3	
M2	4.50	1.02	4	1	2	1	4	2

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M4	4.46	1.08**	2	4	1	2	4	1
M5	4.44	1.08**	2	1	4	3	3	1
CP4	4.37	0.91	2	5	-	2	4	1
M6	4.37	0.81**	5		2		2	5
CP1	4.34	1.19**	-	4	3	3	3	1
CP5	4.20	1.07	-	3	4	1	4	2
CCP3	4.17	1.01	-	2	5	-	5	2
Abava	4.17	0.84**	2	3	2	1	2	4
CP2	4.15	0.99	1	2	4	-	2	5
Rasa	4.11	1.01	-	2	5	1	3	3
SP3	4.08*	0.99	-	2	5	1	2	4
SP4	4.07*	1.01	-	3	4	1	-	6
SP2	3.98*	0.89**	-	2	5	-	-	7
SP1	3.82*	0.89**	-	1	6	-	-	7
CP3	3.81*	1.01	-	-	7	-	1	6
CCP4[&]	4.38	0.94	3	1	2	1	4	–
CCP5[&&]	4.02	0.91	3	1	–	–	2	1

*Significantly distinctive from average yield over 14 environments 4.34 t/ha ($p<0.05$) ($LSD_{0.05}=0.25$); **significantly different from 1 ($p<0.05$); b – coefficient of regression; I, II, III – ranked in the upper, middle, and lower third, respectively; [&] results over 11 environments; results over 7 environments. Hulless barley genotypes are not included in this table

To improve *weed suppression ability*, mixtures M3, M4 and M5 were created by combining different plant traits, however, they did not compete with weeds significantly better than component mean value in pure stand. For mixture M3 containing genotypes with different plant growth habit (erect, intermediate and planophyle) there was a trend to better ground cover at the beginning of plant development and ability to suppress weeds (ME_{aver}=5.3% and 1.6%) (Figure 3, Table 5 supplementary). For mixture M5, although the ground cover was lower than the average of the components, in most of cases S_{var} indicator was insignificantly higher (average ME_{aver}=-10% for ground cover and 4.0% for weed suppression). Comparatively best results among the tested mixtures were obtained for two-component mixture M7 (ME_{aver}=12.9% and 1.7%) and three-component mixture M8 (ME_{aver}=12.5% and 5.2%). There were no significant differences between M7 and M8 components in crop ground cover and weed suppression ability. Other traits of components that are important to provide competitiveness against weeds were assessed and should be further analysed to better explain the mixing effects.

To evaluate possibilities for *leaf disease (mainly net blotch)* control by combining genotypes with different susceptibility/resistance mixtures M6, M7 and M8 were created. According to the results of this study, components of M6 did not correspond to the characterisation given before and the mixing effect for net blotch was the worst out of all mixtures. In comparison to the average of components, only for M8 tendency to lover infection was observed (Table 6 supplementary). In only one out of six cases (3 years x 2 environments = 6 cases) M6 infection with net blotch was significantly lower than that of the most susceptible component, whereas for M7 and M8 it was in all cases. Average mixing efficiency (both O and C) in comparison to average of components for M6 ME_{aver}=12.4%, for M7 ME_{aver}=-3.3% and for M8 ME_{aver}=-11.6%. (Figure 4 suppl.). Negative ME shows reduction of disease severity. Comparatively best results were obtained for M1, M2 and M3 – in all cases there was a tendency to less infection than the average of components in pure stand (even in cases with no significant differences between the components), average mixture efficiency –14.8%, -8.9% and -14.3% respectively.

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Therefore, further studies should be carried out, to determine what traits may affect the development of net blotch. Results of M3 may be explained by combining genotypes with different plant growth habit, thus forming such plant structure that delayed development of the disease. The fact, that other plant traits besides susceptibility/resistance could play an important role in controlling spread of disease in mixture, probably also shows the result of M5 under O conditions, where were significant differences between the components, but in two out of three cases infection level increased (in one case significantly).

Infection with *powdery mildew* under O conditions observed only in one of four years but under C conditions in two years in small amount, therefore convincing conclusions cannot be made, but under C conditions mixtures tended to infect less than average of components in pure stand.

Populations. Broad *adaptability* and average yield over environments above the mean of the trial was found for 3 hulled barley CCPs. All the tested CCPs were more stable than two out of tree check varieties (Table 1). Average yield of simple and complex populations was below the mean in most of the cases. All CCPs yielded significantly higher than two checks with lower yield under drought stress in the site PR_O in 2018 (Figure 1).

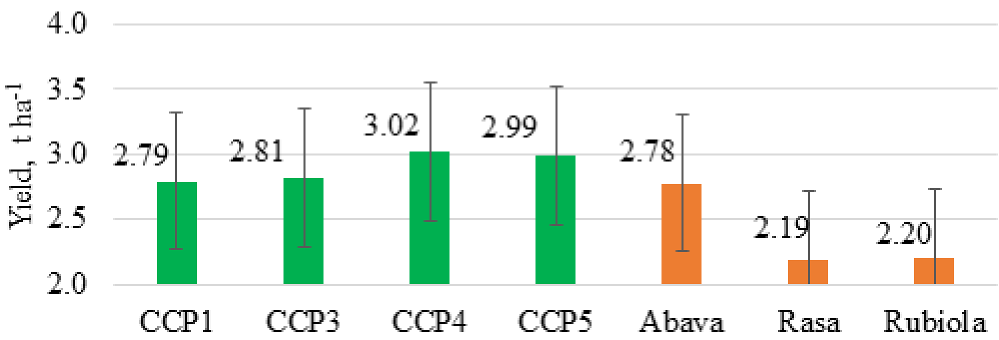

Figure 1. Yield of CCPs and check varieties under Pr_O environment in 2018 (under drought stress)

Significantly lower *infection with net blotch* compared to two more susceptible check varieties was observed for all populations in most of the cases. One simple and one complex population were infected significantly higher with *powdery mildew* than all checks, but obtained results for other populations varied, and no trend to better resistance was observed. No convincing advantages regarding *weed suppression ability* of populations over the checks were found.

While comparing two hulless barley (HB) populations (complex cross and CCP) to check variety, no advantages in respect of yield were found (CCP insignificantly over-yielded check in one environment only). Advantages were grain weight and protein content. For CCP, which was created with the aim for improved beta-glucan content, it was higher, however, the mean difference was only 0.4%.

Evaluating *quantitative plant traits* in selected parallel populations a trend has been observed showing that the development under O and C conditions caused differences. Also, significant differences were found for some traits in some cases (Table 9, 10, 11, 12 suppl.), the results are not convincing and should be further investigated. In case of simple sub-populations opposite trends were found for plant height and spike length while tested organically and conventionally which can indicate to specific adaptability to the cultivation environment. For complex population and CCP a slightly greater diversity of traits was observed in parallel population grown under O conditions if compared to conventional sub-populations (Table 5).

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Table 5. Average Shannon-Weaver diversity index of parallel sub-populations of three populations, average of O and C testing environments, 2016 and 2018

Population	F	Number of cultivation years**	Plant length	Length of the main spike	Number of grains in the main spike	Coefficient of tillering
BZ12C* (simple)	15	10	2.0	1.5	2.6	1.2
BZ12O* (simple)			1.8	1.4	2.5	1.3
1018-12C (complex)	8	4	1.7	1.4	2.4	1.1
1018-12O (complex)			2.0	1.5	2.5	1.3
CCP1C (composite cross)	6	4	1.7	1.4	2.5	1.2
CCP1O (composite cross)			1.8	1.5	2.4	1.5
*C – cultivated under conventional conditions, O – cultivated under organic conditions; F- generation in 2018, ** number of cultivation years in the respective environments in 2018						

DH lines. Comparing yield stability of three DH lines with their advanced initial lines over 11 environments (one pair of lines over 7 environments) no significant differences between the adaptability according to regression analysis were found (Table 6) and it was different for each pair of lines, including broad adaptability, adaptability to favourable and unfavourable environments. Also, the trends in adaptability were similar for both types of lines in each pair. However, the mean yield of DH lines was lower if compared to the initial lines with significant differences in two cases.

Table 6 Comparison of yield and yield stability of doubled haploid (DH) lines and their initial lines over 11 (7) environments

Genotype	Average yield (t ha^{-1})	Coefficient of regression (b)
DH(PR-5602)**	4.38	0.99
PR-5602**	4.44	1.03
DH(PR-5614)**	4.02	1.20*
PR-5614**	4.16	1.16*
DH(PR-5279)**	3.76b	0.65*
PR-5279**	4.10a	0.75*
DH(PR-5779)***	3.38b	1.11
PR-5779***	3.80a	–****

*Significantly different from 1 ($p<0.05$); b – coefficient of regression; **average from 11 environments; ***average of 7 environments; ****regression model not significant; a, b – significant difference within a pair of lines

How was resilience improved?

Improve Y stability over E:

- diversity incorporated in CCPs can improve yield stability over environments.
- Yield advantage found especially under stress environments.
- 3 out of 8 mixtures were stable yielding over the environments (but not all of them).

Participatory/multi-actor approach

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Field trials were demonstrated to organic farmers during the meeting of farmer's association (2018). Information on temporary experiment on marketing of heterogeneous populations was provided and four farmers are involved in the experiment and grow CCP population on their farms (2018-2020). Results were presented to interested farmers and discussed with them in a workshop on heterogeneous material (13.02.2020).

How information was transferred to the other situations

- Breeding for CCPs with well-grounded selection of parents can be recommended;
- Several CCPs will be recommended and offered for farmers;
- Mixtures can have a positive effect with larger probability in respect to the reduction of leaf diseases.

Discussion

The most superior mixture for yield M7 (ranked in the top third in all organic environments) was made of two lines with distinctive resistance to netblotch, one of them with powdery mildew resistance with the main aim to investigate the mixing effect on disease incidence; one of the components was characterised with broad adaptability and yield above average and the other with adaptability to unfavourable environments and yield below average. CCP III made by crosses to male sterile parents provided comparatively lower yield to other CCPs; it makes to think that male sterile parents might have negative effect on yield potential.

Results suggest that combining in mixture genotypes with different adaptability to environments, cannot provide prediction of the mixture adaptability and combination of different traits related to competitive ability against weeds (rapid plant development, good soil covering ability, good tillering, early heading, leaf bending) did not promote better weed suppression ability of the mixture if compared to components.

Only few of the mixture case studies turned out to fulfil the original aim, why they were composed: for M2 broad adaptability was achieved; M7 and M8 were always less infected by net blotch than the most susceptible component. The most superior mixture for yield M7 was made of two lines with distinctive resistance to net blotch, one of them additionally with powdery mildew resistance, with the main aim to investigate the mixing effect on disease incidence; one of the components was characterised with broad adaptability and yield above average and the other with adaptability to unfavourable environments and yield significantly below average.

CCP3 made by crosses to male sterile parents provided comparatively lower yield to other CCPs; it makes to think that male sterile parents might have negative effect on yield potential. Under optimal growing conditions for development of barley, it was more difficult for the populations to compete with the most productive control variety, but it was less affected by drought stress conditions than this productive homogeneous variety.

Outcomes and conclusions

Mixtures can provide advantages over growing in pure stand in respect to incidence of leaf diseases and to lesser extent in respect to yield and weed suppression ability.

It can be possible to obtain stable and good yielding mixtures by mixing components with distinctive disease resistance, yield potential and adaptability but it is not true for all cases. Stable yielding

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mixtures can be produced by mixing components with broad adaptability and adaptability to lower yielding environments.

CCPs were stable yielding over various environments can provide comparable yield results to the check varieties under organic farming and stress conditions. Hulless barley CCP was lower yielding but with higher grain weight, protein and beta-glucan content if compared to the hulless check variety. Diversity of plant traits of organically cultivated sub-populations was slightly larger than that of conventionally cultivated ones in case of complex and CCP population, but not for simple population cultivated under the respective farming system for longer period.

No effect of complete homogeneity of DH lines on adaptability was found, however, they yielded on average lower than the initial advanced lines.

Final conclusions and analysis of mixture component traits and their possible connections to mixture performance will be made after additional analysis of data.

Main outcomes

•Mixtures: largest mixing effect for leaf disease (net blotch), reduction for 7 out of 8 mixtures; smaller effect with positive/negative variation between mixtures and environments for yield and competitive ability traits
•Populations containing greater genetic diversity (CCPs) did ensure better yield performance and stability than those with lower diversity level
•slightly higher average diversity as the effect of organic vrs conventional cultivation for 2-4 years for pops with higher diversity level (CCP, CP), but opposite for simple pops cultivated for 8-10 years. Generally taller plants and better tillering for organically cultivated pops.
•No effect of complete homogeneity of DH lines on adaptability was found, however, they yielded on average lower than the initial advanced lines.
•More data analysis planned/needed (limited resources and knowledge)

Main lessons learned

•Strategy to combine mixtures with components differing for 3 target traits did not ensure expected effect in all cases and therefore cannot be generalized. Stable mixtures arose by combining components with different adaptability (but not in all cases).
•CCPs have a potential for organic and stress environments (yield and stability might be better than for homogenous varieties, diseases and weed competition - similar)

Publications

Peer-reviewed article on populations (EN), 2 shorter papers on populations (Latvian), abstract on mixtures and populations at the EUCARPIA-LIVESEED conferences (2018, 2021), PhD Thesis (defence in May 2021), paper on mixtures planned in 2021.

Supplementary material

Figure 2. Relative mixing effect for yield over 7 organic and 7 conventional environments

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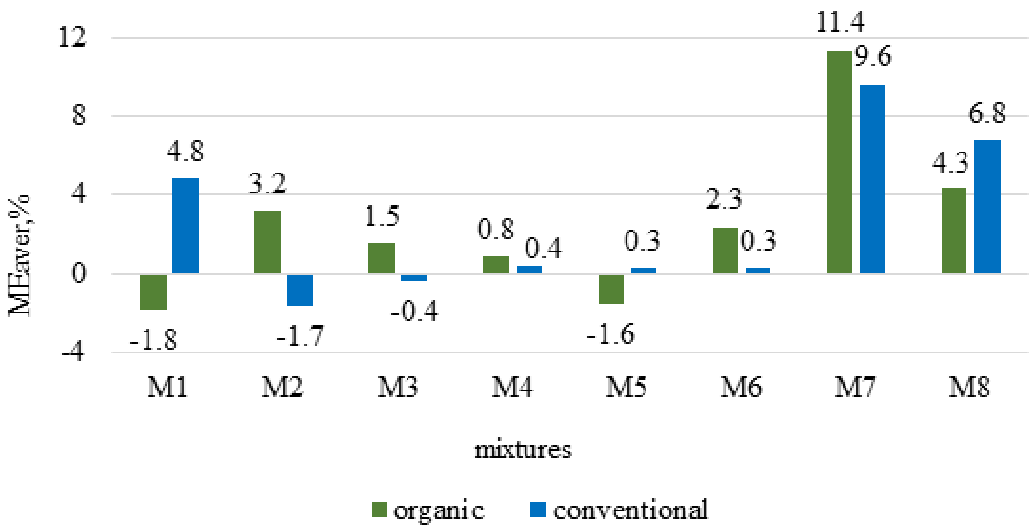

Table 7 Deviation of the average yield of mixtures from the average yield of the components (number of cases)

Mixture	Organic (n=7)		Conventional (n=7)	
	+/- average of components**	+/- average, %	+/- average of components	+/- average, %
M1 (3)*	-5;+2	**-3**	**-1;+6(1)***	**5**
M2 (3)	-3;+4	**3**	**-4;+2;=1**	**-2**
M3 (3)	-4;+2	**2**	-2;+2;=3	**0**
M4 (5)	-3;+3	**1**	-3;+3;=1	**0**
M5 (2)	-4;+3	**-2**	-2;+2;=3	**0**
M6 (3)	-1(**1**);+5;=1	3	-3;+4	**0**
M7 (2)	-2;+5	6	-1;+6(**1**)	9
M8 (3)	+4;-2	3	-1;+6(**2**)	7

*Number of components; ** number of cases when yield was lower (-)/higher (+)/equal (=) than that of average of the components; *** in brackets in red bold – number of cases when differences are significant (p<0.05)

Figure 3. Relative mixing effect for crop ground cover and competitive ability against weeds (4 organic environments)

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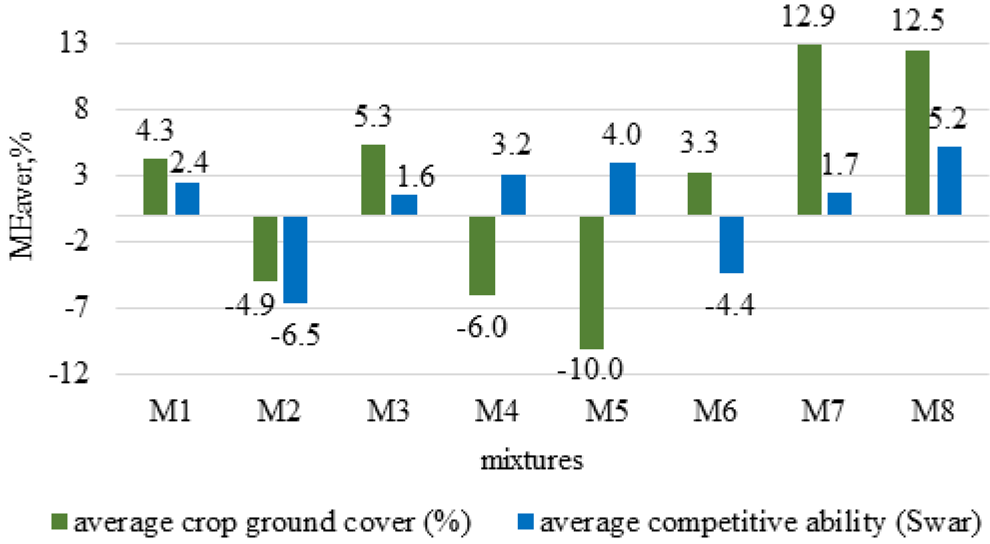

Table 8 Deviation of the average crop ground cover (CGC) and average ability to suppress weeds (S_{var}) of mixtures from the average of the components (number of cases)

S_{var} – 4 environments × 3 observations = **12 cases**

CGC – 4 environments × 2 observations= **8 cases**

Mixture	+/- Average of components**	
	S_{var} (n=12)	CGC (n=8)
M1 (3)*	-4;+7(**1**)***;=1	-2;+6
M2 (3)	-6;+6	-7;+1
M3 (3)	**-4;+7(2);=1**	**-2;+6**
M4 (5)	**-7;+5**	**-6;+1;=1**
M5 (2)	**-4;+8**	**-7;+1**
M6 (3)	-10;+2	-3;+5
M7 (2)	-5;+7	**8**
M8 (3)	-3;+9	**8**

*number of components; ** number of cases when indicator was lower (-)/higher (+) or equal (=) than that of average of the components; *** in brackets in bold – number of cases when differences are significant ($p<0.05$)

Figure 4. Relative mixing effect for net blotch severity (4 organic, 4 conventional environments)

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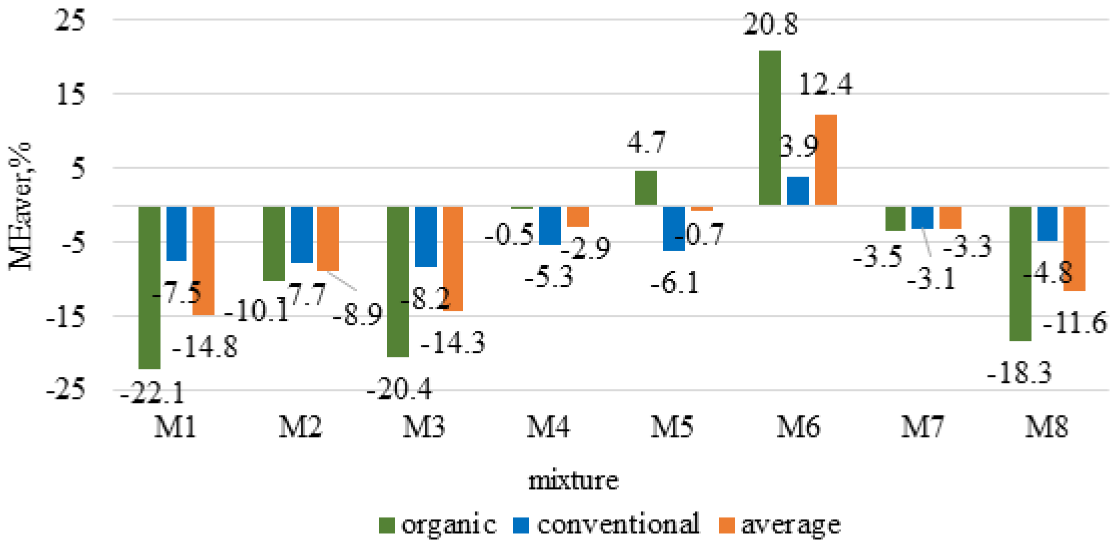

Table 9. Deviation of the average net blotch infection level of mixtures from the average infection level of the components

Mixture	+/- Average of components**	
	organic	conventional
M1 (3)*	**-3(1)*** **	**-3**
M2 (3)	**-3**	**-3**
M3 (3)	**-3(1)**	-3
M4 (5)	-1;+2	-1;+1;=1
M5 (2)	-1;+2(**1**)	-2;+1
M6 (3)	**+3**	**-2;+1**
M7 (2)	**-2;+1**	**-2(1);+1(1)**
M8 (3)	**-3(1)**	**-2;+1**

*number of components; **number of cases when indicator was lower (-)/ higher (+) or equal (=) than that of average of the components; *** in brackets in bold – number of cases when differences are significant (p<0.05)

Table 10 Comparison of net blotch infection level of populations and check varieties (number of cases)

6 environments (3 O and 3 C) × 4 simple populations = **24 cases**

6 environments (3 O and 3 C) × 5 complex populations = **30 cases**

Type of populations	Comparison with check variety*		
	+/- Abava	+/- Rasa	+/- Rubiola
simple n=4	-24(**23**)**	-22(**16**);+1;=1	-18(**9**);+5;=1
complex n=5	-30(**30**)	-29(**16**);+1	-25(**9**);+4;=1
CCP1	-6(**6**)	-6(**5**)	-4(**4**);=2

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CCP3	-6(**6**)	-6(**5**)	-5(**2**);+1
CCP4	-6(**6**)	-6(**4**)	-6(**4**)
CCP5***	-4(**4**)	-4(**4**)	-4(**2**)

*number of cases when indicator was lower (-)/higher (+) or equal (=) than that of average of the components; **in brackets in bold – number of cases when differences are significant (p<0.05);***results from four environments

Table 11 Average crop ground cover and weed suppression ability of populations and check varieties

Type of populations/ variety	Crop ground cover (%)		Weed suppression ability S_{var}		
	GS 25-29	GS 29-31	GS 31-39	GS 59-65	GS 87-92
simple n=4*	11.5	17.9	47.0	71.7	65.7
complex n=5*	11.0	13.1	51.7	70.8	64.4
CCP1*	10.8	17.8	45.1	69.4	65.2
CCP2*	9.1	15.8	48.0	65.9	61.7
CCP3*	10.9	17.1	47.1	70.0	64.0
Abava*	16.8	22.1	52.5	74.3	67.2
Rasa*	10.3	17.1	48.4	69.5	62.4
Rubiola*	12.5	17.2	50.2	66.1	62.7
CCP4**	16.3	21.0	50.3	62.3	67.9
Abava**	16.8	24.2	49.0	68.3	65.8
Rasa**	11.1	18.1	42.8	61.6	62.2
Rubiola**	14.1	19.1	46.4	59.2	61.9
CCP5***	16.3	24.0	42.5	70.2	72.9
Abava***	16.8	25.1	40.6	69.1	70.3
Rasa***	11.8	20.0	31.5	62.1	67.4
Rubiola***	14.9	21.5	43.9	62.0	64.0

*average from four years; **average from three years; ***average from two years

Table 12 Average plant length and its diversity in parallel populations

Year	Population	Organic			Conventional		
		average length, cm	minimum/ maximum length, cm	H'[&]	average length, cm	minimum/ maximum length, cm	H'
2016	BZ12C^	77.8	59-98	2.04	78.5	60-99	1.92
	BZ12O^	77.1	56-107	2.07	77.1	58-100	2.04
2018	BZ12C	61.3**	43-83	1.84	71.6*	42-99	2.01
	BZ12O	64.7**	52-84	1.49	64.7*	52-84	1.49
2016	1018-12C	78.2	56-101	2.01	65.7*	56-95	1.46
	1018-12O	80.9	57-100	1.93	68.0*	46-87	1.78
2018	1018-12C	65.2**	52-82	1.64	56.6**	43-75	1.65
	1018-12O	76.6**	52-103	2.27	73.8**	51-95	2.01
2016	CCP1C	64.7	47-82	1.82	69.2	53-87	1.68
	CCP1O	67.8	46-82	1.80	69.2	54-85	1.52
2018	CCP1C	60.5**	49-73	1.41	55.9*	42-75	1.75

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	CCP1O	70.4**	52-62	2.02	58.5*	44-74	1.75

*average plant length of parallel populations differs significantly within a given year and environment (p<0.01); ** (p<0.001); & Shannon-Weaver diversity index; ^C – grown under conventional conditions, O – grown under organic conditions

Table 13 Average length of the main spike and its diversity in parallel populations

Year	Population	Organic			Conventional		
		average length, cm	minimum/ maximum length, cm	H'&	average length, cm	minimum/ maximum length, cm	H'
2016	BZ12C^	7.3*	4-12	1.68	6.5*	4-10	1.55
	BZ12O^	6.9*	5-10	1.59	6.1*	3-9	1.52
2018	BZ12C	6.8*	5-9	1.39	6.7***	5-9	1.33
	BZ12O	7.0*	4-9	1.26	6.3***	5-8	1.20
2016	1018-12C	6.9	4-9	1.65	6.1	4-9	1.41
	1018-12O	7.0	4-10	1.62	6.1	3-8	1.47
2018	1018-12C	7.2	5-11	1.41	6.5*	4-9	1.31
	1018-12O	7.4	5-11	1.48	6.7*	5-9	1.37
2016	CCP1C	7.5**	5-10	1.53	6.5	4-9	1.51
	CCP1O	6.9**	4-10	1.62	6.5	4-10	1.58
2018	CCP1C	7.1***	5-10	1.30	6.5	4-10	1.30
	CCP1O	8.4***	7-12	1.44	7.0	5-10	1.48

*average plant length of parallel populations differs significantly within a given year and environment (p<0.05); ** (p<0.01); ***(p<0.001) ; &Shannon-Weaver diversity index; ^C – grown under conventional conditions, O – grown under organic conditions

Table 14 Average number of grains in the main spike and its diversity in parallel populations

Year	Population	Organic			Conventional		
		average	minimum/ maximum	H'&	average	minimum/ maximum	H'
2016	BZ12C^	22.0	10-32	2.77	17.8**	10-28	2.63
	BZ12O^	21.0	10-31	2.76	16.4**	6-25	2.55
2018	BZ12C	22.7***	15-32	2.58	21.2***	14-28	2.41
	BZ12O	20.4***	13-25	2.30	17.9***	10-24	2.24
2016	1018-12C	19.0	10-29	2.58	15.2	10-22	2.43
	1018-12O	19.2	10-27	2.52	15.7	10-24	2.44
2018	1018-12C	22.2**	15-28	2.35	19.5***	11-26	2.30
	1018-12O	22.9**	16-30	2.43	21.0***	14-27	2.42
2016	CCP1C	19.8	10-27	2.70	18.1	10-26	2.56
	CCP1O	19.7	10-30	2.58	17.9	10-26	2.67
2018	CCP1C	22.1*	16-29	2.39	19.8	10-27	2.40
	CCP1O	23.1*	18-29	2.17	19.0	12-24	2.17

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*average plant length of parallel populations differs significantly within a given year and environment (p<0.05); ** (p<0.01); ***(p<0.001) ; &Shannon-Weaver diversity index; ^C – grown under conventional conditions, O – grown under organic conditions

Table 15 Average coefficient of productive tillering and its diversity in parallel populations

Year	Population	Organic			Conventional		
		average	minimum/ maximum	H'&	average length, cm	minimum/ maximum	H'
2016	BZ12C^	4.7*	2-13	2.00	3.2	2-6	1.30
	BZ12O^	3.9*	2-7	1.55	3.2	2-5	1.16
2018	BZ12C	2.3***	2-4	0.61	2.4	2-5	0.79
	BZ12O	3.2***	2-8	1.56	2.5	2-6	0.87
2016	1018-12C	3.1***	2-7	1.38	3.2	2-5	1.04
	1018-12O	4.8***	2-10	1.78	3.4	2-6	1.20
2018	1018-12C	2.5***	2-4	0.85	2.7**	2-7	1.16
	1018-12O	3.3***	2-9	1.53	2.4**	2-5	0.75
2016	CCP1C	3.4***	2-7	1.48	3.5***	2-6	1.16
	CCP1O	3.9***	2-7	1.68	3.9***	2-7	1.46
2018	CCP1C	2.7***	2-7	1.05	2.8***	2-6	1.20
	CCP1O	3.1***	2-7	1.48	3.5***	2-6	1.28

*average plant length of parallel populations differs significantly within a given year and environment (p<0.05); ** (p<0.01); ***(p<0.001) ; &Shannon-Weaver diversity index; ^C – grown under conventional conditions, O – grown under organic condition

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IPC- Maize - Azorean trials and characterization 2018-2020 (Trial #7)

Lead: IPC– Portugal
Emanuel Ferreira, Duarte Pintado, André Pereira Pedro Mendes Moreira – pmm@esac.pt

Abstract

Portugal has a distinct type of maize landraces. Most of their uses are related to the human consumption and animal feeding.
The main objective of our work was the regeneration, multiplication, characterization, agronomic evaluation of the germplasm collection of Azores Islands plus VASO germplasm to provide varieties adapted to organic and agroecological agricultural systems.
In 2018 and 2019, two agronomic evaluation tests were done with 20 maize populations from Azores. The trials in addition with seed multiplication allowed to characterize the Azorean germplasm and provide the production of 2 CCPs to be introduced in Participatory Plant Breeding (PPB) in low input organic systems.
In 2020 two trials were performed; Organic vs Conventional: in two locations distance 1.3 km and intended to compare two agriculture systems, the low input organic agriculture (Caldeirão) and conventional agriculture; On-farm PPB: 2 locations distance 15 km in Sousa Valley region, Macieira de Lixa and Lousada (agroecological sites).

Introduction

In Portugal maize landraces are being used for human consumption for centuries, not only in mainland but also in Islands. Maize is used for maize bread, polenta, and other specialties. IPC-ESAC has been working on participatory plant breeding towards the valorisation of landraces for niche markets and adapted germplasm for marginal areas and to agroecological and organic agriculture.
The market of organic seeds in Portugal (DATA BASE ORGANIC SEEDS, DGADR) only have two varieties of maize (sweet corn), with recommended region for Netherlands and Europe, registered in 2018. Which means that most organic maize seed in Portugal is based on derogations. This fact highlights the opportunity to produce varieties adapted to organic and food purposes for the seed market. In 2020 the agricultural area for organic was referred as 8.1%, which means a lot of effort until 2030 to reach 25%, in addition until 2036 it is expected to reach 100% of organic seed.
With this trial we intend to both multiply some germplasm that was collected in the 70s and 80s plus recently collected germplasm that was in cold storage, and to create two CCPs that could help us to obtain germplasm for organic and low input agriculture that could be adapted to Azores and Portugal mainland. With this purpose we did adapt the crossing methodology that was described in Mendes Moreira et al. 2009 (Mendes-Moreira P, Vaz Patto MC, Mota M, Mendes-Moreira J; Santos JPP, Santos JPNS, Andrade E, Hallauer A, Pêgo S (2009) 'Fandango': long term adaptation of exotic germplasm to a Portuguese on-farm-conservation and breeding project. Maydica 54: 269-285. ISSN: 0025-6153).

Material and Methods

Genetic resources

The germplasm used for trials included 50 accessions: 40 populations were collected in 1979 at Azores (Bettencourt and Gusmão, 1982) and multiplied in 2018 and 2019 at ESAC; 7 populations from the Participatory Plant Breeding Program "VASO" in Sousa Valley, Portugal; 3 Composite Cross Population (CCP).

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The list in the Table 1 displays the germplasm used in the maize trials and in seed multiplication.

Table 1. List of germplasm used in seed multiplication and in trials (2018-2020).

			2018-2020	2018-2019	2020	
			Seed Multiplication	Evaluation trial	Organic vs Conventional trial	PPB Trial Agroecological site
1	2444	Azores, Portugal	x		X	
2	2445	Azores, Portugal	x			
3	2446	Azores, Portugal	x			
4	2448	Azores, Portugal	x	x	X	
5	2449	Azores, Portugal	x		X	
6	2488	Azores, Portugal	x	x	X	
7	2489	Azores, Portugal	x	x	X	
8	2492	Azores, Portugal	x			
9	2493	Azores, Portugal	x	x	X	
10	2494	Azores, Portugal	x	x	X	
11	2496	Azores, Portugal	x	x	X	
12	2497	Azores, Portugal	x	x		
13	2498	Azores, Portugal	x		X	
14	2499	Azores, Portugal	x		X	
15	2500	Azores, Portugal	x			
16	2501	Azores, Portugal	x		X	
17	2502	Azores, Portugal	x		X	
18	2503	Azores, Portugal	x			
19	2504	Azores, Portugal	x		X	
20	2505	Azores, Portugal	x	x	X	
21	2506	Azores, Portugal	x			
22	2507	Azores, Portugal	x		X	
23	2508	Azores, Portugal	x		X	
24	2509	Azores, Portugal	x	x	X	
25	2510	Azores, Portugal	x		X	

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26	2511	Azores, Portugal	x			
27	2512	Azores, Portugal	x			
28	2513	Azores, Portugal	x		X	
29	2514	Azores, Portugal	x		X	
30	2515	Azores, Portugal	x	x	X	
31	2516	Azores, Portugal	x		X	
32	2517	Azores, Portugal	x		X	
33	2518	Azores, Portugal	x	x	X	
34	2519	Azores, Portugal	x		X	
35	2520	Azores, Portugal	x			
36	2521	Azores, Portugal	x			
37	2522	Azores, Portugal	x		X	
38	2524	Azores, Portugal	x	x	X	
39	2525	Azores, Portugal	x		X	
40	2526	Azores, Portugal	x		X	
41	2527	Azores, Portugal	x	x	X	
42	2528	Azores, Portugal	x	x	X	
43	2529	Azores, Portugal	x		X	
44	2530	Azores, Portugal	x		X	
45	2531	Azores, Portugal	x		X	
46	Amc397	Vale do Sousa Portugal			X	x
47	Fn 2014	Vale do Sousa Portugal			X	x
48	Pg COSO 18	Vale do Sousa Portugal			X	x
49	Pg COSO 19	Vale do Sousa Portugal			X	x

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50	Pg C0S0 19 - (Lousada 2019)	Vale do Sousa Portugal			X	x
51	SinPre -	Vale do Sousa Portugal			X	x
52	VA C0S0 17 - (Sequeiro Lousada 2017)	Vale do Sousa Portugal			X	x
53	VA C0S0 19 - (Regadio Lousada 2019)	Vale do Sousa Portugal			X	x
54	VA C0S0 19 - (Sequeiro Lousada 2019)	Vale do Sousa Portugal			X	x
55	VT17 - (Caldeirão 2018)	Azores, Portugal	x	x	X	
56	MT17 - (Caldeirão 2018)	Azores, Portugal	x	x	X	
57	MONJ-3 - (Caldeirão 2018)	Azores, Portugal	x	x	X	
58	MONJ-2 - (Caldeirão 2018)	Azores, Portugal	x	x	X	
59	BT18 - (37-2019 Caldeirão 2019)	Azores, Portugal	x		X	
60	BT17 - (Caldeirão 2018)	Azores, Portugal	x	x	X	
61	BSM17 - (Caldeirão 2018)	Azores, Portugal	x	x	X	
62	Bulk-Azores1 - (Caldeirão 2018)	Coimbra, Portugal	x	x	X	
63	Bulk-Azores 2	Coimbra, Portugal	x		X	x
64	Milho Vermelho	Vale do Sousa Portugal			x	x

Location

The trials were managed at the low input organic field (40.21709426119619; -8.44779968261719 W; 15 m) and conventional field (40.214724 N, -8.480292 W) of IPC-ESAC in Bencanta, Coimbra, Portugal. On-Farm Trial and PPB mass selection were done at Alvarenga, Lousada, Portugal (41.29412380145764; -8.26727628707886 Altitude 286 m) and Macieira de Lixa, Felgueiras, Portugal (41.345100, -8.169200; 406 m elevation).

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Experimental design

2018

The trial was sowed on 15 May 2018 (BT17, BSM17, 2497 and MONJ-2) and 16 of May 2018 (the other populations).

The field had maize as the preceding crop and no fertilization inputs were made.

The plots were design (Figure 1) in lines of 10, 20, 30 and 40 linear meters (depending on the number of seeds available), 2 rows with 0.75 m inter-rows, and 0.2 m between plants (making a standard density with approximately 60 000 seeds/ha).

The mechanical weed control was made on 29 of May and 16 of June 2018, and the manual weed control was occurred from June to July.

The trial was harvested by hand on 3rd November of 2018 (2497 popcorn) and the other populations on 15th November of 2018.

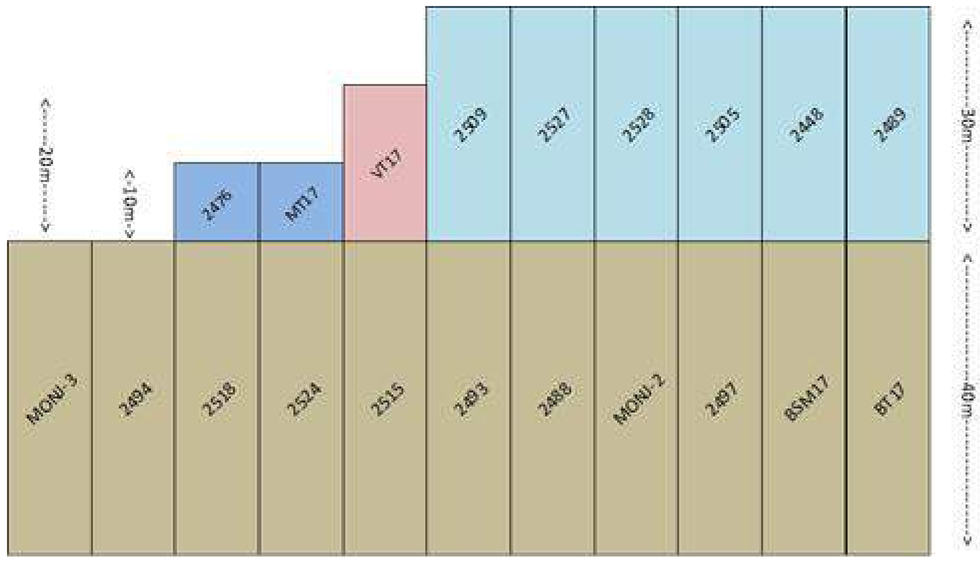

Figure 1. Azorean trial design 2018.

2019

The trial was sowed on 15th May 2019. The field hadn't preceded crops and no fertilization was done since the last year maize campaign and the 2019.

The 20 accessions were evaluated in a randomized complete block design at Coimbra (Figure 2), with a 60 000 stand, in plots of two lines with 6.4 length and 0,75 interrow distance. Only the CCP were evaluated in the three locations (Alvarenga, Azores and Coimbra). For Alvarenga and Azores the evaluation consisted in a sampling of three plots of 9,6 m2 (similar to the plots) randomly distributed and within the field (for Alvarenga harvest occurred in early November of 2019). The production system used were respectively low input and organic agriculture in Azores and Coimbra, and low-input conventional system in Alvarenga).

Figure 2. Azorean Trial design in a randomized complete block of 3 repetitions per population.

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2020

The Organic Vs Conventional trial was sowed on the 2nd June 2020 in two locations distance 1.3 km and intended to compare two agriculture systems, the low input organic agriculture (Caldeirão) and conventional agriculture (Vagem Grande). The 50 accessions were evaluated in a randomized complete block design with 60 000 stand, in plots of two lines with 6.4 length and 0,8 interrow distance.

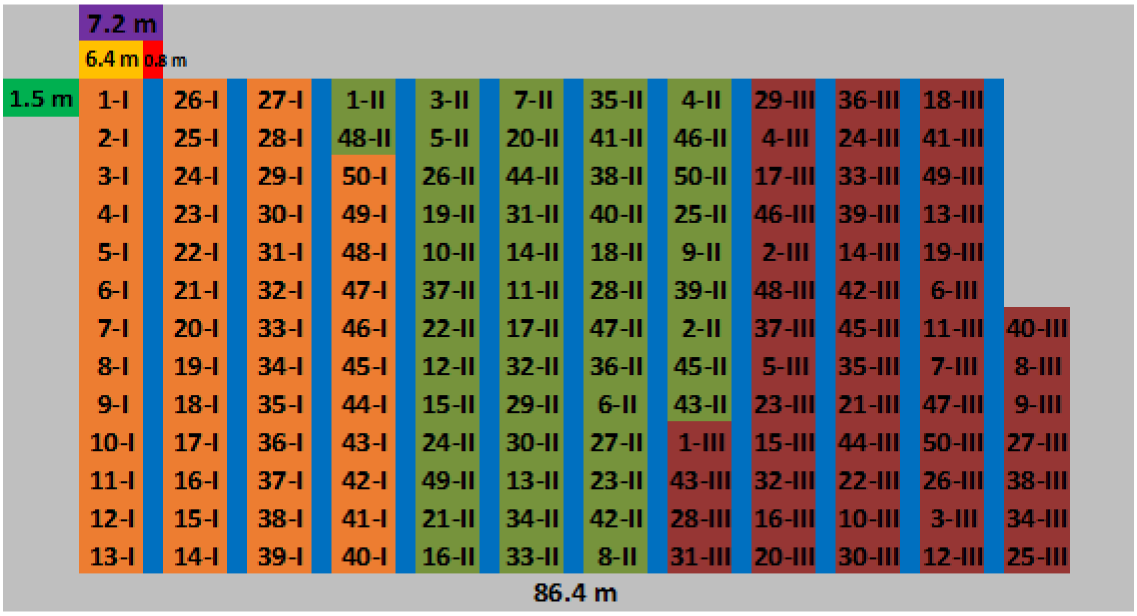

Figure 3. Azorean trial design 2020 (Organic vs Conventional).

The On-farm PPB trial was sowed on the 28st May 2020 in 2 locations distance 15 km in Sousa Valley region, Macieira de Lixa and Lousada (agroecological sites). The 10 accessions were evaluated in a randomized complete block design with 60 000 stand, in plots of two lines with 6.4 length and 0,8 interrow distance.

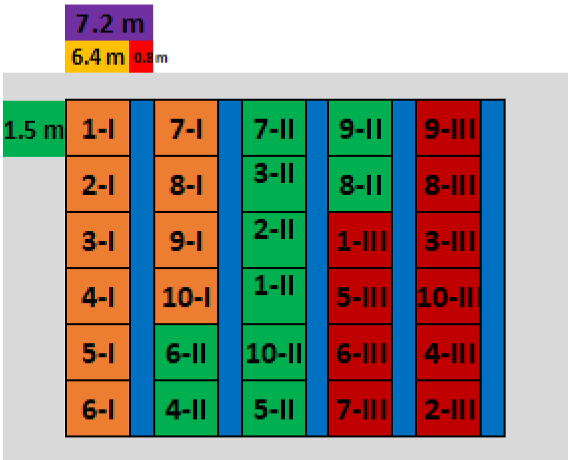

Figure 4. Azorean trial design 2020 (Agroecological sites).

Statistical methods

ANOVA, descriptive statistic, and correlations analysis was done with IBM SPSS Statistics program and Excel.

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Evaluation

The HUNTERS descriptor or scale (High, Uniformity, aNgle, Tassel, Ear, Root lodging and Stalk lodging) was developed by Silas Pego (Table 5, p84). Below we describe in Table 2-4 the List of the traits collected in the trials.

Table 2. List of the traits collected for maize concerning crop development and agro-ecological performance

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Field emergence	Counting of plants	Quantitative (plants/m2)

Table 3. List of the traits collected for maize concerning crop characterisation

Crop characterization				
Traits	**Codes**	**Scale**	**Type of data available**	**How it has been assessed / Description**
Stand*		Plants ha-1		Thousands of plants per hectare;
Moisture	MO	%	Plot (considering 4 ears)	The grain moisture was measured with the ISOELECTRIC GRAIN CHECK® moisture meter, using the grain of the average four ears;
Overlap Index	OI		Index	This method enables the knowledge of a population concerning the relative amount of theoretical allogamy versus autogamy;
Height	H	cm	20 plants	Plant height, from the stalk basis to the last leaf insertion before the tassel;
Height of the First Ear	H1E	cm	20 plants	Ear height, from the stalk basis to the highest ear bearing node;
Uniformity	U	1 to 9	Plot	1-minimum uniformity and 9 – maximum; 1-4 to pure lines and 5-9 to populations;
Leaf Angle	N	1 to 9	Plot	Angle of the adaxial side of the leaf above the ear with the stalk (5=45°, <5 =<45° and >5 = >45°C);
Tassel Branching	T	1 to 9	Plot	1- absent tassel (Inbreeds and hybrids) 9- a much branched tassel (frequent in populations with abnormal fasciated ears);

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Ear Placement	E	1 to 9	Plot	5- indicates that the ear is in the middle of the plant, if <5 bellow and if >5 above the middle of the plant;
Root Lodging *	R	%	%	Percentage of plants leaning more than 30º from vertical;
Stalk Lodging *	S	%	%	Percentage of plants broken at or below the primary ear node, related with the quality of the stalk and the stalk damage caused by some insect attack;
Ear Length	L	cm	20 ears	Ear length;
Ear Diameter 1 and 3	DE1, DE3	cm	20 ears	Large diameter in the 1/3 bottom and top of the ear respectively;
Ear Diameter 2 and 4	DE2, DE4	cm	20 ears	Small diameter in the 1/3 bottom and top of the ear respectively (90° rotation from large diameter) (cm);
Kernel-Row Number 1 and 2	R1, R2		20 ears	Row number in the 1/3 bottom and top of the ear respectively (nº);
Fasciation	Fa	1 to 9	20 ears	1 – without fasciation and 9 = maximum of fasciation;
Determinated vs Indeterminated	D_I		20 ears	Top of the ear full of grain, case of determinate ears (2) or not, case of indeterminate ears (1), average value is calculated;
Convulsion	CV	0 to 5	20 ears	Kernel row arrangement in the ear (0 - without convulsion, regular kernel row arrangement, 5 – maximum of convulsion, without kernel row arrangement);
Flint vs Dent	F/D		20 ears	1- Popcorn, 2-flint, 3-medium flint, 4-low flint, 5 - 50% flint and 50%dent, 6 - low dent, 7-medium dent, 8-high dent, 9-sweet maize;
Ear Weight	EW15	g	20 ears	Ear weight adjusted to 15% of grain moisture;
Kernel Weight	KW15	g	20 ears	Kernel weight per ear, adjusted to 15% moisture;
Cob Weight/Ear Weight	CW_EW	%	20 ears	Indicates the percentage of cob weight in the ear weight;
Ears Moisture	HR%		20 ears	The grain moisture was measured with the ISOELECTRIC GRAIN CHECK® moisture meter, using the grain per ear and calculating the average;
Kernel Depth	KD	cm	20 ears	Measure of one kernel in the middle of the ear;
Kernel Number	KN		20 ears	Kernel number per ear;
Thousand Kernel Weight	SW15	g	20 ears	Thousand kernels weight at 15% moisture content;

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Kernel per Row	NC		20 ears	Kernel number per row;
Cob Diameter 1 and 3	DC1, DC3	cm	20 ears	Large diameter in the 1/3 bottom and top of the cob respectively;
Cob Diameter 2 and 4	DC2, DC4	cm	20 ears	Small diameter in the 1/3 bottom and top of the cob respectively (90° rotation from large diameter);
Medulla 1 and 2	M1, M2	cm	20 ears	Large and small length of medulla respectively;
Rachis 1 and 2	Ra1, Ra2	cm	20 ears	Large and small length of rachis respectively;
Cob Color	CC		20 ears	Cob colour: 1 is red and 2 is white.

Table 4. List of the traits collected for maize concerning crop quality performance.

Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**
Estimated Yield (15% moisture) *	Using KW at 15% moisture, average of 20 plants, an estimation was made to a production to a density of 60000 plants per ha.	Quantitative (Mg/ha)

*- Due to storm Leslie, on 12 October 2018, it was not possible to collect the data

How resilience was improved?

- The germplasm used have been multiplied on a marginal land (low input organic system) for two years (2018 and 2019).
- 2 CCP were created from 40 populations that were collected in Azores – BulkAzores1 and BulkAzores2; incorporating diversity and heterogeneity.

Participatory/multi-actor approach

2018-2019

The multiplication trials made possible the recovery of long-term stored germplasm and the creation of 2 CCPs. This CCPs will be used under multi-actor approach, from the seed (continuation of building the CCP) to farmers, processors, and consumers.

2020

PPB field:

Farmers and stakeholders were requested to give inputs having as basis the on-farm field at Macieira da Lixa location and indicated from the 10 populations, their preferred germplasm for food and feed.

Culinary breeding

EXPLORING PORTUGUESE MAIZE LANDRACES AS FRESH MAIZE

The aim of this work was to make a sensory analysis with five maize landraces grown in organic agriculture. Colour, texture, and flavour were some of the attributes observed and studied to know what the purchase intention would be among those who tried the maize landraces.

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

-The maize landraces (‘Milho Vermelho’, 2495, ‘Amiúdo’, ‘Pigarro’, ‘Broa 70’) were harvested on August 20th, 2020, at milk stage (R3).
-30 participants contributed to identify the characteristics of the maize that most attracted their attention
-Each sample was presented sequentially per se to participants.
-Each participant had tasted five varieties -The evaluated attributes were: colour, flavour, texture, global impression. The scale used was hedonic with nine points, varying between 9 as I liked it very much and 1 as I disliked it a lot.
-The buy intention test was also carried out, where a five-point scale was also used, ranging from certainly not buying to certainly buying
-Data were submitted to PostHoc methods, Kruskal-Wallis test using the IBM SPSS 26.

How the information can be transfer to the other situations?

Under LIVESEED experiments we create CCPs and made them available to farmers.
Analysis's methods used by us for maize, as MARS, CART, RF can be transferred to other situations.

Results and discussion

2018

The agronomic evaluation and characterization allowed us to separate the early (68 days) and late (83 days) populations. Plant height range from 147 cm to 305 cm. The populations reveal high level of diversity. The angle of the leaf indicate that the populations are mainly adapted to low densities with 30% with value 7. The ear insertion on the plant showed a value 5 for the 20% of the populations. Ear Length range from 15,2 to 21,2 cm. Kernel weigh at 15% moisture, indicates variations across populations from 68,0 to 247,9 g. Also, the cob and ear ratio range 18,6 to 28,0%. The thousand kernel weight range from 408,0 g to 718,0 g which indicates big kernel types and include popcorn (155,5 g). Due to storm Leslie, it was not possible to quantify the yield (Yield), the root lodging (R), the stalk lodging (S) and the stand. However, an estimation was done based on the average of 20 ears kernel weight at 15% moisture considering a density of 60 000 plants (Figure 5). The landrace 2524 was the variety that had the highest expected yield production (14.88 Mg h-1) followed by 2528 (13.26 Mg ha-1), the variety 2497 (popcorn) was the less productive (4.08 Mg ha-1).

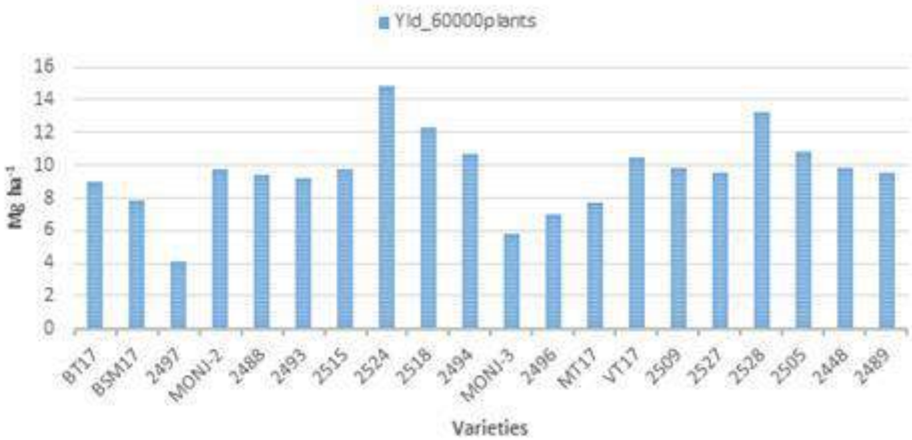

Figure 5. Estimated yield based on the average of twenty ears kernel weight at 15% moisture for a density of 60 000 plants per hectare 2018.

2019

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The CCP height comparisons among environments indicate that Azores and Alvarenga (257.4 cm and 258.2 cm respectively) were significantly higher than Coimbra (211.3cm). The tested populations in Coimbra's trial, indicates that significant differences occurred to plant Height (H), height of first ear (H1E) and Stalk lodging (S) parameters.

The plant Height (H) (Table 1) trait indicates that the values range from 211.3cm (Bulk-Azores in Coimbra) to 260.3 cm (2515). To first ear height (H1E) average there was a variation between 107.9 cm (2505) and 186.5 cm (2515).

The Uniformity (U) average range from 1 to 2 to all varieties in all environments. For the leaves aNgle (N) and Ear insertion (E) the average values range from 5 to 6 and the Tassel (T) presented values between 6 and 7 to all tested accessions in all environments. The Root lodging (R) shown values between 0% (2496) and 9.09% (2509) and the Stalk lodging (S) range from 3.56% (2496) and 17.22% (2527).

Due to the later sowing and consequent harvesting in Alvarenga, the results for Root and Stalk lodging (R & S), Prolificity (Prol.), cob and ear weight ratio (CW/EW) and Yield are ongoing. The populations shown Yield results among 2.2 Mg/ha (MONJ-3) and 5.159 Mg/ha (2489), and the CCP in Coimbra obtained 3.34 Mg/ha and in Azores 4.01 Mg/ha.

Table 5. Characterization of the 20 Azorean maize landraces according to HUNTERS descriptor.

Genotype	H		H1E		U		N		T		E		R%		S%		Pf	
	Mean	CV	Mean	CV	Mean	CV	Mean	CV	Mean	CV	Mean	CV	Mean	CV	Mean	CV	Mean	CV
2524	246,65	0,0699	137,67	0,0974	1,67	0,3464	6	0	6	0	5,33	0,1083	8,32	0,6815	21,93	0,2356	1,04	0,0385
2518	222,75	0,0560	132,57	0,0875	1,67	0,3464	5	0	5,67	0,1019	6	0	5,40	0,7443	9,89	0,4761	0,94	0,0821
BSM17	248,82	0,0854	159,4	0,1153	2,33	0,2474	6	0	6	0	6	0	2,94	1,1924	10,27	1,1625	0,93	0,0455
MONJ-3	228,98	0,1272	143,65	0,1580	1,33	0,4330	5	0	6	0,1667	6	0	4,86	0,8921	12,46	1,2875	0,97	0,0623
BT17	221,65	0,0268	122,27	0,0702	1,33	0,4330	6	0	6	0	5,33	0,1083	3,24	1,2282	3,31	0,8992	1,02	0,0722
2528	228,12	0,1081	142,12	0,1784	1,33	0,4330	5,67	0,1019	6	0	5,67	0,1019	4,96	0,8921	13,75	0,8847	1,04	0,0859
2515	260,27	0,0954	172,15	0,1220	2	0	5,67	0,1019	7	0	5,67	0,1019	7,36	1,0615	13,30	1,3067	1,04	0,0988
MT17	206,73	0,0454	115,33	0,0303	1,67	0,3464	5,67	0,1019	6,33	0,0912	5,33	0,1083	0	0,0000	6,42	0,9099	1,01	0,0377
2505	213,12	0,0083	110,65	0,0220	1,67	0,3464	5	0	6	0	5	0	0	0,0000	5,89	0,9124	1,15	0,1576
2448	225,23	0,0630	129,77	0,1278	1	0	5	0	6,33	0,0912	5,67	0,2038	1,33	0,9007	8,50	1,3593	0,89	0,0325
VT17	209,42	0,0272	113,23	0,0542	2	0	5,33	0,1083	6	0	5	0	3,46	1,0349	11,63	0,3862	1,11	0,0672
2496	225,05	0,0947	122,15	0,0429	2	0,5	5	0	6,33	0,1823	5	0	0	0,0000	3,56	0,9437	0,91	0,1275
Bulk-Azores	211,3	0,0111	117,95	0,0235	1,33	0,4330	5	0	6	0	5,33	0,1083	2,91	0,8840	11,31	0,1700	1,05	0,0881
2509	219,32	0,0438	123,08	0,0863	1,67	0,3464	5	0	6,67	0,0866	5	0	9,09	0,9748	8,31	0,2117	0,99	0,0310
2489	222,07	0,0250	133,2	0,0894	1,67	0,3464	5,33	0,1083	6	0	5,67	0,2038	2,84	0,6597	4,51	0,7537	0,95	0,0179
2494	224,18	0,0313	131,02	0,0277	1	0	5,67	0,1019	6	0	5,67	0,2038	0,74	1,7321	9,81	0,8992	1,05	0,0532
2493	218,38	0,0344	125,52	0,0737	1,67	0,3464	5,33	0,1083	6	0	5,33	0,1083	5,63	0,5346	7,66	0,6581	1,01	0,0569
MONJ-2	248,78	0,0294	140,67	0,0493	2	0	5	0	7	0	5,33	0,1083	5,66	0,6175	8,42	0,4642	1,01	0,0963
2527	235,05	0,0740	130,63	0,0748	1,67	0,3464	5,33	0,1083	6,33	0,0912	6	0,1667	1,11	1,7321	17,22	0,4573	0,96	0,0501
2488	245,32	0,0648	147,48	0,0657	2	0	5	0,2	6	0	5,33	0,1083	4,01	1,0675	15,32	1,0932	1,06	0,0881

The populations showed Yield at 15% moisture results among 2.2 Mg/ha (MONJ-3) and 5.159 Mg/ha (2489) (Figure 6).

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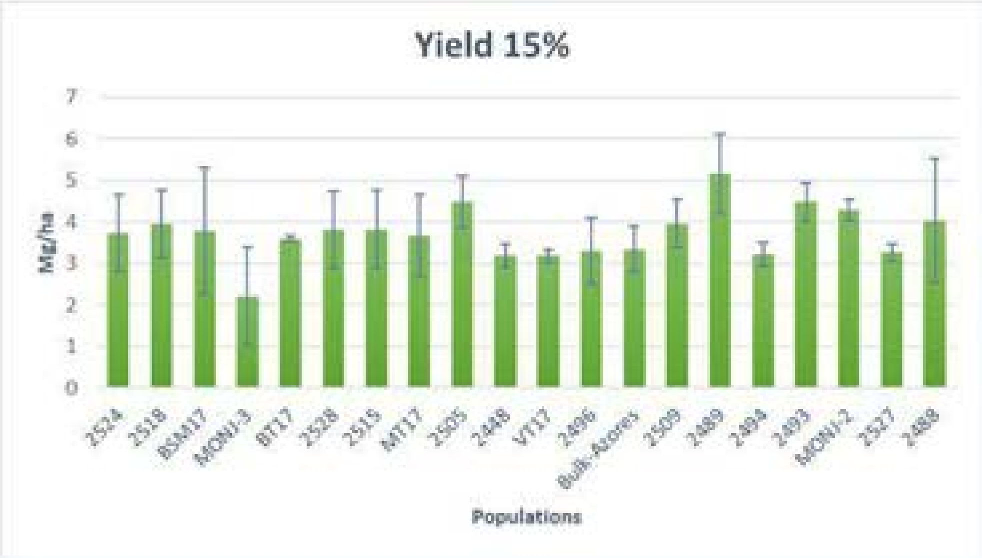

Figure 6 - Yield at 15% moisture per populations in 2019.

2020

From the tested genotypes three of them were in the top ten for both systems (Pg C0S0 19 - (Lousada 2019), 2499 and BulkAzores1).

Comparing the rank of the maize population across the conventional and organic low-input, it was observed a 33-range difference for Verdeal C0S0 17) and 0 for the 'Population' (2505). For CCPs the range interval did decrease, with a maximum of 10 (Sinpre) and a minimum of one (BulkAzores1). The most stable yield across the two systems were obtained by two Azorean populations (2505 and 2494) and one CCP (BulkAzores1).

The low input organic system resulted in significant lower yield compared to conventional system. The VA C0S0 19 - (Regadio Lousada) (4314 kg / ha), 2501 (4249 kg / ha) and CCP-BulkAzores2 (4145 kg / ha) had the higher yield and better adaptation to low input organic system.

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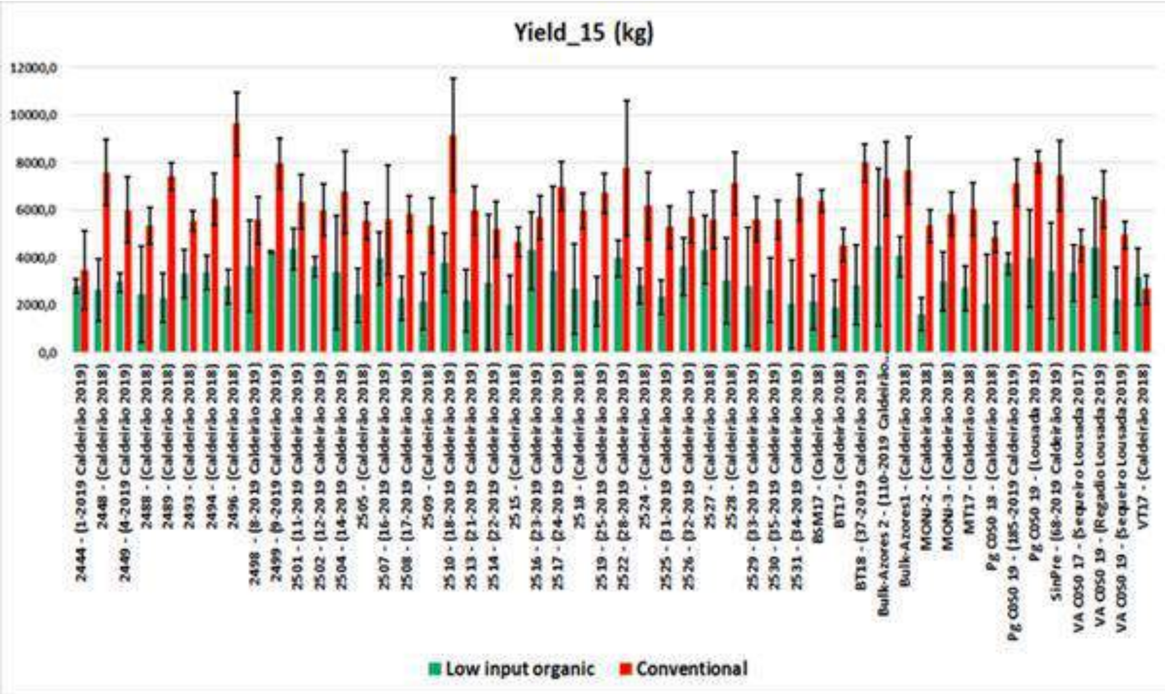

Figure 6. Yield in the Organic vs Conventional trials.

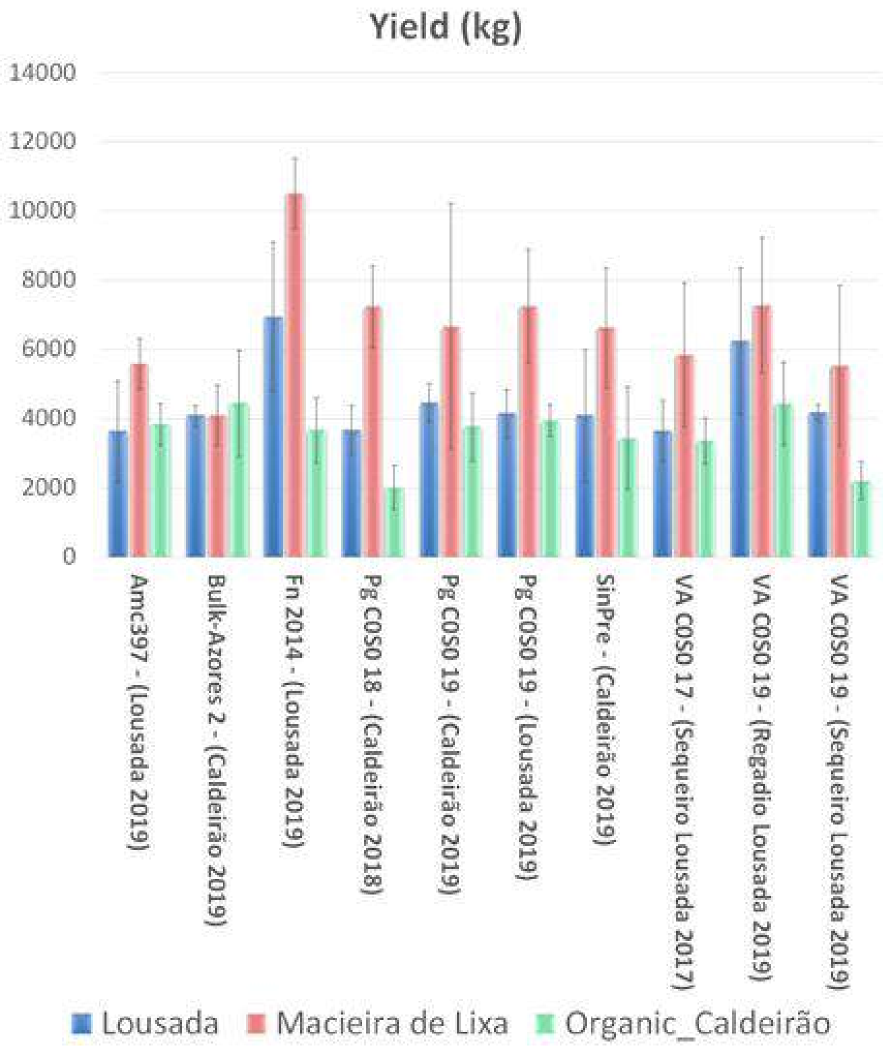

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Figure 7. Yield in Organic vs Conventional trial.

The tested populations didn't show significant phenotypic differences between the environments, although there were significant phenotypic differences among genotypes for Plant Height, Ear placement and Stalk lodging.
The yield ranged from 2.015 Mg/ha to 10.505 Mg/ha, with significant higher values in Macieira da Lixa. From the tested genotypes only VA C0S0 19 - (Regadio Lousada 2019) ranked in top 3 in all environments, being the highest yield in low input organic system (4.314 Mg/ha).
BulkAzores2 (CCP) obtain its higher yield in the low input organic system (4.145 Mg/ha).
AmC397 attain higher yield values in the low input organic system compared to conventional farming system in Lousada.
The population VA C0S0 19 - (Regadio Lousada 2019) showed organic potential and promising yield results across environments, despite not being chosen for gastronomic use or animal feed by farmers and stakeholders.

The exploitation of the Portuguese landrace as fresh maize indicated that the measures related to the color and texture were the attributes that most showed significant differences in consumers perception.

Outcomes and conclusions

2018
We were able to introduce in the field, long term cold storage populations. Populations that indicate some potential for yield and for agroecological and organic systems. In addition, they were used for its quality for food and adaptation to niche markets. These facts make these populations interesting for farmers, breeders, processors, and consumers under multi-actor approach.

2019
The CCPs had a good adaptation to all environments but tended to have a higher height and production values in his original environment (in organic), but also in conventional agriculture system. The performed trials indicate that the genetic resources are diverse and with yield potential. This indicates a good basis for participatory plant breeding, were seeds adapted to low-input organic agriculture and new niche markets and uses can be explored.

2020
Main outcomes:
Test a wide range of Portuguese maize populations (50) in low input organic conditions *versus* conventional system indicate the availability of germplasm to farmers aware of the many choices that can be made in favour of biodiversity and resilience of their ecosystem.
One traditional maize population and one CCP (composite cross populations) shown yield potential and stability (across environments) to be used in organic farming for gastronomic purposes and animal feeding.
· Meetings with farmers and stakeholders who provided the exchange of information and analysis of the potential of the germplasm tested in farmer fields and its possible adaptation to organic market.
Main lessons learned,
Traditional maize populations, due to grain quality traits that allow a wider range of products.
· It's important to share knowledge, keep farmers motivated and interested in germplasm evaluations to adapt their preferred populations to their systems.

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· Culinary breeding networks can be a way to promote underutilized maize landraces.

Publications

Duarte Manuel Fernandes Pintado (2019) Avaliação e Caracterização de Populações de Milho Açorianas – Implicações na sua Gestão Dinâmica e Valorização. Relatório de Estágio Profissionalizante de Mestrado em Engenharia Agro-Pecuária. Coimbra (Intern Report in MSc Agronomic and Livestock Engineering).

Dario Giglio (2019) Characterization and Evaluation of Open Pollinated Azorean Maize Landraces. Corso Di Laurea Magistrale In Scienze E Tecnologie Agrarie. Tesi Di Laurea Sperimentale. Universitá Degli Studi di Napoli Federico II. Dipartimento di Agraria

André Pereira, Duarte Pintado, Ana Neri, Pedro Mendes-Moreira (2021), Maize participatory breeding in Portugal - germplasm evaluation. Poster presentations in the EUCARPIALIVESEED conference - Breeding and seed sector innovations for organic food systems, March 8-10, 2021, Cēsis

André Pereira, Duarte Pintado, Ana Neri, Pedro Mendes-Moreira (2021), Evaluation of 50 Portuguese landraces, open-pollinated populations, and composites of maize (zea mays l.) In low input organic system versus conventional in Portugal. Poster presentations in the EUCARPIALIVESEED conference - Breeding and seed sector innovations for organic food systems, March 8-10, 2021, Cēsis

Felipe Hanower, Rosa Guilherme, João Noronha, André Pereira, Sara Correia, Valkiria Spring, Ana Neri, Pedro Mendes-Moreira (2021), Exploring Portuguese maizes landrace as fresh maize. Poster presentations in the EUCARPIALIVESEED conference - Breeding and seed sector innovations for organic food systems, March 8-10, 2021, Cēsis

Submitted and accepted abstract for poster presentation in the Organic World Congress 2021.
Pintado D, Giglio D, Joaquim C, Leitão R, Veloso M, Guilherme R, Mendes-Moreira P (2019) Evaluation and Characterization of Azorean Maize Landraces Enhancing the Genetic Diversity. Organic World Congress 2020: OWC2020-SCI-1336.

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

FiBL-CH - White lupin (*Lupinus albus*) composite cross population (CCP) (Trial #8)

Lead: FiBL-CH
Christine Arncken, Monika Messmer (FiBL-CH)

Abstract

Grain yield and tolerance to anthracnose disease was measured for three growing seasons in two locations in Switzerland on the F3 (2018), F4 (2019) and F5 (2020) generations of a white lupin CCP developed from crosses performed at FiBL in 2015.

Yield decreased in both locations from season to season, mainly due to the increasing anthracnose susceptibility in the population. Little information on anthracnose resistance of the parental lines was known at the moment of the crosses in 2015. To test the performance of a CCP specifically developed from anthracnose tolerant lines, a new CCP is being set-up at FiBL in 2021 from tolerant lines adapted for spring sowing and carrying the same mutation for low-alkaloid content. This new CCP will be monitored for yield, anthracnose tolerance and alkaloid content in the following growing seasons starting from 2022.

Introduction

Hypothesis and research question

Increased intra-specific diversity may support yield stability and decrease secondary infection of anthracnose. The objective of this activity was to test whether the use of a composite cross population can support the productivity of white lupin in Switzerland in conditions of spring-sowing and high anthracnose pressure.

Objectives

Test the performance of a composite cross population of white lupin in terms of yield production and susceptibility to anthracnose.

Material and Methods

Location(s)

- Switzerland, Kanton Aargau, High Rhine Valley
- Switzerland, Kanton Zürich, near Zürich lake

Creation of experimental population

FiBL white lupin CCP derives from 22 crossings between 14 parents performed in 2015. The CCP was developed based on the selection of 10 genotypes in the F1 and by mixing amounts of seed proportional to seed yield of each genotype. This was propagated in the field from 2016 to 2018 and subjected to grain selection before sowing 2019 and 2020.

Experimental design

For three growing seasons (2018-19-20) the CCP was tested in a cultivar trial, in two locations in Switzerland, each arranged in RCB design with two replicates in 2018 and three replicates in 2019 and 2020.

Evaluation

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In each year the susceptibility to anthracnose was scored at plot level and the grain yield measured.

Statistical methods

ANOVA and post-hoc test to compare the yield of the CCP to the average yield of the commercial cultivars in the cultivar trials at the same site and to the newly introduced and anthracnose tolerant cultivar Frida (2019-2020).

Results

In 2018, best cultivars at the Zurich Lake location were Sulimo, Amiga and the CCP (6,5, 5,5 and 5 t/ha respectively). In 2019, the CCP that was tested in the cultivar trial suffered severely from anthracnose, especially in the High Rhine valley location. The strict selection for sweet grains may have contributed to the reduced performance of the population, as the unselected plots yielded 0,45 t/ha more than the selected ones in Feldbach (data not shown).

In 2020, the CCP performed extremely poor, with only 0,43 t/ha in the High Rhine valley location (overall mean at this site was 1,49 t/ha) and 0,0617 t/ha at the Zurich Lake location (overall mean 0,625 t/ha).

Across the three seasons of the trials, and with a similar trend in both locations, anthracnose infestation increased and the CCP yield decreased (Figure 1). As the population is still evolving, future years will show whether the natural selection under high disease pressure will improve the population's resistance level.

Additionally, the introgression of bitter types into population made necessary a selection for sweet seeds each year after harvest.

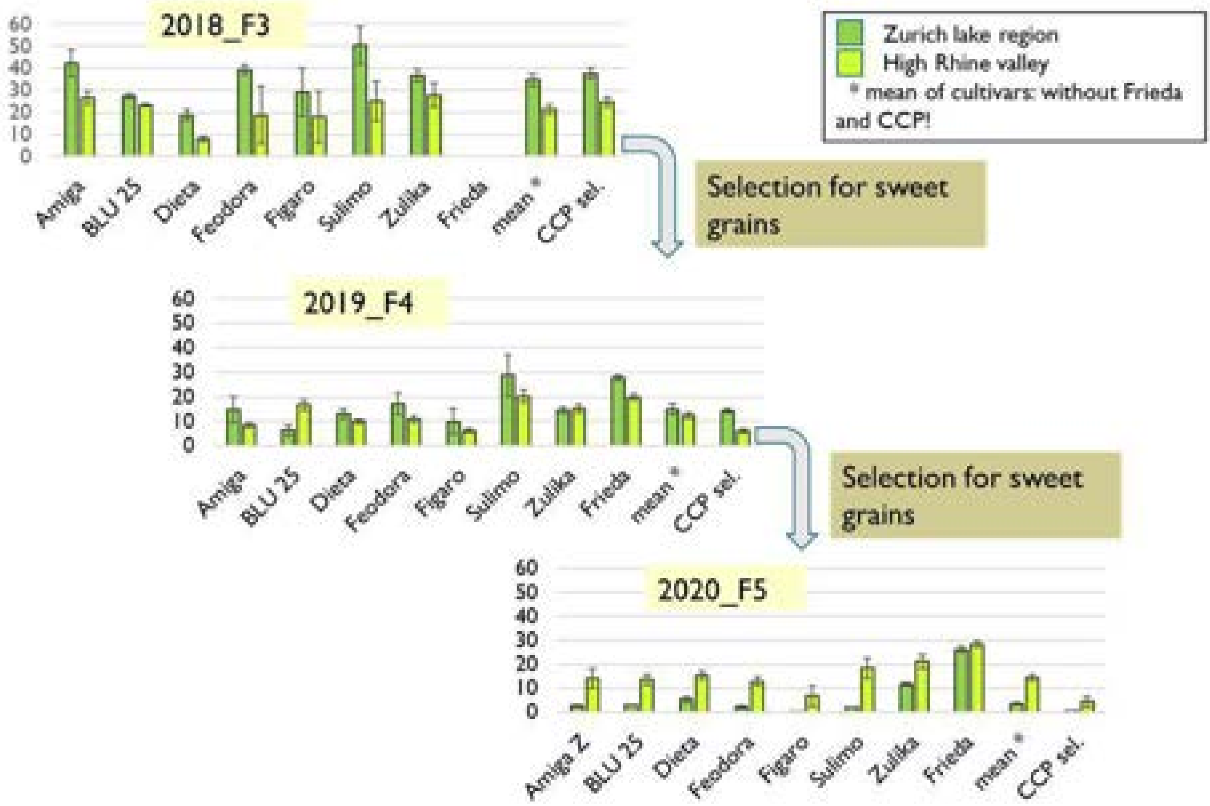

Figure 1. Yield (dt/ha) of CCP compared to cultivars in the same cultivar trial, two sites, 2018-2020. Bars represent the standard error.

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Discussion

The result of the CCP shows that resilience through genetic diversity can only be achieved in a genetic backgroun containing considerable resistance. Outcrossing with bitter plants and selection for sweet plants in 2018 and 2019 probably enhanced the population breakdown in 2020. However, we still think that the CCP concept is worth to be applied in white lupin if based on different parents.

This CCP derives from crosses effectuated in 2015 with little information on the anthracnose resistance of the parental lines, as the pre-breeding program on lupin started at FiBL-CH in 2014 and only 1 year of genetic resources screening was available at that point in time. A new CCP will be created from the most resistant and sweet F3 lines from FiBL pre-breeding program. It will be grown in isolation, and cross-pollination between the components will be supported by exposing them to a population of bumblebees. Furthermore, given the problematic introgression of bitter types into the population derived from the 2015 crosses, the new CCP will be developed from sweet parental lines with the same mutation for low-alkaloid content.

Main lessons learned

In contrast to CCPs developed for cereals in LIVESEED, we faced several challenges for white lupin as a strategy for fast adaptation to local conditions. The CCP of FiBL was created with limited prior knowledge on anthracnose resistance in the parental lines. Unfortunately, this CCP had no sufficient genetic diversity to allow for adaptation to spring sowing in Switzerland when infection pressure is high.

Thus, although this specific CCP did not support the objective to increase yield stability with the use of intra-specific diversity, we suppose that the results can be very different using a CCP developed from anthracnose tolerant parents and bearing a single major mutation for low-alkaloid content. The same hypothesis of this trial will be tested with a newly developed CCP that follows these guidelines.

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AREI – pea varieties mixture with spring wheat (Trial #9, #14 and#22)

Lead: Aina Kokare (AREI)

Abstract

Spring pea mixtures with wheat have been evaluated in field trials during 2017 – 2018 under organic conditions. The objectives are to compare pea and field beans performance in pure stand and with pea- support crop mixture under organic conditions. To compare the ranking of pea genotypes in mixture with support crop with the ranking in pure stand.

Spring pea mixtures with wheat have been evaluated in field trials 2019 under organic conditions. The objectives are to compare pea performance in pure stand and with pea- support crop mixture under organic conditions. To compare the ranking of pea genotypes in mixture with support crop with the ranking in pure stand. Under organic conditions semi leafless varieties grown in mixture took higher ranking positions in comparison to the pure stand, while opposite tendency was observed for leafed varieties.

Introduction

Objectives

The aim of the study is to compare how do the yield and yield components change when peas, beans and vetch are grown in pure stand and in grain legumes mixes.

Hypothesis and research question

Whether there are differences between leafed and semi leafless pea genotypes. Does the ranking of the pea genotypes change in pure stand compared to growing them in pea-wheat mix?

Material and Methods

Location(s)

Experiment was conducted in the experimental field of Priekuli research centre (AREI, Latvia) N 57˚ 18' 57'', E 25˚ 20' 19'', 123 m.

Genetic resources

Eleven pea genotypes, containing eight advanced varieties and three breeding lines were selected for this experiment (Table 1a and b).

Table 1a. selected genotypes of pea and wheat in 2017-18:

Accession name	**Country of origin**	**Description:** tallness, leafe type, flower coulor
Pisum sativum		
Bruno	Latvia	Advanced variety, medium tall, semileafless, pink colored
Clara	Sweden	Advanced variety, short, semileafless, white
Grisel	Portugal	Advanced variety, short, semileafless, white
H 03-05-04	Latvia	Breading line, medium tall, normal leaf, pink colored
H 08-10-9	Latvia	Breading line, medium tall, semileafless, pink colored

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H 86-19-3	Latvia	Breading line, medium tall, normal leaf, white
Kirke	Estonia	Advanced variety, short, normal leaf, pink colored
Leili	Estonia	Advanced variety, medium tall, semileafless, pink colored
Rebekka	Latvia	Advanced variety, short, normal leaf, white
Retrija	Latvia	Advanced variety, very tall, normal leaf, pink colored
Zaiga	Latvia	Advanced variety, tall, normal leaf, white
Vicia faba		
Lielplatones	Latvia	Local variety, very tall, late maturiting
Vertigo	Sweden	Advanced variety, medium tall
Vicia sativa		
Cēsu vietējie	Latvia	Local variety, very tall, late maturiting
Triticum aestivum		
Ufo	Latvia	Advanced variety, medium tall, medium early
Triticosecale		
Nilex	???	Advanced variety, medium tall, early

Table 1b. selected genotypes of pea and wheat in 2019

Accession name	Country of origin	Description: tallnes, leafe type, flower coulor
Pisum sativum		
Bruno	Latvia	Advanced variety, medium tall, semileafless, pink colored
Clara	Sweden	Advanced variety, short, semileafless, white
Grisel	Portugal	Advanced variety, short, semileafless, white
H 03-05-04	Latvia	Breading line, medium tall, normal leaf, pink colored
H 08-10-9	Latvia	Breading line, medium tall, semileafless, pink colored
H 86-19-3	Latvia	Breading line, medium tall, normal leaf, white
Kirke	Estonia	Advanced variety, short, normal leaf, pink colored
Leili	Estonia	Advanced variety, medium tall, semileafless, pink colored
Rebekka	Latvia	Advanced variety, short, normal leaf, white
Retrija	Latvia	Advanced variety, very tall, normal leaf, pink colored
Zaiga	Latvia	Advanced variety, tall, normal leaf, white
Triticum aestivum		
Korneto	Latvia	Advanced variety, medium tall, medium early
	Triticale	
Nilex		

Experimental design

The trials took place during 2017-19. The description of trial design and management is summarized in Table 2.

Table 2. Soil characteristics and crop management systems for trials performed under task 3.2.1. at Priekuli research center, AREI, Latvia)

	2017	2018	2019
Soil (plot) preparation before sowing	Ploughing autumn 2016.6.10, 28th April, 2017 cultivation two times before sowing, 6-8 cm deep.	Ploughing autumn 2017.18.10, 20th April, 2018 cultivation two times before sowing, 6-8 cm deep.	Ploughing autumn 2018, 22nd April, 2019 cultivation two times before sowing, 6-8 cm deep.
Sowing date	28th April, 2017	24th April, 2018	24th April, 2019

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Sowing depth		4-6 cm	4-6 cm	4-6 cm
Humus content (g kg-1)		2.1	1.5	2
P205 (mg kg-1)		156 mg kg-1	133 mg kg-1	164 mg kg-1
K20 (mg kg-1)		148 mg kg-1	69 mg kg-1	233 mg kg-1
pHKCl		5.5	5.3	6.2
Preceding crop		Winter rye for green manure	Potatoes	potatoes
Management of	Diseases and pests	none	none	none
	Weeds	25th May harrowing	Harrowing 9th May, 21th May, 29th May	7th, 15th, 24th May harrowing
Harvest date				Start 15.08

For the 2017-18 trials, plots size 10 m2, 3 replicates. Proportions in mixtures: pea: wheat (leafed pea type) 60:40, semileafless type 80:20. For faba beans: wheat/triticale 80:20, and vetch wheat/triticale 70:30. For the 2019 trials, plots size 10 m2, 3 replicates. Proportions in mixtures: pea: wheat (leafed pea type) 60:40, semileafless type 100:20. For faba beans: wheat/triticale 100:20, and vetch wheat/triticale 70:30.

Evaluation

The list of the traits collected in the trial Table 3-5.

Table 3. List of the traits collected for pea mixtures with wheat concerning crop development and agro-ecological performance

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Soil coverage SC	visual observation in scores 1- poor soil coverage, 9 complete soil coverage at the beginning of pea flowering stage (216-226 decimal code for the growth stages of peas by UPOV)	scores
Days to flowering (FLO).	Number of the days from sowing to the stage when 10 % of plants have begun to flower.	days
Days to full ripening (MAT)	Number of the days from sowing till full ripening were counted	days
Yield (Y)	Seed yield from each plot harvested, dried, and the grain yield weighted in kg. Then each component crop was separated, and the yield of each component was recorded. Yield in t/ha at 14% moisture content calculated.	t/ha
Plants: length (PLG)	Before harvest (Decimal code 320 for the growth stages of peas UPOV) 5 pea plants from each replicate randomly were taken. Plant length of five plants from the base to the top of plant measured and the mean has calculated.	cm
Number of fertile nodes (FNOD)	Nodes per 5 plants per replicate, which have leaves and pods counted, and the average fertile nodes calculated.	number

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Table 4. List of the traits collected for pea mixtures with wheat concerning crop productive performance

Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**
100 seed weight (100 SW)	Average weight of 2 samples of 100 randomly chosen seeds in grams was evaluated	g
Pods per plant (POD)	Counted number of pods per 5 plants and average pods per plant calculated	Number
Seed: average number of seeds per plant (SEED)	Seed were counted of five randomly chosen plants from replicate and average number of seeds per plant calculated.	number

Table 5. List of the traits collected for pea mixtures with wheat crop quality performance.

Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		
Trait	**How it has been assessed**	**Type of data available**
Protein content (PRO)	Representative sample from legumes was measured by FOSS XDS Rapid Content Analyzer. The cereal grains were analysed by NearInfrared Transmittance Infratec 1241 Analyser (Foss, Högenäs, Sweden)	%

Participatory/multi-actor approach

The trial was demonstrated to farmers and producers in annual ''Field Days''. The organic farmers were interested in pea growing in pure stand under organic conditions. On October 8, 2018, we took part in the meeting with company ALOJA-STARKELSEN that is the biggest potato starch producer in the Baltic States. ALOJA-STARKELSEN is interested in plant based organic proteins production from pea and field beans. During the meeting, we discussed the choice of pea and beans varieties for protein production purposes and the legumes management practice in organic farming. Since company focused on gluten-free product, then the option for pea growing in pure stand were discussed.

Statistical methods

Analysis of variance was performed using Genstat 14.0 (VSN International, 2011). The consistency of ranking of the genotypes between pure stand mixture was assessed with Spearman`s rank correlation coefficient. Statistical significance was assessed at the 95% confidence level (a = 0.05).

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Results

Results of trials in 2017-18

Both years of the study characterized by contrasting weather conditions. In 2017 the growing season in Priekuli was characterized by cool weather and with large amount of precipitation at the end of April, in June and in July, while in 2018 the growth period was hot and drier than normal. In 2018 the average pea yield was significantly higher in both growing methods (in pure stand and in mixture with wheat) in comparison to 2017 (Table 6). The year significant influenced the crop development. Differences in yield level between leaves type (normal leaves and semileafless type) were more pronounced in 2018 than that in 2017 grown in both methods. The yield was higher in 2018 in comparison to 2017. The semileafless varieties performed better than leafed ones in pure stand and mixture. In 2017 when yield level was extremely low for both leaves type of pea varieties, the leafed type varieties significantly decreased yield components such as FNOD, POD and SEED in comparison to semileafless type when they were grown in mixture. However, in 2018 significant decrease in yield and yield components was observed for semileafless varieties grown in mixture. For semileafless varieties grown in mixture the decrease in yield was 52% in comparison to pure stand, while for leafed type it was 47%. Climatic conditions influenced protein content in pea, and it depended on pea growing method as well. In 2017, both types (leafed and semileafless) of pea varieties grown in mixture had higher protein content in seeds in comparison to the growing in pure stand, but it was not observed in 2018.

Table 6. The average soil coverage (SC, scores), days to flowering (FLO), days to full ripening (MAT), yield (Y, t/ha), plant length (PLG, cm), number of fertile nodes (FNOD), 100 seed weight (100 SW), pods per plant (POD) and average number of seed per plant (SEED) and protein content in seeds (PRO,%) of six leafed and five semileafless pea genotypes grown at organic site in pure stand (T) and in mix with wheat (M), 2017–2018.

Trait	Year		Normal leaves	Semileafless
SC	2017	M	6.56***[1]	6.13***
		T	5.11	3.87
	2018	M	6.17***	5.83***
		T	4.33	4.07
FLO	2017	M	62	64.
		T	62	63.
	2018	M	51	53
		T	51	53
MAT	2017	M	114	116
		T	113	115
	2018	M	95	94
		T	95	94
Y	2017	M	0.117***	0.124***
		T	0.381	0.303
	2018	M	0,781***	0.93***

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		T	1,481	1.92
PLG	2017	M	45.5	40.2*
		T	45.9	37.2
	2018	M	66.0	61.9**
		T	69.5	75.0
FNOD	2017	M	1.4*	1.3
		T	1.7	1.3
	2018	M	4.4	3.9**
		T	4.4	5.9
100 SW	2017	M	20.5	20.2
		T	20.5	19.2
	2018	M	25.2	24.7
		T	25.1	24.8
POD	2017	M	1.6*	1.4
		T	2.0	1.4
	2018	M	6,5	5.0*
		T	6,2	6.9
SEED	2017	M	4.4*	5.7
		T	5.8	4.5
	2018	M	21.0	22.8*
		T	20.5	35.2
PRO	2017	M	20.45**	20.48**
		T	19.45	19.43
	2018	M	23.38	22.56
		T	23.65	22.46

[1]* * significantly different from pure stand at* *P* = 0.05, ** significant at *P* = 0.01, ***significant at *P*= 0.001

Between the two growing methods (in pure stand and mixture with spring wheat), leafed varieties showed the greatest difference in yield rank order with exception of Retrija, which took the highest-ranking position in both years of testing (Fig.1). The rank changes of semileafless varieties for yield between pure stand and mixture were less pronounced in comparison to leafed varieties. Semileafless varieties Bruno, breeding line H 08-10-9 and Leili took the highest-ranking position for yield either grown in pure stand or in pea-wheat mix in both years of observations (Fig. 2).

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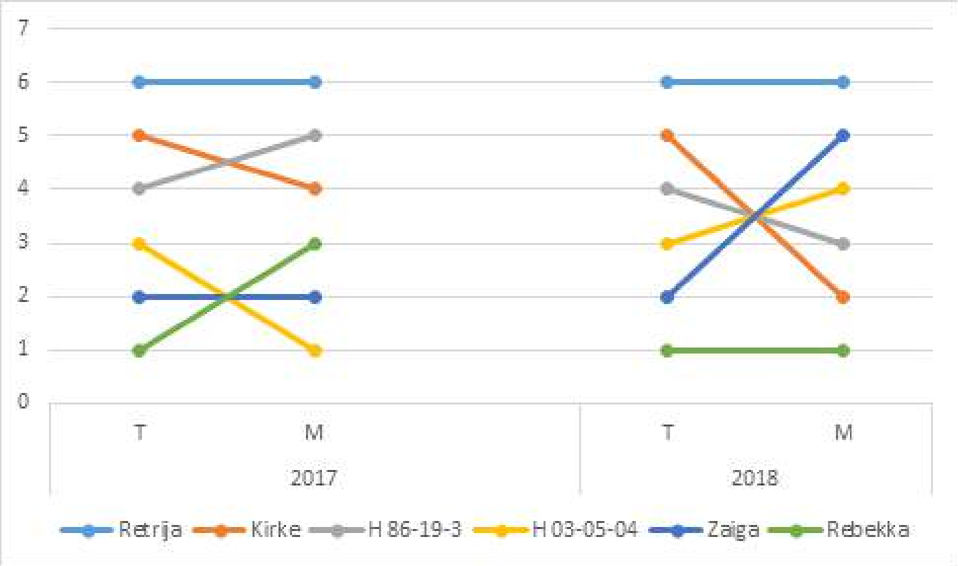

Figure 1. The ranking order of leafed varieties with respect to their mean yield between the growing type (pure stand T and mixture M) in 2017 and 2018. (Higher value of ranking higher yield level)

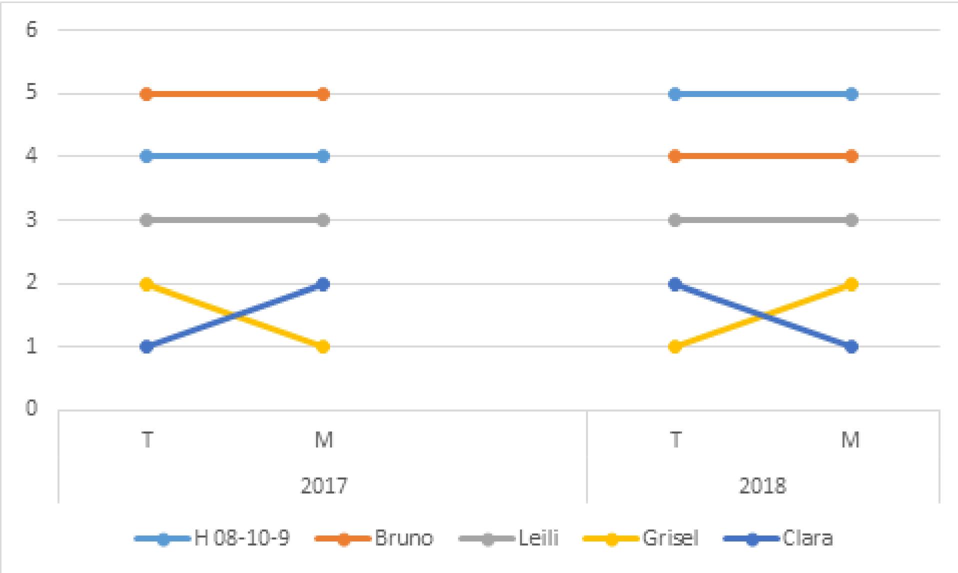

Figure 2. The ranking order of semi leafless varieties with respect to their mean yield between the growing type (pure stand T and mixture M) in 2017 and 2018.

In 2019, the growing season in Priekuli was characterized by warm weather and with large amount of précipitation in the middle of June and at the beginning of July. The average pea yield was significantly higher in pure stand than in mixture with wheat for leafed genotypes, but no differences were found for semi leafless varieties in 2019 (Table 7). The soil coverage increased significantly of semi leafless genotypes when they were grown in mixture. When growing in mixture, the plant productivity elements such as the number of fertile nodes, number of pods and the seeds number per plant decreased significantly for both type (leaved and semi leafless) of pea genotypes. The 1000 seed weight was higher in mixture in comparison with pure stand for leafed genotypes while no differences were observed for semi leafless genotypes. No differences were observed for protein content between mixture and pure stand for both pea types.

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The rank changes of semi leafless varieties for yield between pure stand and mixture were less pronounced in comparison to leafed varieties in 2019 (Fig 3). Semi leafless varieties Bruno, breeding line H 08-10-9 and Leili took the highest-ranking position for yield either grown in pure stand or in pea-wheat mixture. The rank position for leafed varieties grown in mixture had tendency to decrease in comparison to pure stand. Leafed variety Kirke took slightly higher-ranking position grown in mixture compared to pure stand, while Retrija which took the highest rank in pure stand ranked lower in mixture.

Table 7. The performance of six leafed and five semi leafless pea genotypes grown at organic site in pure stand (T) and in mix with wheat (M), in 2019.

Traits	Normal leaves		Semi leafless	
	T	M	T	M
Yield, t ha^{-1}	1,92***[1]	1,17	1,98	1,87
SC, scores	7,1	6,9	7	7,4*
FLO, days	55	56	57	57
MAT, days	110,0	112**	110	113***
PLG, cm	84,5***	74,0	70,7**	64,0
FNOD, number	6,2**	5,1	4,4**	3,6
100 SW, gr	23,9	25,0**	24,6	23,9
POD, number	6,4*	5,1	4,7*	3,5
SEED, number	23,2**	17,7	20**	15,0
PROT, %	22,7	22,8	22,8	22,3

[1] * significantly different from pure stand at* P = 0.05. ** significant at P = 0.01, ***significant at P= 0.001

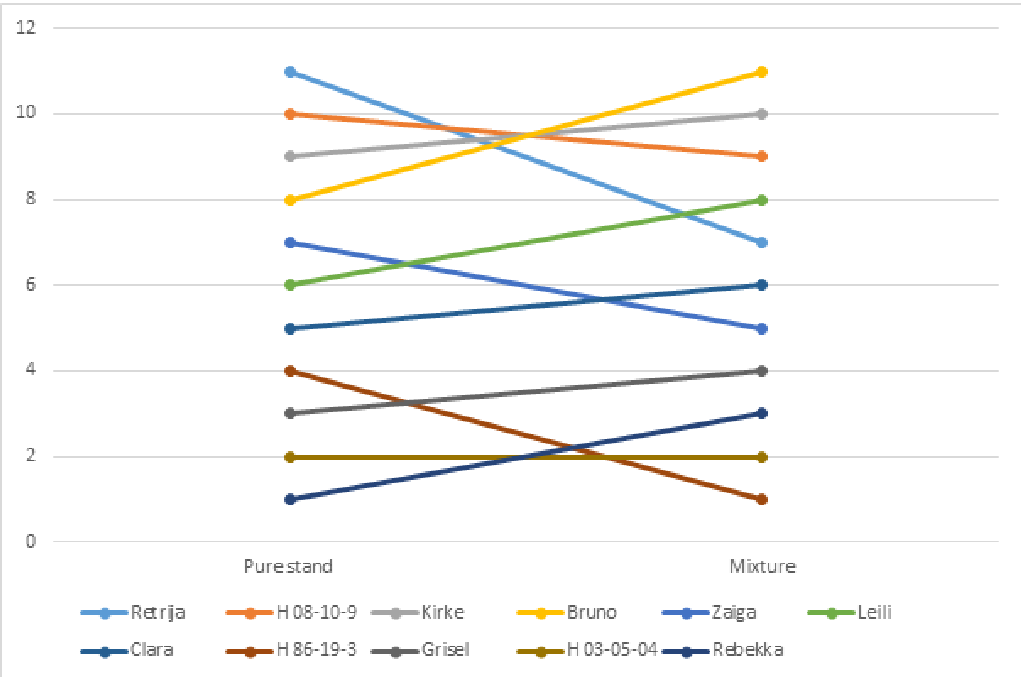

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Figure 3. The ranking order of varieties with respect to their mean yield between the growing method (pure stand T and mixture M) in 2019 (Higher value of ranking means higher yield level).

Field beans grown over three years

Two field bean varieties were grown in mixture with wheat and spring triticale, to find out which of spring cereal would be the best crop for growing field beans in the mixture to enhance the soil cover and achieve the high yield in organic farming system.

Results showed the higher soil coverage was obtained when field beans were grown in mixture with cereals than in pure stand.

However, the yield level of beans grown in mixture with cereals was in average 21% lower in comparison to pure stand. The choice of cereal species for growing in mixture with field beans had no significant impact on the yield level of the beans.

Spring vetch

The spring vetch was grown in mixture with wheat, spring triticale and field beans, to find out which of the support crops would be the best to achieve the high yield in organic farming system. The results showed, that in two years, the highest seed yield of spring vetch was obtained in mixture with field beans.

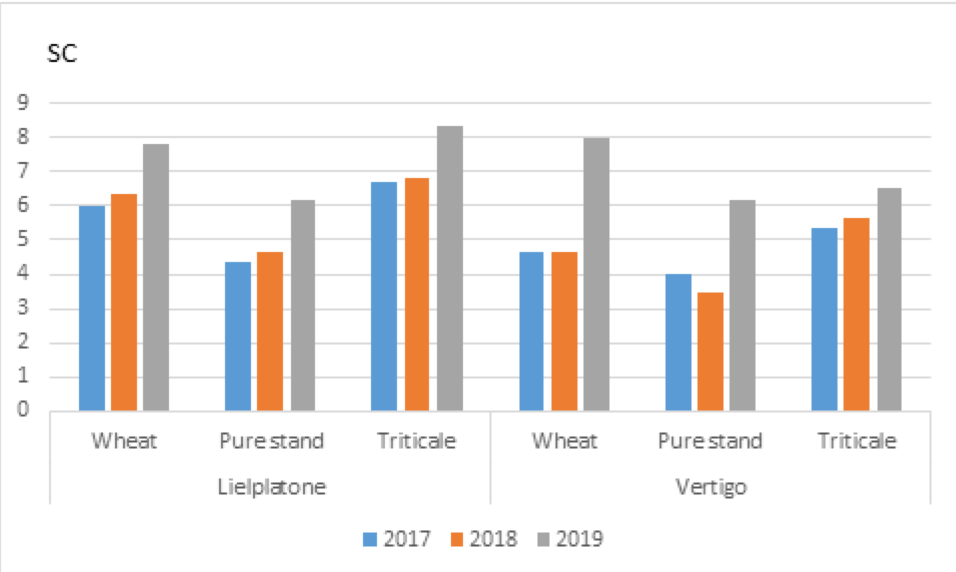

Figure 4. The soil coverage (SC in scores 1 -poor soil coverage, 9 – very high soil coverage) of two field bean varieties grown in mixture with spring wheat and triticale in three years (from 2017 to 2019)

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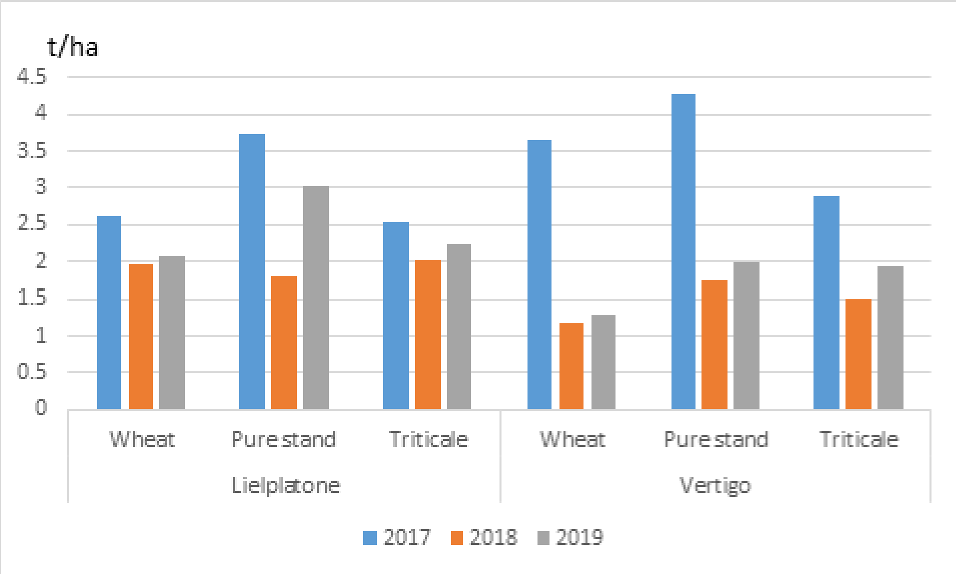

Figure 5. The yield (tha-1) of two field bean varieties grown in pure stand and in the mixture with spring wheat and triticale in three years (from 2017 to 2019)

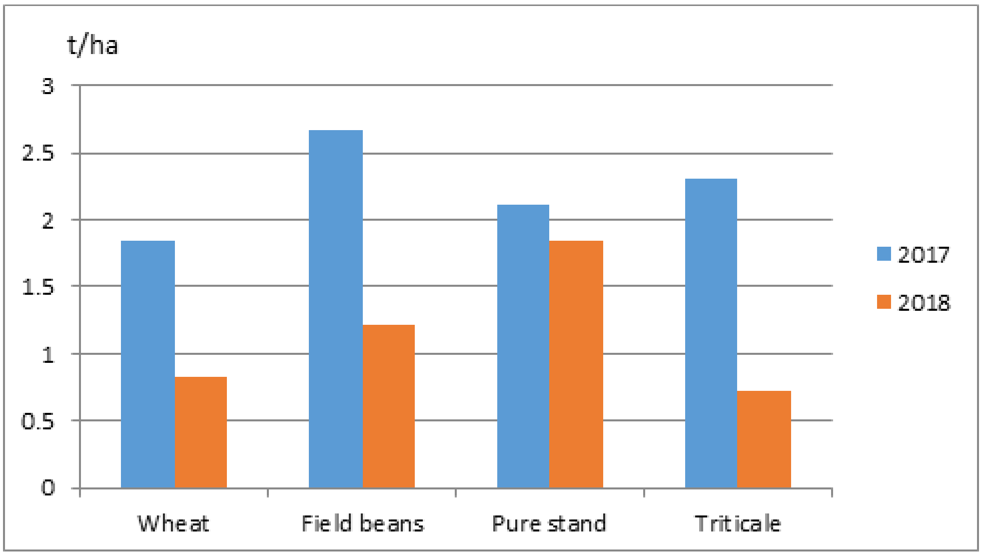

Figure 6. The yield (t ha-1) of spring vetch grown in pure stand and in the mixture with spring wheat, triticale and field beans in two years (from 2017 to 2018)

Discussion

Results from 2017-18

In Latvia pea was used mainly for food and feed purposes. In organic farming, leafed pea type is more attractive due to its ability to produce high green mass. These varieties have advantage in green fodder production in comparison to semileafless varieties which are mainly used for grain production. The leafed varieties had good weed suppression ability; however, they have poor lodging resistance. That is reason why farmers used to grow leafed varieties in mixture with support crop. With the growing interest of processors in organically grown peas, the demand for farmers for organically produced seeds is also increasing. The Semileafless varieties are more attractive to farmers for their grain yield, good lodging resistance what allows to grow them in pure stand, however, the biggest challenge is

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weed management. Results showed that by adding wheat to the semileafless pea, the soil coverage has increased significantly that may help to deal with weeds in organic fields. Depending on weather conditions, yield components (FNOD, POD, SEED) has tendency to decrease in the mixture that may influence the yield level of pea, especially for semileafless varieties. The ranking of varieties for yield showed that leafless varieties with the highest yield in pure stand may also yield well in mixtures.

Results from 2019

Without reduction the sowing rate of peas, but the adding 20% of wheat to the sowing rate of the semi leafless genotypes, the grain yield did not decrease. Besides the soil coverage significantly increased which was highly beneficial for semi leafless genotypes, it could help to withstand with high weed pressure. This could help to increase the production of organic pea, hope with weed pressure and in addition to improve lodging resistance of crop in unfavourable conditions. The ranking of varieties for yield showed that leafless varieties with the highest yield in pure stand may also yield well in mixtures.

In organic farming leafed pea type is more attractive due to its ability to produce high green mass. These varieties have an advantage in green fodder production in comparison to semi leafless varieties which are mainly used for grain production. The leafed varieties have good weed suppression ability; however they tend to lodge, which makes it very difficult to harvest and causes great losses at harvest. That is reason why farmers used to grow leafed varieties mainly in mixture with the support crop. The ranking of leafed varieties showed that there are differences in the ranking between varieties. The genotypes which took the highest-ranking position grown in pure stand, tend to have low rank when grown in mixture. This tendency was observed mainly for tall and most productive genotypes such as Retrija and Zaiga. Besides, for short genotypes with low yield potential the differences in rank position were less pronounced.

Outcomes and conclusions

The 2017-18 results showed that weather conditions had significant influence on pea yield grown in pure stand and in mixture. The cool and humid conditions favoured to the growth of leafed varieties, while in more hot and dry weather conditions semileafless varieties performed better than leafed ones. However, when cultivated in a mixture, the semileafless varieties may significantly reduce number of productive nodes, pods ans numbed of seeds, that could result in decrease in yield level.

The ranking of varieties in the 2017-2019 trials showed that under organic conditions, those semileafless varieties which have high yield in pure stand will also yield high in mixture.

The adding of 20% of spring wheat to the sowing rate of semileafless pea genotypes, without the reduction of pea sowing rate, could be a viable option for growing of semileafless varieties under organic conditions. That would allow to increase weed suppressive ability by better soil coverage and to maintain the high productivity of pea.

The growing field beans in mixture with spring cereals enhances the soil coverage and could help to hope with weeds pressure. The species of the support crop did not have an impact on yield level of field beans.

The field beans could be the best solution for spring vetch growing in mixture, to provide high seed yield under organic conditions.

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Main outcomes

The semi leafless pea genotypes which ranked high for GY in the pure stand, also ranked high when grown in a mixture with wheat. This finding allows the selection of semi-leafless genotypes to perform in a mixture within an organic breeding program and recommends the most suitable genotypes for a mix with spring wheat to the farmers for growing under organic conditions.

Leafed tall pea genotypes are more suitable for growing in a mixture with wheat under organic conditions than short genotypes.

Main lessons learned

- Leafed short genotypes are more sensitive and respond differently to the cultivation in a mixture with spring wheat than the tall genotypes.
- The decrease in yield components was more pronounced for the short genotypes than for tall grown in the mixture.
- We observed that some genotypes are more vigorous after germination than other. A trait such as early vigor may be useful for pea genotype selection for mixed cultivation under organic conditions

How had the resilience improved?

Mixed cropping allows to cope with weed pressure, and in addition to improve the lodging resistance of crop in unfavorable conditions.

How had the resilience improved?

Mixed cropping allows to cope with weed pressure, and in addition to improve the lodging resistance of crop in unfavorable conditions.

How information was transferred to the other situations

Information about pea mixtures recommended to organic pea seed producers and for the farmers growing peas for industrial plant-based organic proteins production
Recommendations were prepared for farmers and the results of the study on pea mixtures were presented for legumes growers in e-field days.

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GZPK - Summer pea - spring triticale – screening of pea gene bank accessions (Trial #10)

Lead: Getreidezüchtung Peter Kunz – Switzerland (LTP of FiBL-CH)
Christine Scheiner c.scheiner@gzpk.ch

Abstract

20 pea accessions from European gene banks were evaluated for intercropping with spring triticale (variety "Mamut") with the goal to find interesting traits to introduce in the GZPK summer pea breeding program. After first evaluation and seed multiplication in 2017, trials were sown 2018, in 1,4m² plots on 2 sites. Due to the very dry growing season, peas have shown very few diseases. Yields were very low due to the strong competition with triticale. Some interesting traits were noticed: high protein content ("Merveille d'Étampes" 24,4 %), late flowering and maturity (Revermont Petit Pois", "Merveille d'Étampes", "Century"), drought resistance ("Arthur", "Revermont Petit Pois", "Merveille d'Étampes", "Century").

In 2020, 9 newly ordered pea accessions from European gene banks were evaluated together with 11 accessions from the previous years for intercropping with spring barley (variety mixture of 1/3 each "KWS Beckie", "KWS Atrika", "Sydney"). The goal was to find interesting traits to introduce in the gzpk summer pea breeding program and multiplicate promising lines from previous year. In the third year 2020 trials were sown in 1.4m² plots on 2 sites. Overall yields showed great variation between accessions, protein content was overall comparatively low. Interesting traits were noticed: high protein content "Procumbens-pro (1968)" 28.3%, extremely early flowering "Pisum sativum 75121".

Introduction

Location(s)

The 20 microplots for seed multiplication were sawn in March 2017 near CH-8225 **Siblingen**, canton Schaffhausen in northern Switzerland after maize (47°42'40.2"N 8°30'37.2"E). The trials with 20 plots of 1,37 m^2 were sawn in March 2018 in two locations (CH-8610 **Uster**, canton Zürich, Swiss Plateau, 47°19'22.8"N 8°42'44.0"E, previous crop maize; CH-8477 **Oberstammheim**, canton Zürich, Züricher Weinland, 47°37'53.7"N 8°48'40.9"E, previous crop maize)

The trials with 20 plots of 1.4m^2 were sown end of March 2020 in two locations (CH-8610 Uster, canton Zürich, Swiss Plateau, 47°19'08.3"N 8°43'13.3"E, previous crop maize; CH-8477 Oberstammheim, canton Zürich, Züricher Weinland, 47°38'00.5"N 8°48'49.9"E, previous crop maize and green manure over winter).

Hypothesis and research question

Peas have a high value for crop rotations especially in organic agriculture. By growing peas in mixture with cereals, a reduction regarding lodging and competition against weeds can be reached. The cultivated pea area in Switzerland nevertheless is still small, caused by the lack of varieties with stable yields and resistances against soil borne diseases. The idea of this trial originated by the need of enlarging the narrow gene pool from where modern pea varieties are being derived from. The reason is to describe accessions and landraces from different gene banks to find interesting traits to introduce

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in the GZPK breeding programme to enhance resilience of this crop and identify interesting varieties for special uses (“Nischensorte”).

Material and Methods

Genetic resources

Pisum sativum L. subsp. sativum convar. medullare Accessionname: “Revermont Petit Pois ”
Genbank Changins (Schweiz)
Pisum sativum L. subsp. sativum convar. medullare Accessionname:”Tante Emma”
Genbank Changins (Schweiz)
Pisum sativum L. subsp. sativum convar. axiphiumAccessionname:”Wintererbse Greifensee”
Genbank Changins (Schweiz)
Pisum sativum L. subsp. sativum convar. medullare Accessionname:“Lötschentaler”
Genbank Changins (Schweiz)
Pisum sativum L. Accessionname:”Montana”
Genbank der Universität Wageningen (Niederlande)
Pisum sativum L. subsp. sativum convar. sativumAccessionname: “Century (DE)”
IPK Genbank Gartersleben (Deutschland)
Pisum sativum L. subsp. sativum convar. sativumAccessionname: “Delwiche Scotch Green“
IPK Genbank Gartersleben (Deutschland)
Pisum sativum L. subsp. sativum convar. sativum Accessionname: “Erbse (PIS 5122)”
IPK Genbank Gartersleben (Deutschland)
Pisum sativum L. subsp. sativum convar. sativum Accessionname : “Merveille d'Étampes”
IPK Genbank Gartersleben (Deutschland)
Pisum sativum L. subsp. sativum convar. sativumAccessionname : “Orgueil du Marché”
IPK Genbank Gartersleben (Deutschland)
Pisum sativum L. subsp. sativum convar. sativumAccessionname: “Triomphe de Maninet”
IPK Genbank Gartersleben (Deutschland)
Pisum sativum L. Accessionname:"Arthur"
Genbank KCRZG (Polen)
Pisum sativum L. Accessionname:"Belmon"
Genbank KCRZG (Polen)
Pisum sativum L. Accessionname:"Century (PL)"
Genbank KCRZG (Polen)
Pisum sativum L. Accessionname:”Dunkelgruen Mai“
Genbank KCRZG (Polen)
Pisum sativum L. subsp. elatiusAccessionname:“77727“
Genbank KCRZG (Polen)
Pisum sativum L. subsp. elatius Accessionname:“75742“
Genbank KCRZG (Polen)
Pisum sativum L. subsp. elatius Accessionname:”77398”
Genbank KCRZG (Polen)
Pisum sativum L. subsp. abyssinsicum Accessionname:“75559”
Genbank KCRZG (Polen)
Pisum sativum L. subsp. abyssinsicum Accessionname:“77386“
Genbank KCRZG (Polen)

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2020

List of accessions

Include their origin and any relevant background information available – (add additional information in Data file (excel spreadsheet) as supplementary material if too long)

IPK Gene Bank Gatersleben (Germany):

- *Pisum sativum L*.: **“Merveille d´Étampes”**
- *Pisum sativum L. subsp. sativum convar. Sativum var. cimitari Alef*.: **“Feltham First Rogue”**

Gene Bank KCRZG (Poland):

- *Pisum sativum L*.: **“Century”**
- *Pisum sativum L*.: **“P. sativum Arthur”**
- *Pisum sativum L*.: **“P. sativum 75121”**

Gene Bank of the University of Wageningen (Netherlands):

- *Pisum sativum L.:* **“Lamm 31; JI 12; L 31”**
- *Pisum sativum L.:* **“Montana”**
- *Pisum sativum L.:* **“P. sativum Turkey (1971)”**

John Innes Centre (United Kingdom):

- *Pisum sativum L.:* **“Nike (1969)”**
- *Pisum sativum L.:* **“Dwarf Sugar (1969)”**
- *Pisum sativum L.:* **“Recette (C-4) (1969)”**
- *Pisum sativum L.:* **“Kronenerbse (1967)”**
- *Pisum sativum L.:* **“Keerau Pea (1961)”**
- *Pisum sativum L.:* **“Filigreen-af (1983)”**
- *Pisum sativum L.:* **“Densinodosum-lkb (2005)”**
- *Pisum sativum L.:* **“Acacia Leaved Purple Dwarf”**
- *Pisum sativum L.:* **“Filby-af,st (1983)”**
- *Pisum sativum L.:* **“Procumbens - pro (1968)”**
- *Pisum sativum L.:* **“Lamm 30 (1964)”**

National Gene Bank of Agroscope in Changins (Switzerland):

- *Pisum sativum L. :* **“Revermont Petit pois”**

Experimental design

2017-18

The 20 micro plots for seed multiplication were sown mid-March 2017 in Siblingen and harvested mid-July. The 20 accessions were sown in mixture with triticale (variety “Mamut”) on two sites in 2018, in Oberstammheim on 25.03.2018 and in Uster ZH on 09.04.2018, in 1,37m² plots. The sowing density was 80 seeds/ m² (pea), and 180 seeds/m² (triticale). The trails were harvested on 12.07.2018 in Oberstammheim and on 15.07.2018 in Uster ZH. Because our trial was a screening-trial, we use no special experimental design and do no statistical analyses.

2020

The 20 accessions were sown in mixture with spring barley (variety mixture with 1/3 each “KWS Beckie”, “KWS Atrika”, “Sydney”) on two sites in 2020, in Oberstammheim ZH on 18.03.2020 and in Uster ZH on 19.03.2020, in 1.4m² plots. The sowing density was 120 seeds/plot (pea), and 161 seeds/plot (barley). Flowering started between 07.05.2020 and 10.06.2020, while end of flowering happened between 22.05.2020 and 20.06.2020. Trails in Oberstammheim were harvested on 21.07.2020 and in Uster ZH on 14.07.2020.

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Evaluation

List of the traits collected in the trial in Table 1-3. The traits were **collected for pea** (not for triticale n 2017-18-19 or for barley in 2020).

Crop development and agro-ecological performance (phenology, weeds, diseases, …)			
2018-2019-2020			
Trait	**How it has been assessed**		**Type of data available**
Beginning of flowering	Observation		Date
End of flowering	Observation		Date
Plant length at beginning of flowering	Measured	Length in cm	
Plant length at end of flowering	Measured	Length in cm	
Lodging at beginning of flowering	Scoring in relation of each other	1=no lodging, 9=full lodging	
Lodging at end of flowering	Scoring in relation of each other	1=no lodging 9=full lodging	

Table 1. Crop development and agro-ecological performance traits

Crop productive performance (yield, yield components)		
2017-19		
Trait	**How it has been assessed**	**Type of data available**
Total yield of combination	Weighing	dt/ha
Yield pea	Weighing	dt/ha
Yield triticale	Weighing	dt/ha
Thousand Grain Weight pea	Counting and weighing	g
2020		
Yield (barley) replaced the triticale	Weighing	dt/ha

Table 2. Crop productive performance traits

Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		

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2017-2020		
Trait	**How it has been assessed**	**Type of data available**
Protein content of pea (NIR)	Near Infrared Spectroscopy (NIRS)	Estimated in %

Table 3. Crop quality performance traits

Results

Table 4 shows the yield results from 2018-19, for pea and triticale, while Figure 1 shows the results for 2020, for pea and barley.

Variety	Uster				Oberstammheim			
	Yield peas (dt/ha)	Yield Triticale (dt/ha)	Yield total (dt/ha)	Protein Peas (%dm)	Yield peas (dt/ha)	Yield Triticale (dt/ha)	Yield total (dt/ha)	Protein Peas (%dm)
Revermont Petit pois	13.97	5.98	19.95	19.47	12.33	27.43	39.77	22.05
Tante Emma	9.04	7.93	16.97	17.62	8.14	29.57	37.71	16.76
Wintererbse Greifensee	7.34	6.60	13.94	18.85	8.31	30.40	38.71	19.70
Lötschentaler	9.14	8.32	17.46	18.55	7.11	25.25	32.36	20.06
Montana	7.94	9.74	17.68	19.00	2.79	27.99	30.78	*
Century (DE)	33.83	2.52	36.36	21.73	9.76	29.67	39.43	19.70
Delwiche Scotch Green	24.40	3.86	28.26	21.83	2.69	28.14	30.83	*
Erbse (PIS 5122)	3.67	7.75	11.41	*	3.62	22.86	26.48	*
Merveille d'Étampes	18.49	2.67	21.16	24.11	8.68	27.64	36.32	23.10
Orgueil du Marché	8.16	11.78	19.94	19.20	0.63	18.48	19.11	*
Triomphe de Maninet	1.36	10.78	12.14	*	0.00	19.39	19.39	*
Arthur	15.17	9.06	24.22	17.78	16.01	29.64	45.64	19.52
Belmon	16.05	5.93	21.99	18.96	4.34	16.45	20.79	*
Century (PL)	30.46	5.23	35.69	22.46	11.73	21.88	33.61	20.90
Dunkelgruen Mai	14.99	7.22	22.20	22.78	4.17	20.65	24.82	*
77727	0.00	12.49	12.49	*	0.00	18.72	18.72	*
75742	2.54	10.12	12.66	*	0.73	21.46	22.19	*
77398	3.66	7.72	11.38	*	0.00	24.91	24.91	*
75559	0.00	10.11	10.11	*	0.00	21.16	21.16	*
77386	9.99	10.12	20.10	22.58	2.37	24.35	26.72	*

**Yield too low for protein measurement.*

Table 4. Results of yield 2018-19, pea and triticale

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

Nr.	Accession name	Uster					Oberstammheim				
		Yield peas (dt/ha)	Yield Barley (dt/ha)	Yield total (dt/ha)	Protein Peas (%dm)	TKM Peas	Yield peas (dt/ha)	Yield Barley (dt/ha)	Yield total (dt/ha)	Protein Peas (%dm)	TKM Peas
1	P.sativum-Turkey (1971)	34.7	17.6	53.8	18.5	234.0	8.7	26.8	36.8	19.7	238.2
2	Filigreen-af (1983)	0.9	18.7	21.1	*	100.0	0.1	35.9	37.3	*	*
3	Densinodosum-lkb (2005)	6.5	18.5	26.5	23.7	179.5	2.9	51.6	55.8	26.9	206.9
4	Aca. Leavd Purple Dwarf	2.4	14.5	18.5	*	205.6	0.1	39.9	41.8	*	*
5	Filby-af,st (1983) *	3.2	11.4	16.1	*	189.8	1.1	44.1	47.2	*	*
6	Procumbens - pro (1968)	4.3	18.4	24.2	29.4	248.5	5.0	50.6	57.7	27.2	275.0
7	Lamm 30 (1964)	4.0	12.4	17.8	20.9	194.0	2.6	40.1	45.0	22.5	*
8	Keerau Pea (1961)	0.1	13.7	15.4	*	*	0.0	38.1	39.9	*	*
9	Kronenerbse (1967) *	13.9	16.5	31.9	22.3	216.4	17.1	44.1	63.5	20.9	275.4
10	Recette (C-4) (1969)	3.2	15.3	20.1	22.7	113	2.2	29.7	33.2	*	130.0
11	Montana *	6.5	26.7	34.7	16.8	215	5.7	35.0	41.5	20.6	105.6
12	Feltham First Rogue *	5.5	7.1	14.1	22.6	217	1.6	28.8	31.2	*	220.6
13	P. sativum 75121	4.1	13.3	18.9	*	157.5	0.2	33.8	35.5	*	*
14	P. sativum Arthur *	12.5	14.9	28.9	20	225.0	17.4	36.3	55.3	18.8	253.8
15	Revermont Petit pois *	13.9	19.7	35.1	18.7	198.4	22.9	32.4	58.1	20.7	215.4
16	Century *	12.2	1.4	15.0	17.5	205.6	16.5	22.9	40.6	21.3	227.4
17	Lamm 31; JI 12; L31 *	3.7	17.1	22.3	20.5	193.0	1.4	30.1	32.9	*	*
18	Merveille d´Étampes *	13.1	13.8	28.4	21.1	160.8	7.9	27.3	36.6	22.9	195.4
19	Dwarf Sugar (1969) *	1.8	15.0	18.3	*	123.2	1.1	25.7	28.6	*	*
20	Nike (1969) *	3.1	13.4	18.0	19.8	158	3.8	19.9	24.5	22	180.5

Figure 1. Yield results in 2020, for pea and barley

Statistical methods

Because our trial was a screening-trial, we used no special experimental design and conducted no statistical analyses.

Discussion

2018-19

Six of the 20 genotypes tested where identified as interesting for special uses *(“Nischensorte”)* and/or crossings with GZPK breeding lines. As the climatic conditions in Oberstammheim where extremely dry, some accession (Arthur, Revermont Petit Pois, Merveille d'Étampes, Century) where identified with a possible drought tolerance. Other interesting traits were high protein content (Merveille d'Étampes 24.4 %), late flowering and maturity (Revermont Petit Pois, Merveille d'Étampes, Century). As predicted, some of the accessions from the gene banks where not suitable for modern agriculture practice, especially caused by their high tendency to lodging and their poor agronomic performance. The available varieties of spring triticale may not be the right partners for mixtures with spring peas, because of their different maturity dates. By breeding for later maturing peas and/or earlier maturing triticale the potential of these partners grown together can be enhanced. Future goals for improving the crop mixture of the two crops are peas with late maturity or triticale with early maturity. By this, a better threshing result could be provided.

2020

As shown in the two previous test years not all accessions are suitable for modern agriculture. However, interesting traits were detected and first crossings with gene bank accessions and GZPK breeding lines were conducted. Accessions, which were identified to have interesting traits in previous years, mostly showed the same pattern again. “Merveille d´Étampes” had the highest protein content in 2018, this year was the fifth best; “Nike (1969)” and “Feltham First

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Rogue" had highest protein in 2019 and were forth best and in the middle this year, respectively. However, three new accessions exceeded the values – "Procumbens-pro (1968), Densinodosum-

lkb (2005) and Recette (C-4) (1969) – and overall protein values were comparatively low. In 2019 "Feltham First Rogue" was the earliest accession to flower, in 2020 two new accessions were even earlier, with "P. sativum 75121" developing extremely early. The risk of lodging was generally low this year and no huge differences occurred. The mixture of three summer barley varieties functioned well as a stabilizer and competitor against weed. However, as some peas did not grow well plots needed hand weeding during the season but yields still varied greatly; no sticks for stabilizing were needed like in previous years.

Outcomes and conclusions

Barley proved to be the best mixing partner for summer peas, providing stability against lodging, suppressing weeds and was ripe at the same time. Many gene bank accessions have interesting traits; however, are not always suitable for modern agriculture.

Early flowering might help to avoid drought stress in summer. Thus, the project helped to ensure an enlargement of the gene pool available for breeding, revealing accessions with traits valuable for future agriculture.

Main lessons learned

Mixed cropping needs infrastructure to separate harvest (if not used as fodder). Finding the right mixing partners is not always easy and might need a lot of weeding by hand.

How the information can be transfer to the other situations?

Information on suitable sowing density for mixtures and on sowing/harvesting can be shared and advice on how to separate harvested seeds. Networking started on marked usability besides fodder and will be useful to improve infrastructure. Gene pool used for crossings has been enlarged, thus, diversified organically bred seeds will be available in near future.

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Cultivari - Winter pea mixture trial with winter triticale (Trial #11)

LEAD: Karl-Josef Mueller, David Gloger (CULTIVARI)

Abstract

To study effects on yield of different combinations between ten normal and semleafless types of winter peas of different plant length with three winter triticale of different length a trial with three replications for each combination (90 plots) was planted in autumn 2018 after selection of ten peas out of fourty in vegetation period 2017/18.

Five combinations of isogenic winter peas (normal leaf type: semi-leafless) with different length (10 samples) were combined with three different triticale varieties. Because of loss of peas during season and their low contribution to harvest, the expressiveness of the trial is low. But statistical analyses on yield showed differences between triticale varieties depending on pea varieties and pea varieties independent from triticale varieties, but no statistical interactions. Short peas were suppressed most, but a medium type in length could reach nearly up to the very tall pea varieties, which suppressed triticale most. For the medium length type of pea, the triticale seemed to be a strong competitor.

The trial was repeated in season 2019/20. Six combinations of isogenic winter peas (normal leaf type/semileafless) with different length (12 samples) were combined with three different triticale varieties. Even despite very limited precipitation during the growing season (65mm/m² from April till July 2020) satisfying yields of about 3t/ha of mixture yield could be obtained. Statistical analyses on yield showed differences between triticale varieties depending on pea varieties and pea varieties independent from triticale varieties, but no statistical interactions. On low level of significance, normal leaved peas supressed the triticale more but led to slightly higher pea share in the yields. Ground covering was higher with normal leafed peas.

Introduction

Under organic farming in the middle-north of Germany growing of winter peas can be done best in mixture with triticale, because it has a better performance and is later ripening than barley, is not as much shadowing like rye and can compensate yield, if peas are reduced by weather conditions. Peas can differ first of all in height and leaf type related to the question, which type would be best in yield to combine with triticale, but also whether there is an influence of triticale to yield.

Hypothesis and research questions

Winter triticale can compensate loss of peas during winter best. It is also a pillar for peas, but a competitor too.

- Whether the type of leaf and the length of the pea plant has an influence on yield of pea and triticale?
- Whether a shorter or taller triticale can compensate missing peas better?
- Which type of winter pea related to plant length and leaf type should be followed in breeding for mixing with winter triticale to suit best?

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The trial shall give hints for which type of winter pea should be used in mixtures and whether height and leaf type have an influence on yield or can show interaction with different triticale.

Material and Methods

Location(s)

2017-18

The 400 microplots with winter peas were sown in Sept. 2017 on a loamy sand near D-21371 Tosterglope, OT Köhlingen, North-East of Lower Saxony in Northern Germany after spring barley (53°12'22.0"N 10°49'43.0"E). The trial with 90 yield plots in Sept. 2018 under similar conditions on a field of the same organic farmer (Andreas Wenk) in a distance of about 1km to previous year (53°12'48.9"N 10°50'29.3"E).

2019-20

53°12'49.3"N, 10°50'29.2"E, DE-21371 Tosterglope-Koehlingen-Bus Stop, loamy sand

Genetic resources

2017-18

Ten accessions which were selected out of 40 samples containing breeding lines and varieties during vegetation 2017/2018 for yield trial starting in autumn 2018:
Molly – dark green, short, normal leaf, coloured
Polly – dark green, short, semileafless, coloured
DZP1009d4 – medium-short, normal leaf, white
DZP1009e - medium-short, semileafless, white
DZP0801f41 – medium-tall, normal leaf, white
DZP0801f43 – medium-tall, semileafless, white
DZP0801f64 – tall, normal leaf, white
DZP0801f63 – tall, semileafless, white
Karolina – very tall, normal leaf, white
Szarvasi Andrea – very tall semileafless, white

These were sown for season 2018/19 in all combination with winter triticale
Agostino - short
Vuka – medium length
Securo - tall

In 2018/19
Winterpeas (seed density 70 seeds/m²) all with white flowers
DZP 11 03 b V short (3), normal leaf
DZP 11 01 e R short (3), semileafless
DZP 10 09 d4 V short to medium (4), normal leaf
DZP 10 09 e1 R short to medium (4), semileafless
DZP 08 01 f6 V medium to tall (6), normal leaf
DZP 08 01 f6 R medium to tall (6), semileafless
DZP 08 01 f4 V tall (7), normal leaf
DZP 08 01 f4 R tall (7), semileafless
Karolina tall to very tall (8), normal leaf
Szarvasi Andrea tall to very tall (8), semileafless

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<u>All of them in combination with</u>:
Wintertriticale (seed density 120 seeds/m²)
Agostino medium after winter, late ear emergence, short
Vuka planophile after winter, medium ear emergence, broad leaves, tall
Securo erectophile after winter, medium ear emergence, very tall,

<u>Changes for 2019/2020</u>
Winter peas (seed density 75 seeds/m²)
<u>Implemented new</u>
DZP 08 03 c7 V medium (5), normal leaf
DZP 08 03 c7 R medium (5), semileafless
DZP 08 01 e V very tall (9), normal leaf
DZP 08 01 e R very tall (9), semileafless
Pandora V tall (7), normal leaf
Kolinda R tall (7), semileafless
<u>Excluded</u>
DZP 11 03 b V short (3), normal leaf
DZP 11 01 e R short (3), semileafless
DZP 08 01 f6 V medium to tall (6), normal leaf
DZP 08 01 f6 R medium to tall (6), semileafless

Experimental design

2017-18

Forty different winter peas were sown on Sept, 2017 each with ten descendants on ten microplots (400 microplots). Out of these 10 were selected for yield trials in combination with three different winter triticale and sown on Sept. 17th 2018 with 3 replications for each combination in a randomized bloc trial on plots of 3m² each and with a sowing density of 70 seeds/m² for winter pea and 120 seeds/m² for winter triticale.

For the selection of 10 peas out of 40 the following characters were considered: winter hardiness, plant height, leaf type, ripening related to ripening of winter triticale, amount of grain at harvest.

Important for the selection was the availability of the combination of similar types as normal leaf type and semileafless type. For this combination, a short, a medium-short, a medium-tall and a tall couple out of the breeding material were chosen and the combination of two tall varieties from the same breeding station with normal and semileafless type. For fasciata-types there was not enough seed available because of weak winter hardiness. Also, for winter triticale a short, a medium-tall and a tall variety with improved winter hardiness were chosen for the combination with the different winter peas.

2018-19

Ten winter peas, representing 5 different types in height, each as one with normal leaf type and one with semi leaf type and where possible as isogenic lines, were combined with three different triticale varieties on plots of 3,125m² with three replications (90 plots in total as randomised block design). Date of sowing was Sept. 17th, 2018. A mechanical hoeing was done on Nov. 6th, 2018. Harvested with plot combine on July 19th, 2019. Dried with cold air and separated with sieves and spiral separator. Statistical variance analyses were done as cross classification for yield (pea – triticale – combined).

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Additional to the yield trial, all accessions were maintained each in five single plant descendant plots with single plant harvests and rest plot threshing.

2019-20

Twelve winter peas, representing 5 different types in height, each as one with normal leaf type and one with semi leaf type and where possible as isogenic lines, were combined with three different triticale varieties on plots of 3,125m² with three replications (108 plots in total as complete randomised block design). Date of sowing was Sept. 18th, 2019. A mechanical hoeing was done on Oct. 31st, 2019. Harvested with plot combine on July 18th, 2020. Dried with cold air and separated with spiral separator. Statistical analyses were done as a multi-factorial ANOVA. Three principal factors were tested for their effects: Triticale Variety, Pea-Type (Group of similar types as one) and Pea Leaf-type.

Evaluation

2017

List of the traits collected in the trial Table 1-3.

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
2017-2018		
Trait	**How it has been assessed**	**Type of data available**
Winter hardiness	Scoring in relation to each other	1=best
Plant height of pea at beginning of shooting of triticale	Measured	cm transformed into scores
Beginning of flowering of pea	Observation	Day in May
Plant length at end of pea flowering	Measured	cm transformed into scores
2019		
Height in April	Measure in cm transferred into score	cm and score
Height in June	Measure in cm transferred into score	cm and score
Date of ear emergence	Date	date and score
2020		
Height in April	Measure in cm	cm
Height in June	Measure in cm	cm
Ground cover of Pea	Visual notation	%
Ground cover of Triticale	Visual notation	%

Table 1. List of the traits collected for pea and triticale concerning crop development and agro-ecological performance

Crop productive performance (yield, yield components)		
2017-18		
Trait	**How it has been assessed**	**Type of data available**

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Total yield of combination	Addition of yield of pea and triticale	dt/ha
Yield of pea variety Yield of triticale cv. Thousand Grain Weight	Weighing Counting and weighing	dt/ha g
2019		
Yield of pea samples	Weight after threshing and drying	dt/ha
Yield of triticale	Weight after threshing and drying	dt/ha
Yield combined	Weight after threshing and drying	dt/ha
2020		
Yield of pea samples	Weight after threshing, drying and separation	dt/ha
Yield of triticale	Weight after threshing, drying and separation	dt/ha
Yield combined	Weight after threshing and drying	dt/ha

Table 2. List of the traits collected for pea and triticale concerning crop productive performance

Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		
Trait	**How it has been assessed**	**Type of data available**
Crude protein	Near infrared Spectroscopy (NIRS)	Estimated %

Table 3. List of the traits collected for pea and triticale concerning crop quality performance.

Statistical methods

Statistics were done with PLABSTAT, Statistical model used for 2020: R + T + P + L + TP + LP + TL + TPL + RTPL the factor R was taken as random. Model for the 2-year analysis: Y + R:Y + T + P + TP + TY + PY + PTY + L + LT + PL + TLP + LY + PLY + TLY + TPLY + RTPLY factors R and Y were taken as random. Y=Year; R= Repetitions; T=Triticale Variety; P=Group of Pea; L= Leaf type, last term acts as residual/error term.

In addition to the yield trial, all accessions were maintained each in five single plant descendant plots with single plant harvests and rest plot threshing.

Participatory/multi-actor approach

On a pea field day on May 25th, 2019, the trial was presented to farmers and consultants, where the discussions faced to the trail and with respect to farmers experiences during the last three extremely different years related to very wet and very dry conditions ended in the following preferred type of winter pea: A winter pea for combination with triticale from farmers view should be white flowering with white kernels with high protein for threshing, relatively tall (score 6-9) and give also a good plant/leaf mass production beneath grain yield. The reason for this is the possibility to use the green plants alternatively for feeding in very dry years, where not enough feed is available at all. This outcome will be the reason to reduce the number of short types of peas for the season 2019/20 and extend the number of medium to tall types.

Due to the Corona-Virus pandemic, a possibility for contactless field visits was created during May and June. A brochure guiding through the winter-pea trials was created. Only few discussions with single

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visitors were possible. One discussion point was the possibility of having different maturity types of winter pea to be flexible in the choice of mixing partners. Barley and triticale are the main mixing partners used in practice, differing in their possibilities to be used as animal feed. As an outcome of this discussion winter pea types with earlier maturity, more suitable for the mixing with winter barley were identified in the breeding nursery and will in future be selected with focus on mixing-ability with winter barley additionally. The main breeding focus will stay on the mixing with triticale. But also, triticale should be looked for better total yield in combination with winter pea.

Results and discussion

Unfortunately, peas in 2018-19 disappeared more or less during winter and spring. Where the plots looked best with good growing of peas, there was also no satisfying pea yield, which reached from 0.9 to 4.8 dt/ha on average, where up to 25 dt/ha was expected. The extremely dry spring in combination with stem diseases seemed to be the reasons. Nevertheless, variance analyses could help to show, that there was a very high significant difference in pea yield related to the pea varieties (<0,01), but not related to the combining triticale. For yield of triticale from 21.7 to 25.5 dt/ha on average there was a very high significant difference (<0,01) related to triticale varieties and a low significance related to the combined peas. For the total yield of the combinations there was only a very high significant difference for the different triticale varieties. No significance could be found for any kind of interaction between pea and triticale related to separated or total yields.

With triticale VUKA, which has very broad leaves, highest yield of combinations and of triticale on average could be reached. Vuka could compensate best, in particular with a medium type in length and a semileafless type of pea (DZP1009e1R). Highest pea yield could be found with the very tall normal leaf type KAROLINA.

In the 2019-02020 trials, even despite very limited precipitation during the growing season (65mm/m² from April to July 2020), satisfying yields of about 35 dt/ha of mixture yield and pea yields from 4,5 to 26 dt/ha on average could be obtained. Statistical analyses on yield showed differences between triticale varieties depending on pea varieties and pea varieties independent from triticale varieties, but no statistical interactions.

Triticale Variety VUKA showed highest yield in mixtures and on its own as triticale in 2018/19 as well as in 2019/20. VUKA in 2018/19 compensated best, with the medium type in length and a semi leafless type of pea (DZP1009e1R). Best pea yields of about 19 dt/ha in 2020 were obtained with the longer and normal leafed varieties DZP0801f4V, KAROLINA and PANDORA in mixture with Triticale VUKA.

The highest yielding triticale VUKA as broad-leafed single ear type with erect overwintering gave the highest yields for pea, for mixture and, as seen in 2018/19, when necessary, a good compensation of pea losses, whilst SECURO as an ear density type with small leafless and flat overwintering, late ear emergence, produced lowest mixing yields as well as the weakest compensation in 2018/19.

As a tendency, normal leaved peas supressed the triticale yield more but led to slightly higher pea share in the yield. Results differed only on low level of significance (<0,1). Longer Varieties suppressed the triticale yield most, but also led to a higher share of pea in the mixture.

Ground covering in April was significantly higher with normal leafed peas (41,5%) compared to semi-leafless types (36,2%).

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In 2018/19 with an extremely dry spring in combination with stem diseases the pea yield was reduced to a low level. Nevertheless, a small dataset with three groups of peas (six varieties) could be analysed over the two trial years, the better ground cover of normal leafed pea and their stronger suppression of the triticale in yield were confirmed. The influence on total yield was mostly referring to the triticale yield and therefor on the yield performance of the triticale-variety itself.

Discussion

For interpretation of results from 2018-2019 it must be taken into account the very low yield of the peas in this season and the more or less normal development of the triticale. Total yield under these circumstances was influenced mainly by the triticale varieties, but not the pea varieties, which means that peas and triticale compensate each other, whatever the type of pea is, but the level of triticale yield brought the highest influence on total yield at all. The yield of the pea varieties was different, but not significantly influenced by triticale varieties. The shortest peas gave the lowest yield and the tallest with normal leaf type the highest. But it could not be shown that the leaf type has an influence on yield over all five types of plant length. The yield of triticale itself was also influenced by the type of pea. In particular, the very tall pea varieties Karolina and Szarvasi Andrea suppressed the triticale yield significant. But for total grain yield it could not be distinguished whether also a medium tall semileafless type will be best for balanced high yield at all.

In season 2019/20 we saw a parallel development of pea and triticale. Yield share in average was balanced with 50% for each mixing partner. Good circumstances to detect possible interactions between pea and triticale in mixed cropping. In case of the trait yield no significant interactions between the pea and the triticale varieties were found. Together with the results from the two-year analysis the picture is getting even more clear: No statistically significant interaction between single genotypes. It means that it doesn’t matter which type of triticale is used to select for best yield of pea varieties in mixture, but it is important to find the best triticale for best total yield at all.

Especially for organic farming systems, crops and varieties with a good weed suppression are crucial. The experiment could show the positive effect of a normal leafed pea to better supress weeds from the beginning of the growing season on and there seems to be a tendency, that normal leaf type of pea can reach better yield. But normal leaf type is also more sensitive for lodging, which might limit the use related to the fertility of the soil, the location, and the weather conditions. Therefore, more variety trials with mixtures at different locations would be preferable.

Total yield in 2020 and in the two-year analysis was influenced mainly by triticale varieties, but not pea varieties, which means that peas and triticale compensate each other, whatever the type of pea is. But the level of triticale yield brought the highest influence on total yield at all. The yield of the pea varieties was different, but not significantly influenced by triticale varieties. The shortest peas gave the lowest yield and the tallest with normal leaf type the highest. The yield of triticale itself was influenced by the type of pea. The leaf type has an influence on yield over all five length-types of peas. The very tall pea varieties KAROLINA and SZARVASI ANDREA suppressed the triticale yield significantly.

Outcomes and conclusions

For more or less extensive locations like the ones available for the trials as well yielding triticale variety should be combined with more or less tall pea varieties. Normal leafed types due to their enhanced weed suppression and their slight yield advantage should be preferred under organic farming.

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To breed for better mixing ability, a medium to tall normal-leafed winter pea will be the first choice and a high yielding triticale with good compensation should be the one to combine with in yield trials for pea selection.

As triticale has such an influence on the total yield, it should be given more research on more different varieties of triticale to be combined with medium to tall peas to increase the mixed yield with a focus on the pea share and the compensation ability of the triticale. In trials for searching better triticale in mixtures a special view should be given on morphological characters like broadness of leaves, date of ear emergence and rapidness of juvenile growth, plant length and ears per square meter to develop a 'triticale mixture type'.

How information was transferred to the other situations

The results showed that length of pea in relation to length of triticale is relevant for relation of yield among both and that yield of triticale in the mixture is important for total yield.

How resilience was improved

Resilience of pea-triticale-mixtures will become better as more types of winter-pea-varieties suitable for the mixture with winter triticale will become available and more emphasis has to be laid on choosing the most suitable winter triticale varieties for the mixtures. Last lead to a new project to test different winter-triticale-varieties and breeders-lines to develop a new breeding aim "suitability for mixtures" by describing the most suitable morphological type for pea-triticale-mixtures.

Publications

The webpage of www.cultivari.de provides articles on the first years of the trials, but due to poor yield and only one-year of results from one location were not enough to produce a scientific report. It was a pity, that the start of the project for winter pea with the necessity to get first enough seeds of the different isogenic pea types allowed only two trial-years and only one location in the project time.

Supplementary material

2019.

Variance analyse		Discard nullhypothesis	
	yield pea	yield triticale	total yield
between peas	Yes ***	Yes *	No
between triticale	No	Yes ***	Yes ***
Intereaction peaXtriticale	No	No	No
rest	No	No	No
level of significance	*=<0,05	**=<0,01	***=<0,001

Figure 1. Results of variance analysis in 2019

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YIELD in dt/ha	yield of pea (STDDEV: 0,24 dt/ha)			
Name	Agostino	Vuka	Securo	Average
DZP1103bV	1,3	0,6	0,9	0,9
DZP1101eR	1,0	0,2	1,4	0,9
DZP1009d4V	1,7	1,0	1,8	1,5
DZP1009e1R	3,1	2,7	3,4	3,0
DZP0801f6V	0,7	2,4	1,0	1,4
DZP0801f6R	1,0	0,0	1,3	0,8
DZP0801f4V	1,8	1,6	3,0	2,1
DZP0801f4R	1,9	1,6	3,3	2,2
KarolinaV	4,8	6,1	3,5	4,8
SzarvasiAndreaR	3,5	1,8	2,8	2,7
Average	2,1	1,8	2,2	
YIELD in dt/ha	yield of triticale (STDDEV: 0,69 dt/ha)			
Name	Agostino	Vuka	Securo	Average
DZP1103bV	25,2	28,2	21,9	25,1
DZP1101eR	24,4	27,8	22,6	24,9
DZP1009d4V	26,5	28,1	21,8	25,5
DZP1009e1R	23,9	28,0	20,5	24,1
DZP0801f6V	26,6	26,3	21,0	24,7
DZP0801f6R	24,7	25,7	21,0	23,8
DZP0801f4V	24,2	26,5	20,7	23,8
DZP0801f4R	24,3	26,3	20,1	23,5
KarolinaV	22,7	23,2	19,4	21,7
SzarvasiAndreaR	23,4	23,9	18,6	22,0
Average	24,6	26,4	20,7	
YIELD in dt/ha	combined yield (STDDEV:0,63 dt/ha)			
Name	Agostino	Vuka	Securo	Average
DZP1103bV	26,3	28,6	22,6	25,9
DZP1101eR	25,2	27,8	23,7	25,6
DZP1009d4V	28,0	28,9	23,4	26,8
DZP1009e1R	26,7	30,5	23,7	27,0
DZP0801f6V	27,1	28,5	21,8	25,8
DZP0801f6R	25,6	25,5	22,2	24,4
DZP0801f4V	25,7	27,9	23,5	25,7
DZP0801f4R	25,9	27,7	23,2	25,6
KarolinaV	27,3	29,1	22,7	26,4
SzarvasiAndreaR	26,7	25,5	21,2	24,5
Average	26,5	28,0	22,8	

Figure 2. Results on yield 2019

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2020

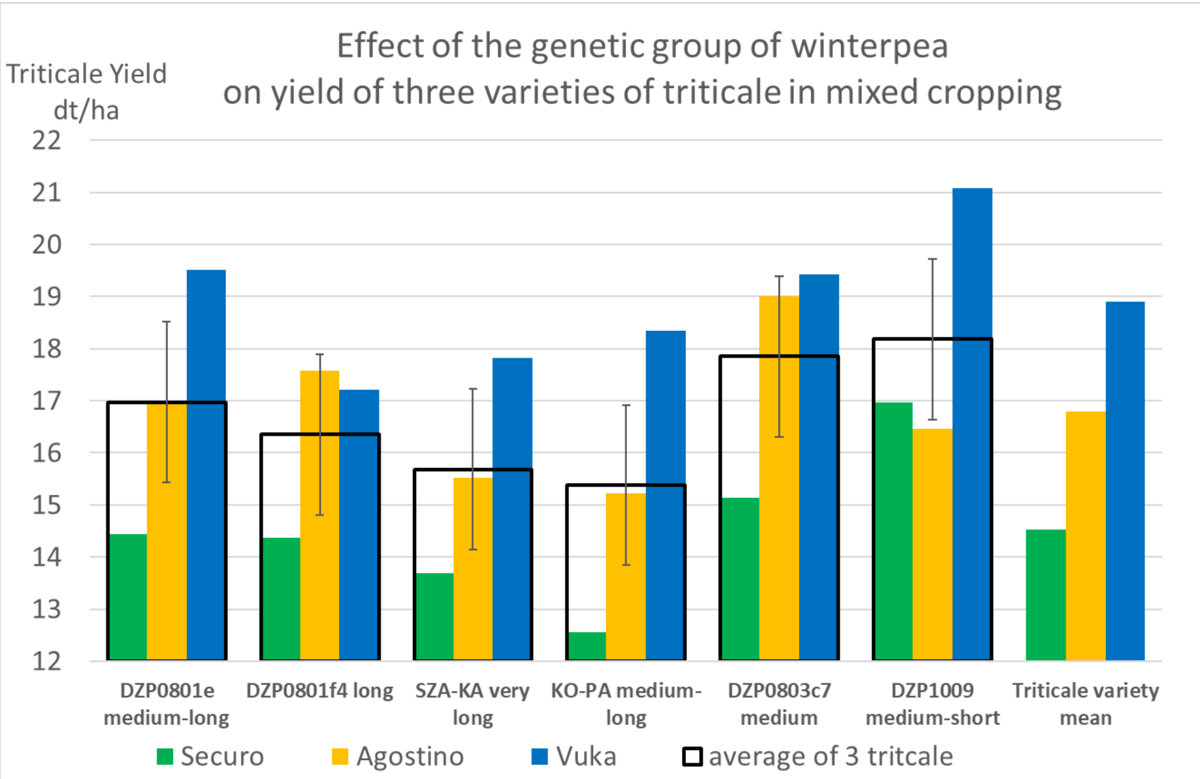

Figure 3. Effect of the genetic group of winter pea on triticale yield of three varieties in mixed cropping. Error bars indicate LSD 5%, different letters above variety means indicate significant differences according to LSD5%.

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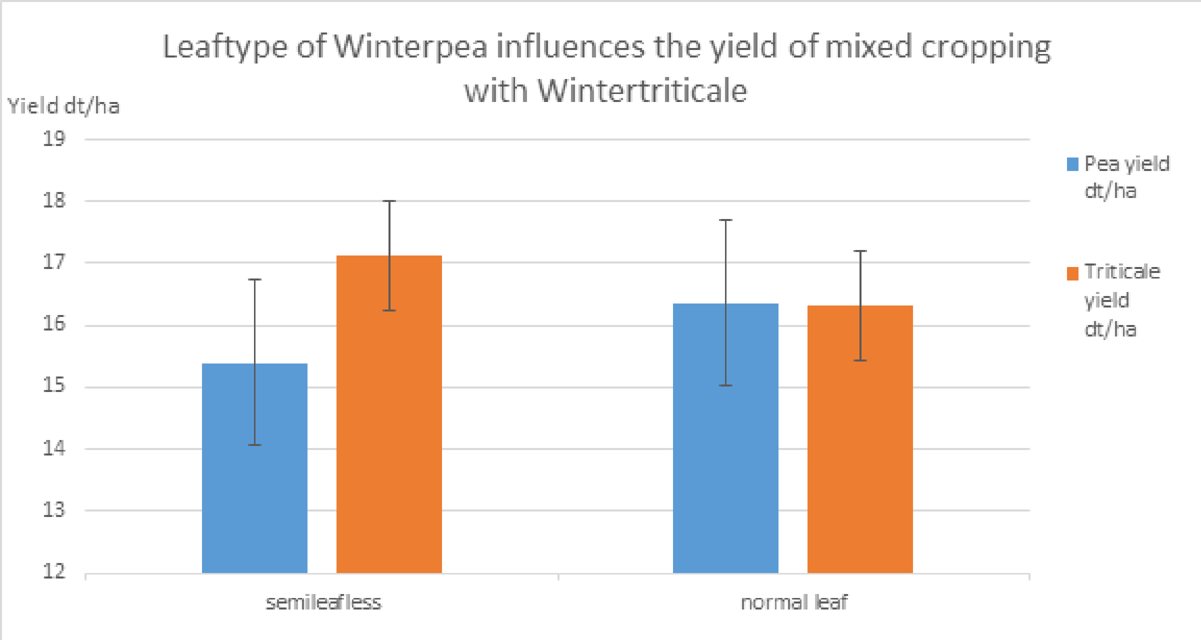

Figure 4. Leaf type of winter pea influences the yield of mixed cropping with winter triticale. Means of Leaf-type of winter pea (n=12 var., 6 normal-leafed and 6 semileafless) tested. Error bars indicate LSD 5%.

Results 2020: Pea Varieties	Pea plants/ 2m row	Early plant height pea (cm) 08.04.2020	Early plant Height Triticale (cm) 08.04.2020	Ground covering pea (%) 08.04.2020	Ground covering triticale (%) 08.04.2020	Ground covering total (%) 08.04.2020	Final plant lenght (cm) Triticale before Harvest 16.06.2020	Final Plant lenght (cm) pea before Harvest 16.06.2020	Lodging Index at harvest 16.06.2020	Pea yield dt/ha	Triticale yield dt/ha	Total yield dt/ha	Harvest Index Pea/Triticale	TKM Pea (Mixed probe all REP.)
Min	11,0	5,0	15,0	10,0	30,0	55,0	55,0	40,0	0,78	4,5	8,5	21,1	0,2	117,6
Max	33	35	30	60	70	90	105	125	1	26	27	45	2,2	215,5
Mean	21.30	15.88	21,11	38,84	45,05	83,89	97,87	90,02	0,96	15,75	16,74	32,77	0,99	153,58
LSD 0.05	n.s.	1,18**	n.s	6,85**	5,76**	n.s.	n.s.	n.s.	n.s.	3,25*	n.s.	3,91+	n.s.	Means only
DZP1009e11R	21,2	10,0	22,2	22,8	55,6	78,3	98	55,6	1	10,34	18,61	29,14	0,57	193,8
DZP1009d4V	20,2	10,6	20,6	22,2	53,3	75,6	98	55,0	1	13,04	17,74	32,26	0,73	207,8
DZP0801f4R	20,2	16,1	21,1	40,6	42,8	83,3	98	101,7	0,97	17,62	17,17	34,98	1,05	130,1
DZP0801f4V	20,4	16,7	21,1	43,9	43,9	87,8	98	105,2	0,96	19,19	15,6	34,93	1,24	131,7
DZP0803c7R	19,4	11,1	20,0	26,7	53,3	80,0	98	69,4	0,99	12,28	17,94	30,44	0,71	137,9
DZP0803c7V	20,3	11,7	20,0	31,1	50,0	81,1	98	62,2	0,99	13,69	17,76	31,58	0,8	137,3
Szarvasi Andrea	23,1	26,1	22,8	56,7	32,2	88,9	98	118,9	0,92	19,06	16,04	35,27	1,26	171,6
Karolina	19,0	20,6	21,1	50,6	38,3	88,9	98	113,3	0,89	15,62	15,32	31,08	1,04	200,9
DZP0801eR	21,3	17,8	20,0	41,7	44,4	86,1	98	98,9	0,96	16,23	17,21	33,58	0,97	121,3

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DZP0801eV	23,7	18,3	20,6	46,7	38,9	85,6	98	105,6	0,95	16,88	16,72	33,77	1,08	125,6
KolindaR	23,3	13,3	21,7	28,9	52,2	81,1	98	99,4	0,97	15,33	15,76	31,32	1,06	148,2
PandoraV	23,2	18,3	22,2	54,4	35,6	90,0	93	95,0	0,94	19,73	15	34,99	1,32	136,7

Figure 5. Results 2020: Pea Varieties

Results 2020: Group of Pea, Leafty pe, Triticale-Variety	Pea plants/ 2m row	Early plant height pea (cm) 08.04.2020	Early plant Height Triticale (cm) 08.04.2020	Ground covering pea (%) 08.04.2020	Ground covering triticale (%) 08.04.2020	Ground covering total (%) 08.04.2020	Final plant lenght (cm) Triticale before Harvest 16.06.2020	Final (Plant lenght (cm) pea before Harvest 16.06.2020	Lodging Index at harvest 16.06.2020	Pea yield dt/ha	Triticale yield dt/ha	Total yield dt/ha	Harvest Index Pea/Triticale	TKM Pea (Mixed probe all REP.)
LSD 0.05	n.s.	2,36**	n.s.	4,84**	4,07**	4,44**	n.s.	7,24**	0,03**	2,3**	1,54**	2,77*	0,19**	Means only
DZP1009	20,7	10,3	21,4	22,5	54,4	76,9	98	55,3	1	11,69	18,18	30,7	0,65	200,8
DZP0801f4	20,3	16,4	21,1	42,2	43,3	85,6	98	103,4	0,96	18,41	16,35	34,96	1,14	130,9
DZP0803c7	19,9	11,4	20,0	28,9	51,7	80,6	98	65,8	0,99	12,98	17,85	31,01	0,75	137,6
SZA-KA	21,1	23,3	21,9	53,6	35,3	88,9	98	116,1	0,91	17,34	15,68	33,17	1,15	186,2
DZP0801e	22,5	18,1	20,3	44,2	41,7	85,8	98	102,2	0,95	16,56	16,97	33,67	1,03	123,5
KO-PA	23,3	15,8	21,9	41,7	43,9	85,6	96	97,2	0,96	17,53	15,38	33,11	1,19	142,4
LSD 0.05	n.s.	n.s	n.s.	2,80**	2,35**	n.s.	n.s.	n,s,	0,02+	1,33+	0,89+	n.s.	0,11+	Means only
semileafless	21,2	15,7	21,3	36,2	46,8	83,0	98	90,7	0,97	15,4	17,12	32,44	0,94	150,5
normal leaf	21,4	16,0	20,9	41,5	43,3	84,8	97	89,4	0,96	16,36	16,32	33,1	1,04	156,7
LSD 0.05	n.s.	1,67*	1,23**	3,42**	2,88**	3,14**	2,26**	n.s.	0,02**	n.s.	1,09**	1,96**	0,13*	Means only
Agostino	20,3	15,8	21,1	35,7	47,1	82,8	90	89,3	0,94	15,67	16,79	33,0	0,96	152,4
Vuka	22,3	16,9	25,4	37,8	49,6	87,4	100	91,4	0,97	16,46	18,9	35,5	0,9	156,1
Securo	21,3	14,9	16,8	43,1	38,5	81,5	104	89,3	0,97	15,13	14,52	29,8	1,1	152,3

Figure 6. Results 2020: Group of Pea, Leaf type, Triticale-Variety

Results 2020: Further significant Effects.	Pea plants/2m row	Early plant height pea (cm) 08.04.2020	Early plant Height Triticale (cm) 08.04.2020	Ground covering pea (%) 08.04.2020	Ground covering triticale (%) 08.04.20 20	Ground covering total (%) 08.04.2020	Final plant lenght (cm) Triticale before Harvest 16.06.2020	Final Plant lenght (cm) pea before Harvest 16.06.2020	Lodging Index at harvest 16.06.2020	Pea yield dt/ha	Triticale yield dt/ha	Total yield dt/ha	Harvest Index Pea/Triticale	TKM Pea (Mixed probe all REP.)
Further significant effects	none	none	Repetitions**	Repetitions*	Repetitions*; PEA-Group x Triticale*	Repetitions **	none	none	none	none	none	none	Repetitions*	

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MODEL R + T + P + L + TP
+ LP + TL + TPL + RTPL
R= Repetitions; T=Triticale Variety;
P=Group of Pea; L= Leaftype, last term
acts as residual/error term.

LSD 0,05 and significance
according to f-statistics.
+=significant at level
of confidence: 0,1
*=significant at level
of confidence: 0,05
**=significant at
level of confidence:
0,01

Figure 7. Results 2020: Further significant Effects.

yield in dt/ha	yield of pea (STDDEV: 1,07 dt/ha)				yield of triticale (STDDEV: 0,54 dt/ha)				combined yield (STDDEV:1,30 dt/ha)			
Name	Agostino	Vuka	Securo	Average	Agostino	Vuka	Securo	Average	Agostino	Vuka	Securo	Average
DZP1009d4V	12,3	14,7	12,2	13,0	16,3	20,4	16,6	17,7	32,6	35,2	28,9	32,2
DZP1009e11R	10,8	10,5	9,8	10,3	16,7	21,8	17,4	18,6	27,8	32,5	27,2	29,1
DZP0801f4V	20,0	20,6	17,0	19,2	15,8	17,0	14,0	15,6	36,0	37,7	31,1	34,9
DZP0801f4R	17,0	19,2	16,6	17,6	19,4	17,4	14,8	17,2	36,6	36,8	31,5	35,0
DZP0803c7V	12,8	14,3	13,9	13,7	19,1	19,0	15,1	17,7	32,2	33,5	29,0	31,6
DZP0803c7R	11,3	12,6	13,0	12,3	18,9	19,8	15,2	18,0	30,5	32,5	28,3	30,4
Karolina	14,6	16,5	15,8	15,6	15,1	16,1	14,7	15,3	29,9	32,8	30,6	31,1
Szarvasi Andrea	18,2	21,5	17,5	19,0	15,9	19,5	12,6	16,0	34,4	41,2	30,2	35,3
DZP0801eV	20,4	12,8	17,4	16,9	16,6	19,6	14,0	16,7	37,2	32,6	31,5	33,8
DZP0801eR	14,7	19,4	14,6	16,2	17,4	19,4	14,9	17,2	32,3	38,8	29,6	33,6
PandoraV	20,4	21,2	17,6	19,7	14,7	17,0	13,3	15,0	35,6	38,3	31,0	35,0
KolindaR	15,6	14,2	16,2	15,3	15,8	19,7	11,8	15,8	31,6	34,1	28,1	31,2
Average	15,7	16,5	15,1		16,8	18,9	14,5		33,0	35,5	29,8	

Figure 8. Means of all pea-triticale combinations for yield 2020

Results: 2-years	Ground covering pea (%) 08.04.2020	Early plant height pea (cm) 08.04.2020	pea yield dt/ha	triticale yield dt/ha	total yield dt/ha	Harvest Index Pea/Triticale	TKM
Min	8	5	0,12	8,5	16,6	0,01	86
Max	60	38	24,1	37,5	44	2,02	215,5
Mean	31,37	20,31	8,98	20,13	29,31	0,54	142,71
LSD 0.05	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	means only
DZP1009e11R	18,67	12,11	6,31	21,69	28,1	0,34	149,25
DZP1009d4V	19,72	12,39	6,98	21,89	29,61	0,39	153,07
DZP0801f4R	32,22	20,72	9,59	20,27	29,97	0,56	115,55

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DZP0801f4V	30,94	19,61	10,41	19,15	29,62	0,66	113,67
Karolina	45	30,33	10,73	18,99	29,81	0,69	162,97
Szarvasi Andrea	41,67	26,72	9,88	18,82	28,76	0,62	161,77
LSD 0.05	n.s.	9,83*	n.s.	1,23*	n.s.	n.s.	means only
DZP1009	19,19	12,25	6,65	21,79	28,85	0,36	151,16
DZP0801f4	31,58	20,17	9,68	19,71	29,79	0,61	114,61
SZA-KA	43,33	28,53	8,48	18,9	29,28	0,66	162,37
LSD 0.05	0,94+	n.s.	n.s.	n.s.	n.s.	n.s.	means only
semileafless	31,96	21,06	8,88	20,32	29,29	0,53	142,59
normal leaf	30,78	19,57	9,09	19,95	29,33	0,56	142,83
LSD 0.05	n.s.	n.s.	n.s.	2,68*	2,60*	n.s.	means only
Agostino	29,42	19,33	8,79	20,32	29,55	0,52	141,12
Vuka	28,33	21,56	9,68	22,14	31,91	0,53	147,92
Securo	36,36	20,06	8,48	17,94	26,47	0,58	139,1
Further significant effects	Year**; Triticale x years*; Pea-Group x Years **; Leaftype x Pea Group x Years+; Pea Genotype x Triticale+	Year**; Pea-Group x Years **; Pea Genotype x Year +	Years**; Repetitions: Years*; Pea-Group x Years **; Pea Genotype x Years**	Year+; Repetitions: Years**	Year*; Repetitions: Years**; Pea Group x Years **; Pea Genotype x Years*	Year**; Repetitions: Years*; Pea Group x Years **; Pea Genotype x Years**	

MODEL Y + R:Y + T + P + TP + TY + PY + PTY + L + LT + PL + TLP + LY + PLY + TLY + TPLY + RTPLY

RANDOM R Y

Y=Year; R= Repetitions; T=Triticale Variety; P=Group of Pea; L= Leaftype, last term acts as residual/error term.

LSD 0,05 and significance according to f-statistics.

+=significant at level of confidence: 0,1

*=significant at level of confidence: 0,05

**=significant at level of confidence: 0,01

Figure 9. Combined results of 2-years

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LBI - Pea - Cereal mixtures (Trial #12)

Lead: LBI - The Netherlands.
Floor van Malland - f.vanMalland@louisbolk.nl, Abco de Buck - a.debuck@louisbolk.nl

Abstract

The mixing ability of different pea varieties and breeding lines with different spring wheat varieties, barley and oats is determined in organic field trials in 2019 and 2020. Mixtures of species are compared with species in pure stand based on yield, development of diseases and plant development. The results of the trials are indicative, because of the small trial setup (only 2 repetitions, and small plots). The results of the trial in 2020 were unfortunately not representative due to the high weed pressure and the presence of diseases. The trial 2019 showed that mixed cropping with peas is certainly possible in the Netherlands with the right pea varieties and cereals. This should be further investigated within a larger trial setup. The results will allow a better understanding of traits important for successful mixed cropping of species.

Introduction

Objectives

The main aim of this trial is to identify traits important for mixed cropping of cereals and peas. What are the differences in yield, diseases, and plant development between de cultivars in mixed stand and pure stand? Are commercial varieties (peas and cereals) suitable enough for mixed cropping in the Netherlands and is there a difference between varieties how they adapt to mixed cropping?

Material and Methods

Location

The trial field 2019 was located within a commercial field with barley at an organic farm in the central polder area (Bant, The Netherlands) on a sandy soil with loam. The trial field 2020 was located within a commercial field with Triticale and pea at an organic farm in the central polder area (Rutten, The Netherlands) on a sandy soil with loam.

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Figure 1. Field pea cereals trial 2019 (photo taken by Floor van Malland 27-06-2019)

Genetic resources

<u>Cereal varieties, pea varieties and breeding lines and mixtures:</u>

Cereals:
- *Wheat Lavett (Agrifirm, The Netherlands)*
- *Wheat Lennox (Agrifirm, The Netherlands)*
- *Wheat HSWS 616-15 (Dottenfelderhof, Germany)*
- *Wheat Saludo (Dottenfelderhof, Germany)*
- *Barley Irina (Agrifirm, The Netherlands)*
- *Oats Elyann*
- *Oats Olympic (Agrifirm, The Netherlands)*

Pea:
- *Audit (Agrifirm, The Netherlands)*
- *Westrupper Grijze (Zaderij, The Netherlands)*
- Nordic Maize Breeding 1
- Nordic Maize Breeding 2
- Nordic Maize Breeding 1.1
- Nordic Maize Breeding 5

Creation of experimental population

Intercropping

Experimental design

Year	Sowing	Harvest
2019	11-04-2019	08-08-2019
2020	01-04-2020	07-08-2020

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The trial was sown in 2 repetitions (plot size gross 11,25 m2, net 10,1 m2).

Sowing density:
- wheat pure stand: 300 seeds/m2
- Pea pure stand: 85 seeds/m2 (2019) and 100 seeds/m2 (2020)

mixture of wheat and white lupins:
- wheat: 100 seeds/m2
- pea: 30 seeds/m2 and 50 seeds/m2 (2020)

Evaluation

List of the traits collected for lupin and wheat concerning Crop development and agro-ecological performance, Crop productive performance and Crop quality performance.

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Germination pea	Counting plants	%
Weed pressure	(9=no weeds to 1=only weeds present)	Scoring
Disease's pea	Brown spots and powdery Mildew (1=dead heavy infected to 9=not infected)	Scoring
Plant height pea and cereal	Measuring height	Cm
Ripening plants	Ripening, state of yellowing (Yellowing of total plant 1=green, 5=yellow)	Scoring

Table 1. Crop development and agro-ecological performance traits

Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**
Yield	Harvesting per plot, dried (mixtures with pea are separated) and cleaned	t/ha
Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		
Trait	**How it has been assessed**	**Type of data available**
Protein content pea	Laboratory Gent University	%

Table 2. Crop productive performance and Crop quality performance traits

Statistical methods

None, only indicative results, because of the small trial setup

Participatory/multi-actor approach

Organisation of workshops and annual field days to inform and discuss results with growers, researchers, breeders, advisors, and other stakeholders of the value chain.

Results and discussion

Main results

Figure 2 shows the average total yield (t/ha) of the different treatments in 2019.

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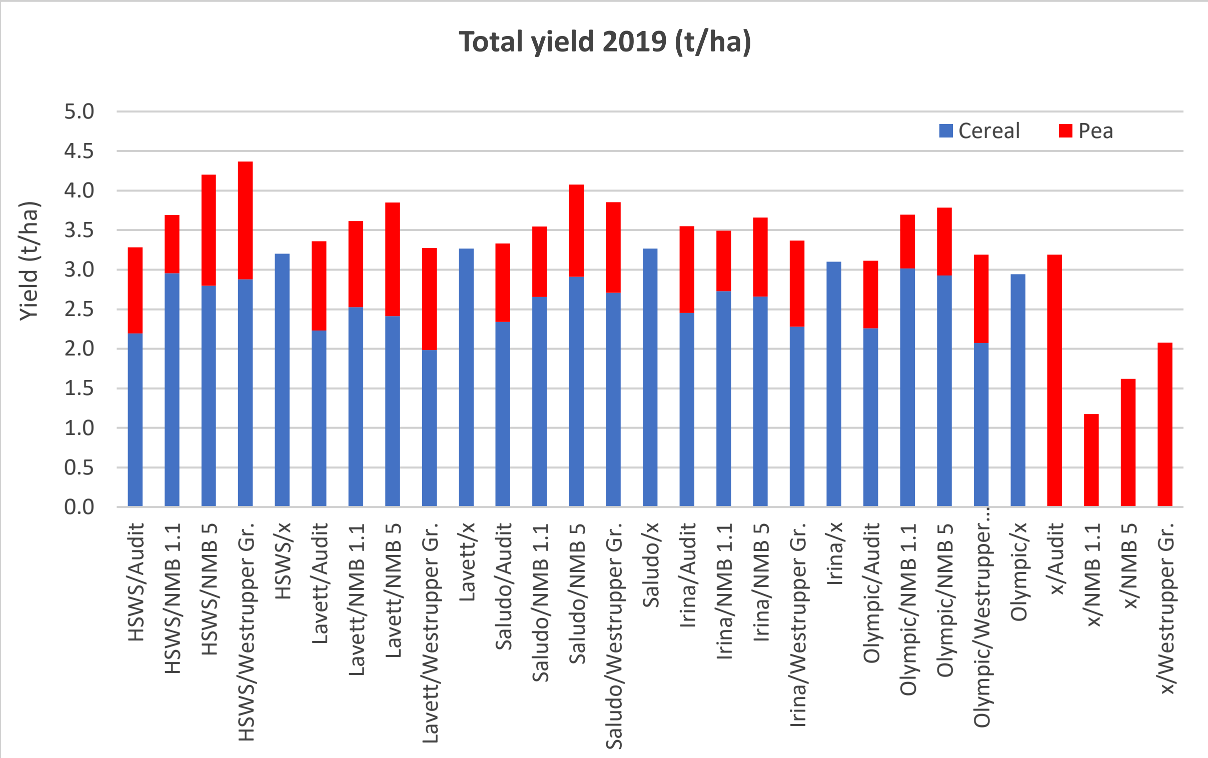

Figure 2. Average total yield (t/ha) for the different treatments in the mixed cropping trial with pea and cereals 2019. Treatment notation = cereal variety / pea variety or breeding line.

Figure 3 shows the average weed pressure of the different treatments in 2019.

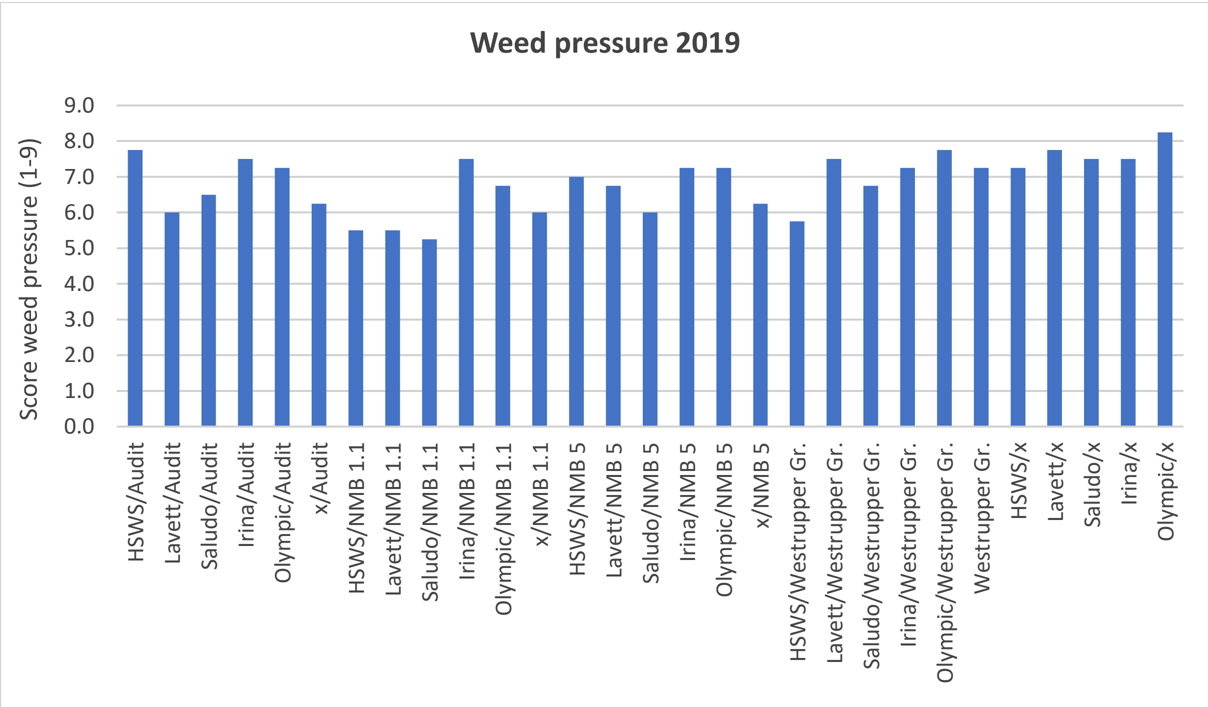

Figure 3. Average weed pressure ((9=no weeds present to 1=only weeds present) for the different treatments in the mixed cropping trial with pea and cereals 2019. Treatment notation = cereal variety / pea variety or breeding line.

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Conclusions

Yield

- Differences found between the treatments for total yield.
- Also differences in yield for pure stand and mixed treatments.
- Mixed stands show a higher yield compared to the yield of pure stand pea and pure stand cereal.

Weed pressure

- Less weed pressure in pure stand cereal treatments compared to most pea pure stand treatments (except the pea variety 'Westrupper Grijze'.
- Some mixed treatments show more suppression of weed compared to the pure stand pea treatments. Especially the mixed treatment with barley and oats shows a good suppression of weeds.

Discussion and next steps

Yield

The mixed pea/cereal treatments showed in almost all treatments a higher total yield (t/ha) compared to the pure stand pea treatments. This was probably caused by the higher weed pressure in most pure stand pea treatments and because of lodging of the plants in the pure stand treatments of pea. In the mixed crop treatments, the plants can grab onto the cereals to prevent lodging. So intercropping shows to be a good solution for lodging of pea plants.

Weed suppression

Weed suppression was not consistently lower in the mixed treatments compared to the pure stand pea treatments. The barley and oats mixed treatments show less weeds compared to the pure stand pea treatments, but differences were visible within the wheat treatments. For this reason, it is important to setup a larger trial to find out which varieties are suitable for mixed cropping with pea or that we have to setup a breeding program to breed for better varieties for mixed cropping with pea.

The transition from meat to plant-based proteins and the reduction of the use of imported soy, as a part of agro-ecology, is on the political agenda in the EU and in the Netherlands. This is one of the reasons for the interest of the agro and food community for the field days, workshops and magazine articles on protein crops and mixed cropping systems. The trials described are important for the development of suitable pea cultivars (in pure stand or in mixed cropping) under the prevailing pedo-climatic conditions.

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LBI - Faba bean - wheat mixtures (Trial #13)

Lead: LBI - The Netherlands.
Floor van Malland - f.vanMalland@louisbolk.nl, Abco de Buck - a.debuck@louisbolk.nl

Abstract

The mixing ability of different Faba bean varieties with different spring wheat varieties is determined in organic field trials in 2018, 2019 and 2020. Mixtures of species are compared with species in pure stand based on yield, protein content, development of diseases, plant height, ripening and general development of plants. The outcome of this research showed that for example the height of wheat and Faba bean is an important factor to perform well when growing intercropped. Wheat has the ability to grow higher and to compete with Faba bean. The results will allow a better understanding of traits important for successful mixed cropping of species.

Introduction

Objectives

The main aim of this trial is to identify traits important for mixed cropping of spring wheat and Faba bean. What are the differences in yield, diseases, plant development and ripening between de cultivars in mixed and pure stand? Are commercial varieties (Faba bean and spring wheat) suitable enough for mixed cropping and is there a difference between varieties how they adapt to mixed cropping?

Material and Methods

Location

The trial fields (2018-2020) were located within a commercial field with wheat at an organic farm in the central polder area (Kraggenburg, The Netherlands) on a limey, light marine clay soil.

Figure 1: Field Faba bean-wheat trial 2019 (photo taken by Floor van Malland 13-06-2019)

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

Genetic resources

<u>Wheat varieties, wheat populations, white lupin varieties and mixtures:</u>
Accessions only mentioned when tested in all 3 trial years.

Spring wheat:
- *Lavett (Agrifirm, The Netherlands)*
- *Quintus (Wiersum)*
- *Saludo (Dottenfelderhof, Germany)*

Faba bean:
- *Tiffany (Wiersum)*
- *Taifun (Wiersum)*
- *Columbo (DLF)*
- *Fuego (Wiersum)*
- *Fanfare (Wiesum)*
- *Victus (Wiersum)*
- *Lynx (Wiersum)*
- *Louhi (Boreal)*
- *Kontu (Boreal)*

Creation of experimental population

Intercropping

Experimental design

Year	Sowing	Harvest
2018	26-03-2018	30-07-2018
2019	02-04-2019	22-08-2019
2020	27-03-2020	19-08-2020

The trials were sown within a production field with a wheat population in 3 repetitions (plot size net 15m2, gross 12.75m2).

Sowing density:

- wheat pure stand: 300 seeds/m2
- Faba bean pure stand: 30 seeds/m2 (‘Louhi’ and ‘Kontu’ 40 seeds/m2)

mixture of wheat and Faba bean:
- wheat: 100 seeds/m2
- Faba bean: 30 seeds/m2 (‘Louhi’ and ‘Kontu’ 40 seeds/m2)

Evaluation

List of the traits collected for lupin and wheat concerning Crop development and agro-ecological performance, Crop productive performance and Crop quality performance.

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Germination	Counting plants	%

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Disease's wheat	Yellow rust (*Puccinia striiformis)*, Brown rust (*Puccinia recondita*) (1 = dead heavy infected to 9 = not infected), *Fusarium ssp.* and *Septoria tritici* (counting infected plants)	Scoring
Disease's Faba bean	Brown spots	Scoring
Plant development wheat	Development of spikes (1= stretching of flag leaf, 2= spike developing in flag leaf, 3=spike in top of flag leaf, 4=spike emerging, 5= spike totally emerged)	Scoring
Plant height	Measuring height Faba bean and wheat	Cm
Ripening plants wheat	Ripening, state of yellowing (Yellowing of total plant 1=green, 5=yellow)	Scoring

Table1. Crop development and agro-ecological performance traits

Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**
Yield	Harvesting per plot, dried (mixtures with Faba bean are separated) and cleaned	t/ha
Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		
Trait	**How it has been assessed**	**Type of data available**
Protein content	Laboratory Gent University	%
Falling number wheat	Laboratory Gent University	Sec
Zeleny wheat	Laboratory Gent University	Ml

Table 2. *Crop productive performance and Crop quality performance traits*

Statistical methods

Statistical analyses: ANOVA randomised block design

Participatory/multi-actor approach

Organisation of workshops and annual field days to inform and discuss results with growers, researchers, breeders, advisors, and other stakeholders of the value chain.

Results and discussion

Main results

Table 3 shows the average total yield (t/ha) of the different treatments in 2018, 2019 and 2020. Significant differences were found between the treatments (P<0,001, LSD=0,617, significant differences shown in table 3), between the three years (P<0,001, LSD=0,158, 2018; 4.64 t/ha, 2019; 5,80 t/ha (b), 2020; 3,86 t/ha (a)).

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Table 3: Average total yield (ton/ha) for the different treatments in 2018, 2019 and 2020. Treatment notation = wheat variety / Faba bean variety.

	Total yield (t/ha)				
Treatment	**2018**	**2019**	**2020**	**Mean**	
Quintus 1/3 + Lynx	5,55	6,80	*	6,26	(a)
Saludo 1/3 + Lynx	5,34	6,16	*	5,81	(ab)
Quintus 1/3 + Tiffany	5,56	6,68	5,04	5,77	(ab)
Quintus 1/3 + Fuego	6,10	6,65	4,57	5,73	(ab)
Quintus 1/3 + Victus	*	6,72	4,75	5,72	(ab)
Quintus 1/3 + Fanfare	*	6,49	4,73	5,60	(b)
Lavett 1/3 + Lynx	5,24	5,86	*	5,59	(b)
Saludo 1/3 + Fanfare	*	6,16	4,83	5,49	(bc)
Quintus 1/3 + Taifun	5,47	6,18	4,30	5,29	(bc)
Lavett 1/3 + Fuego	5,38	5,88	4,52	5,24	(bc)
Saludo 1/3 + Victus	*	5,94	4,54	5,23	(bc)
Lavett 1/3 + Fanfare	*	5,98	4,47	5,22	(bc)
Quintus 1/3 + Columbo	5,11	6,24	4,23	5,19	(bc)
Saludo 1/3 + Fuego	5,32	5,94	4,34	5,18	(bc)
Saludo 1/3 + Tiffany	4,85	6,19	4,37	5,15	(bc)
Lavett 1/3 + Tiffany	4,96	5,89	4,33	5,06	(bc)
Quintus 1/3 + Trumpet	*	*	5,06	5,06	(bc)
Lavett 1/3 + Victus	*	5,78	4,19	4,97	(c)
Saludo 1/3 + Taifun	4,79	5,74	4,30	4,95	(c)
Quintus + x	4,54	6,45	3,78	4,95	(c)
Lavett 1/3 + Taifun	4,76	5,45	4,39	4,87	(c)
Saludo 1/3 + Trumpet	*	*	4,83	4,83	(c)
Lavett 1/3 + Trumpet	*	*	4,75	4,75	(cd)
Lavett 1/3 + Columbo	5,17	5,45	3,74	4,74	(cd)
Lavett + x	4,18	6,14	3,59	4,66	(cd)
Saludo 1/3 + Columbo	4,79	5,43	3,81	4,66	(cd)
x + Lynx	4,38	4,73	*	4,58	(cd)
Saludo + x	4,16	6,02	3,44	4,56	(cd)
Lavett 1/3 + x	*	4,92	3,47	4,19	(d)
x + Fuego	3,90	5,02	3,08	4,00	(de)
x + Fanfare	*	4,96	3,00	3,97	(de)
x + Tiffany	4,06	4,88	2,87	3,91	(de)
x + Trumpet	*	*	3,72	3,72	(de)
x + Victus	*	5,08	2,11	3,58	(de)
x + Taifun	3,59	3,80	3,18	3,51	(e)
x + Columbo	3,36	4,09	1,73	3,02	(e)

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

Figure 4 shows the protein content of the different wheat pure stand and mixed treatments in 2018, 2019 and 2020.

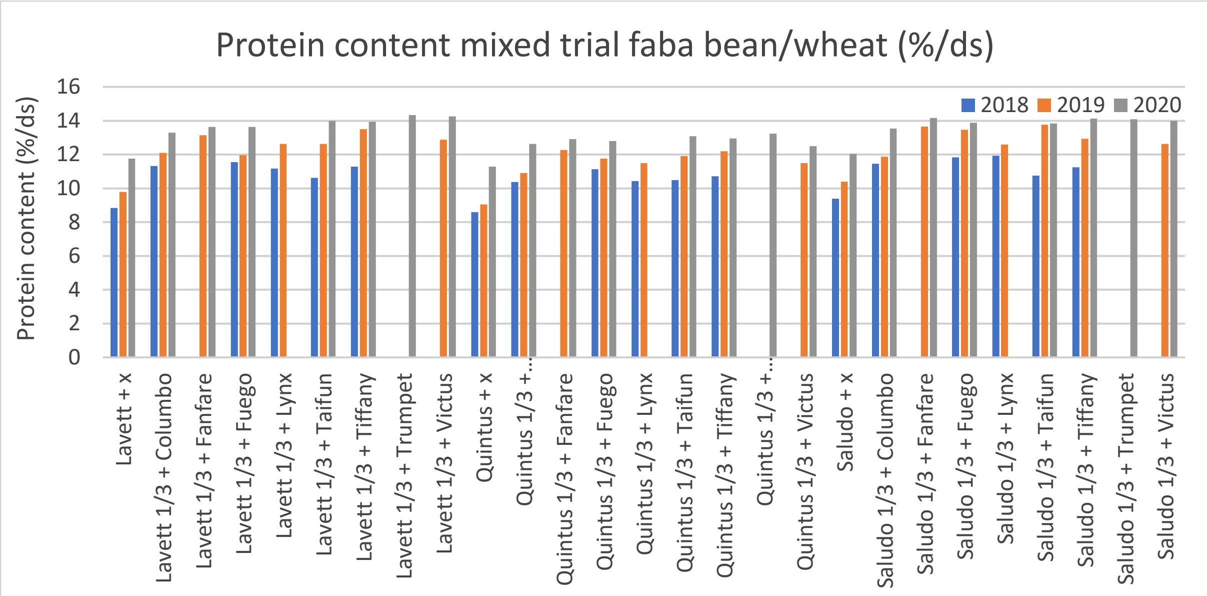

Figure 4. Protein content of the different wheat pure stand and mixed treatments in 2018, 2019 and 2020. Treatment notation = wheat variety /Faba bean variety.

Significant differences were found between the treatments for protein content using a two-way ANOVA (P<0,001, LSD=0,8524) and between the three years (P<0,001, LSD=0,2752). Also, significant differences were found between the three years (P<0,001, LSD=0,2752) but not for the treatment*year interaction (P=0,699). The average protein content of the wheat in 2018 was 10,69%, in 2019 12,05% and in 2020 13,24%.

Figure 5 shows the height of the wheat plants for the different treatments in 2018 and 2019. In 2020 the germination was very poor due to the dry spring, so this data is not considered when performing the statistical analysis.

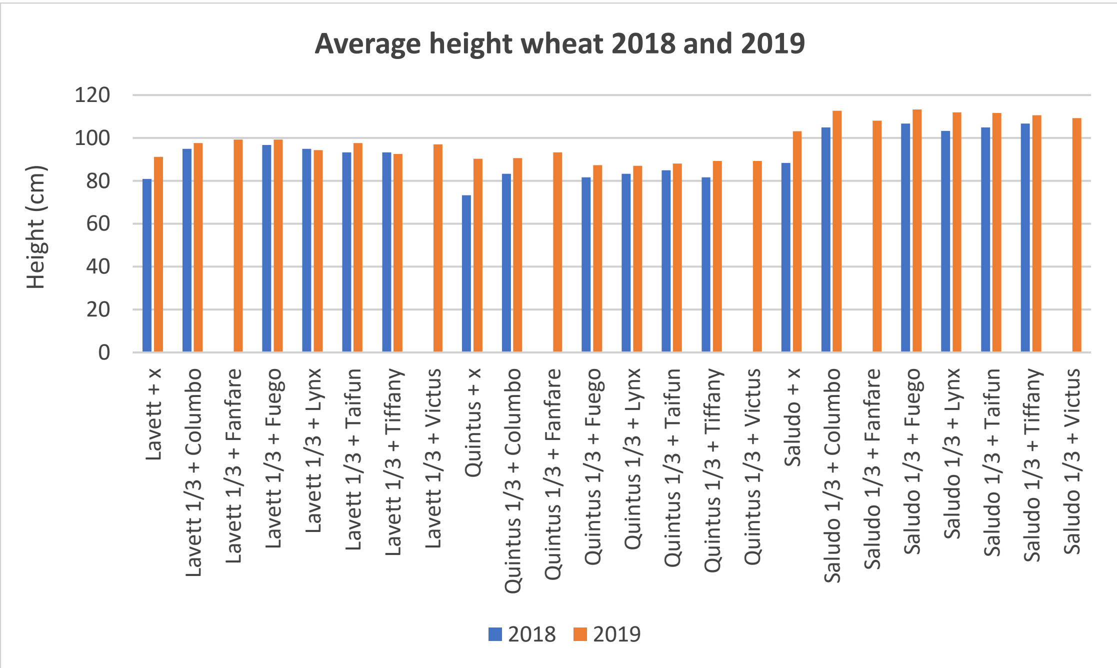

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Figure 5. Average wheat height of the different pure stand and mixed treatments in 2018 and 2019. Treatment notation = wheat variety / Faba bean variety.

Significant differences were found between the treatments for wheat height using a two-way ANOVA (P<0,001, LSD=0,8524) and between the three years (P<0,001, LSD=5,962). Also, significant differences were found between the two years (P<0,001, LSD=1,667) but not for the treatment*year interaction (P=0,107). The average wheat height in 2018 was 91,97 cm and in 2019 98,29 cm.

Figure 6 shows the height of the Faba bean plants for the different treatments in 2018 and 2019.

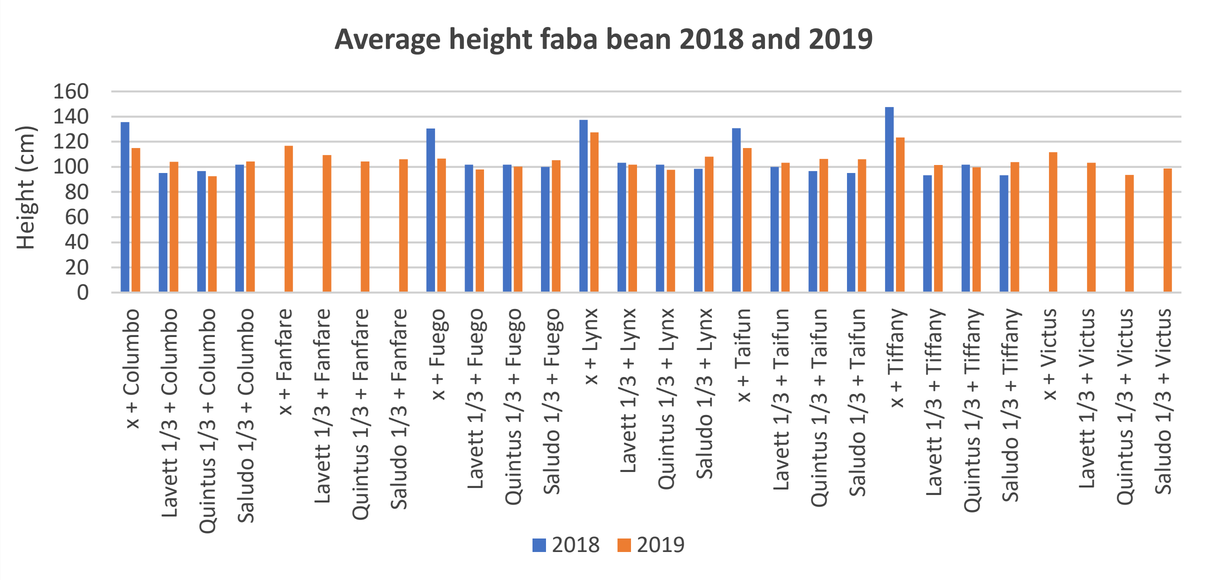

Figure 6. Average Faba bean height of the different pure stand and mixed treatments in 2018 and 2019. Treatment notation = wheat variety / Faba bean variety

Significant differences were found between the treatments for Faba bean height using a two-way ANOVA (P<0,001, LSD=0,8524) and between the three years (P<0,001, LSD=23,48). Also, significant differences were found between the two years (P=0,006, LSD=6,004) but not for the treatment*year interaction (P=0,815). The average Faba bean height in 2018 was 111,3 cm and in 2019 106,9 cm.

Conclusions

Yield

- Significant differences between the treatments for total yield.
- Mixed Faba bean/wheat treatments showed in almost all cases a significant higher total yield (t/ha) compared to the pure stand Faba bean treatments.
- Some mixed Faba bean/wheat treatments showed a significant higher total yield (t/ha) compared to the pure stand wheat treatments.
- Wheat in mixed cropping conditions have a significant higher protein content, compared to wheat in pure stand.

Height

- The mixed Faba bean/wheat treatments showed (for most treatments) significant higher wheat plants compared to the pure stand wheat treatments.

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- The mixed Faba bean/wheat treatments showed significant lower Faba bean plants compared to the pure stand Faba bean treatments.

Discussion and next steps

Yield
The mixed Faba bean/wheat treatments showed (in most cases) a significant higher total yield compared to the pure stand treatments. Intercropping shows to be a good solution to achieve higher yields with less weed management and to increase the biodiversity on the field.

Plant height
To compete with the Faba bean, spring wheat is able to grow higher. Interesting to see is that the Faba bean plants become smaller when growing intercropped.

Relevance
The transition from meat to plant-based proteins and the reduction of the use of imported soy, as a part of agro-ecology, is on the political agenda in the EU and in the Netherlands. This is one of the reasons for the interest of the agro and food community for the field days, workshops and magazine articles on protein crops and mixed cropping systems. The trials described are important for the development of suitable lupin cultivars (in pure stand or in mixed cropping) under the prevailing pedo-climatic conditions.

General information trials LBI

Results transferable to other situations

Compared to mono crops, total grain yield for spring wheat legume mixed crops is higher and more stable and baking quality of wheat improves significantly under the pedo-climatic conditions in the Netherlands. This cropping system has a better competitiveness to weeds compared to a mono legume crop (confirmed for lupin in our trials) and fits well within organic farm management and production for local bread bakers. For production for large-scale value chains, infrastructural issues must be solved (acceptance and sorting facilities for mixed product, product purity standards, etc.). Breeding for mixed cropping requires to evaluate both species in mixed stand, as the ranking in performance of pure stand crops differs from the ranking in mixed stand. When designing mixed stand wheat - legume evaluations, one or a few testers (cultivars) are sufficient (only GMA effects, no SMA).

Planned publications

We are planning a paper for **Sustainability: Special Issue "Evidence-Based Solutions for an Agro-Ecological Transition"** (Guest Editor: Szilvia Bencze and Dora Drexler) or to **Special Issue Sustainability "Breeding and Seed Sector Innovations for Organic Food Systems"** (Guest Editor: M. Messmer).

Improving resilience

In the early development stages, a wheat crop grows faster than a leguminous crop. Therefore, a mixed wheat-leguminous crop has a better resilience to wheat pressure. This was found statistically significant in the wheat – lupin mixed crop and in the wheat – pea mixture as an indication.
The wheat crop has the ability to fill up empty space within a crop. We found a higher proportion of wheat a mixture with lupin or Faba bean (about 60%) when growing conditions were less favourable for the legume species and a stabilisation of total grain yield.

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The hypothesis that genetic diversity in the field enhances resilience against diseases was not confirmed due to the low disease pressure in the wheat and legume crops. In the spring wheat-lupin mixed crop, we observed some brown rust in the lower parts of wheat plants in some plots, which was not present in the pure stand crop. This could be due to the higher relative humidity inside the mixed crop. This finding is contrary to the hypothesis, whilst only an indication.

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IUNG – PIB, JAROSŁAW STALENGA (Trial #15, 16, 20, 21)

Abstract
The purpose of the planned trial is to assess the productivity of various cereal-grain legumes mixtures and to indicate the most favorable variants of these mixtures in the conditions of organic farming. The trial was conducted in 2018-2020 (three seasons of vegetation) in Osiny IUNG-PIB Experimental Station (Eastern Poland). Additionally, in the trial cereal and grain legumes in pure stand have been used as control objects. The following crops have been considered: spring triticale, oat, narrowleaf (blue) lupin and yellow lupin. The research was focused on determination of the crop yields and calculation of LER.

Introduction

Hypothesis and research question
The hypothesis assumed was formulated as follows: mixtures of lupins with oat and spring triticale are better adapted to the cultivation in organic farming than the crops sown in pure stands.

Objectives
The objective of the trial was to assess the productivity of various cereal-grain legumes mixtures and to indicate the most favorable variants of these mixtures in conditions of organic farming, especially for poor/sandy soils. The following crops were considered: spring triticale, oat, narrowleaf (blue) lupin and yellow lupin. The focus in the research was on the crop yields. The trial should result in evaluation of the traits involved in general mixing ability of cereal-legumes mixtures.

Material and Methods

Location(s)
1. Osiny Experimental Station (Eastern Poland) N: 51°27'53.96'' E: 22°3'11.71''

Genetic resources
4 crops (2 cereals: oat and spring triticale and 2 grain legumes: yellow lupin and narrowleaf lupin)
1 cultivar of oat (Harnaś) and 1 cultivar of spring triticale (Mazur)
2 cultivars of yellow lupin: Perkoz (self-determinate cultivar) and Baryt (traditional cultivar)
2 cultivars of narrowleaf lupin: Regent (self-determinate cultivar) and Kurant (traditional cultivar)

Experimental design
The trial has been conducted between 2018 and 2020 within a long-term experiment aimed to compare different crop production systems established in 1994 in the IUNG-PIB Experimental Station in Osiny (eastern Poland).
The trial included 8 treatments with cereal-grain legumes mixtures, 2 with cereals in pure stand and 4 with two species of lupins in a pure stand. Each treatment had 4 repetitions. A randomized block design was applied to arrange the trial. In total there were 56 plots of 30 m² (3x10m) area each. These were the following treatments:

1. oat + yellow lupin (self-determinate cultivar),
2. oat + yellow lupin (traditional cultivar),
3. spring triticale + yellow lupin (self-determinate cultivar),

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4. spring triticale + yellow lupin (traditional cultivar),
5. oat + narrowleaf lupin (self-determinate cultivar),
6. oat + narrowleaf lupin (traditional cultivar),
7. spring triticale + narrowleaf lupin (self-determinate cultivar),
8. spring triticale + narrowleaf lupin (traditional cultivar),
9. yellow lupin in pure stand (self-determinate cultivar),
10. yellow lupin in pure stand (traditional cultivar),
11. narrowleaf lupin in a pure stand (self-determinate cultivar),
12. narrowleaf lupin in a pure stand (traditional cultivar),
13. oat in a pure stand,
14. spring triticale in a pure stand.

Evaluation

List of the traits collected in the trial Table 1.

Table 1. List of the traits collected for mixtures of oat, spring triticale, yellow lupin and narrowleaf lupine concerning agro-ecological and crop productive performance.

Trait	How it has been assessed	Type of data available
Days from sowing to full emergence	*BBCH 29-30 for cereals* *BBCH 15-20 for lupins*	days
Days from sowing to tillering (2-3 tillers detectable)	*BBCH 22-23 for cereals*	days
Days from sowing to end of heading (cereals)	*BBCH 59-60*	days
Days from sowing to beginning of flowering (lupins)	*BBCH 60-61*	days
Grain yield	grain yield from each plot harvested in t/ha at 15% moisture content: measurement from 4 randomly taken samples	t/ha
Land equivalent ratio (LER)	It is the sum of the relative yields of component species in the mixture as compared to their respective sole crops. LER= Y1M1+Y2M2 where Y1 and Y2 are the yields (per unit of total area of the intercrop) of species 1 and 2 in the mixture, and M1 and M2 are the yields of the species in sole crops (per unit area of the respective sole crop).	

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Statistical methods
The analysis of variance of grain yields was done with use of the statistical programme STATISTICA ver. 7.1. The significance of the difference was evaluated at the α = 0.05 significance level with the Tukey test.

How resilience was improved
The results of the research revealed higher resilience of the mixtures in comparison to the crops sown in pure stands.

Participatory/multi-actor approach
The species (lupins and cereals) and their mixtures were selected for the trial based on the consultations with the agricultural advisors closely cooperating with organic farmers.

Results
In the period of 2018-2020 high yields of mixtures of spring cereals with lupins were noted. Mixtures of oat with lupins yielded about one ton higher than mixtures of spring triticale with lupins and these differences were statistically significant. Narrowleaf lupin in a pure stand gave about twice higher yields than yellow lupin, whereas in case of cereals oat yielded better and gave about one-ton higher yield that spring triticale.
The results of own research showed that there were no significant differences in yielding between self-determinate and traditional cultivars of lupins.
It was shown higher values of LER for mixtures with yellow lupin than with narrowleaf (blue) lupin for both cereals. However, LER for spring triticale with yellow lupin was higher than for oat (Table 2).

Table 2. Yields and Land Equivalent Ratio (LER) of different mixtures of spring cereals with lupins (2018-2020)

Crop or mixture of crops	Yield in t/ha*	Land Equivalent Ratio (LER)
Mixtures of oat with lupins		
Oat + Yellow lupin (SDC**)	5,37a	**1,26**
Oat + Yellow lupin (TC**)	5,53a	**1,17**
Oat + Narrowleaf lupin (TC)	5,47a	**1,08**
Oat + Narrowleaf lupin (SDC)	5,58a	**1,12**
Mixtures of spring triticale with lupins		
Spring triticale + Yellow lupin (SDC)	4,33b	**1,38**
Spring triticale + Yellow lupin (TC)	4,60b	**1,24**
Spring triticale + Narrowleaf lupin (TC)	4,34b	**1,10**
Spring triticale + Narrowleaf lupin (SDC)	4,35b	**1,14**
Crops in a pure stand		
Spring triticale	4,25a	
Oat	5,45a	
Yellow lupin (TC)	0,78b	
Yellow lupin (SDC)	0,82b	
Narrowleaf lupin (TC)	1,50b	
Narrowleaf lupin (SDC)	1,56b	

*a, b - values with different letters significantly differ at $p < 0.05$. ** TC – traditional cultivar, SDC – self-determinate cultivar

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

Discussion

Lupins cover the largest sown area of all grain legumes cultivated in Poland. Within this group yellow and blue lupins dominate.

In 2021 31 cultivars of blue lupin were registered in Poland, of which 25 traditional and 6 self-determinate ones. Blue lupin due to its higher yielding, faster ripening, and low susceptibility to anthracnose, is sown on better, more fertile soils. The cultivars are quite diverse. The most important features differentiating them include utilization profile, growth type, flower and seed color and their quality features, length of the growing season. However, there is one important common feature of almost all cultivars, i.e., low content of alkaloids, therefore they can be a valuable protein component in fodder production. Within traditional cultivars there were 2 ones with high concentration of alkaloids, which makes them very useful as catch crops or green manure.

As far as the yellow lupin is concerned in 2021 there were only 10 cultivars registered in Poland, of which 8 were traditional and 2 self-determinate. Yellow lupin has the highest protein content in seeds and therefore can be used for fodder production, but also as a catch crop or a green manure. It may be sown on light, sandy soils with low pH. Plants of this species develop a strong and deep root system, which has the ability to uptake nutrients from deeper soil layers, as well as accumulate symbiotically fixed nitrogen.

Self-determinate cultivars for both yellow and blue lupins have been recently developed to widen the potential cultivation area for these crops. These cultivars are characterised by a shorter vegetation period and earlier, more uniform ripening and the absence or strong reduction of lateral shoots. However, having these features resulted in their lower yielding level in comparison to the traditional cultivars (Prusiński 2007). The results of own research did not confirm better yielding performance of traditional forms of both lupins. Slightly higher yields were only noted for traditional cultivar of yellow lupin in a mixture with oat and with a triticale.

The results of the research showed high yielding performance of mixtures of spring cereals with lupins, but especially with yellow lupin, as expressed in high values of LER. Nelson et al. (2012) indicated that organic intercrops consisting of cereals and grain legumes had the ability to give higher yields and reached also high LER at the level of 1.64 compared with their component monocrops. Similarly, Bulson et al. (1997) reported high LER value for wheat–field bean intercrop in the organic system.

Outcomes and conclusions

Main outcomes:

- High values of LER noted for mixtures of oat/triticale with lupins demonstrated their good mixing ability
- Mixtures of lupins with oat performed better (higher yields and lower variability in years) than mixtures with spring triticale
- LER results showed that yellow lupin in a mixture performed better than narrowleaf lupin
- Self-determinate cultivars have a good potential for their wider utilization in organic farming

Main lessons learned:

Cultivation of grain legumes in pure sown is a big challenge in conditions of organic farming, especially in the context of the climate change.

Mixtures of lupines with cereals can be recommended for organic farmers, especially on poor, sandy soils, however specific measures should be developed and implemented to encourage stockless organic farmers to cultivate them.

How the information can be transferred to the other situations?

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The outcomes of the research can be easily transferred to the organic practice during trainings, field days and by electronic and paper dissemination materials.

Publications

No references yet, one presentation planned for OWC in Rennes in September 2021

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FiBL-CH – Blue (*Lupinus angustifolius*) and White (*Lupinus albus*) Lupin mixed cropping and undersowing trials (Trial #17 and #18)

Lead: Christine Arncken, Monika Messmer (FiBL-CH)

Abstract

FiBl conducted plot trials in 2015-16-17 (trials from 2017 reported here) to select most suitable cereal for mixed cropping with blue lupin. Give the results of these trials, FiBL now supports introduction of blue lupin into Swiss organic agriculture with a mixed cropping regime. From 2018, larger-scale experiments with farmers started.

Both mixed cropping (2017-2018) and undersowing (2020-2021) trials were conducted in white lupin. As crop mixtures did not render the envisaged success in 2017 and 2018, from 2019 the main white lupin cultivar trial is conducted in pure stand. An undersowing trial was conducted in 2020 to test for suitable living mulches to suppress weeds in the late summer in white lupin. The trial is repeated in 2021.

Introduction

Hypothesis and research question

Mixed cropping may support the improvement of yield stability, especially when cereals are mixed with grain legumes, as the cereals can counter-balance low grain-legume production in unfavourable years. Mixed cropping of lupin with a cereal cash-crop or with an undersowing companion crop may reduce weed pressure. Mixed cropping and undersowing may reduce anthracnose pressure through the barrier that the companion crop poses to secondary infection.

This activity aimed at identifying suitable companion crops for mixed cropping and/or undersowing with blue and white lupin in Swiss growing conditions.

Objectives

In Switzerland, local production of grain legumes is important in organic farming for reducing soybean imports. Mixed cropping of grain legumes with cereals has become a success among commercial organic farmers in Switzerland (e.g., barley-pea mixtures for feed production) thanks to the support of FiBL extension service that provides advise to farmers on this practise.

In LIVESEED, with the aim to increase grain legume diversification, FiBL activities focused on blue and white lupin with the objective to test whether mixed cropping or undersowing are useful practises to use in lupin production in Switzerland.

Material and Methods

Location(s)

Table1. Locations of trials in 2017, 18 and 2020

Location	2017	2018	2020
Mellikon, Kanton Aargau, High Rhine Valley, Switzerland	X	X	
Feldbach, Kanton Zürich, near Zürich Lake, Switzerland		X	
Full-Reuenthal, Kanton Aargau, High Rhine Valley, Switzerland			X

Plant materials used in the trials

2017

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Blue lupin (Lupinus angustifolius) trial
Blue lupin cv. Boruta (Saatzucht Steinach, DE)
Spring oat Buggy (Nordsaat Saatzucht, DE)
Spring barley Eunova (Saatzucht Düdingen, CH)
Triticale Trado (FiBL extension service western Switzerland)
Spring wheat Fiorina (FiBL extension service western Switzerland)
Red fescue cv. Reverent (Otto Hauenstein Samen AG, CH)
Spring triticale ARTI 8 (breeding line, Getreidezüchtung Peter Kunz (GZPK), CH)

White lupin (Lupinus albus) trial
White lupin cv. Feodora (Jouffrai-Drillaud, FR)
Winter triticale 06TC07.49 (breeding line, Getreidezüchtung Peter Kunz, (GZPK), CH)
Triticale Trado (FiBL extension service western Switzerland)
Winter oat Delfin (Nordsaat Saatzucht, DE)
Spring oat SH 395-12 (breeding line, Getreidezüchtung Dottenfelder Hof, DE)
Red fescue cv. Reverent (Otto Hauenstein Samen AG, CH)
Isatis tinctoria (Zollinger.Bio Seeds, CH)

2018
White lupin cv. Amiga (Florimond-Desprez, FR)
White lupin BLU 25 (breeding line, Semillas Baer, Chile)
White lupin Dieta (Soya UK, GB)
White lupin cv. Feodora (Jouffrai-Drillaud, FR)
White lupin cv. Figaro (Jouffrai-Drillaud, FR)
White lupin cv. Sulimo (Jouffrai-Drillaud, FR)
White lupin cv. Zulika (Oseva, CZ)

Spring triticale cv. Clayton (IG Pflanzenzucht GmbH, DE)
White clover cv. Hebe (Otto Hauenstein Samen, CH)
Cameline cv. Omega (Getreidezüchtung Peter Kunz, CH).

2020
Undersowing trial
Lupin cultivars
White lupin cv. Frieda (DSV, DE)
White lupin cv. Sulimo (Jouffrai-Drillaud, FR)

Crops used for the undersowing
White clover (Trifolium repens cv. Hebe (Hauenstein seeds, CH)
Cameline (Camelina sativa) cv. Omega (Getreidezüchtung Peter Kunz, CH)
Mixture DSV Humus plus VORSAAT (90 % festuca rubra (=growing tuft-like), 10 % festuca ovina) (Deutsche Saatveredelung, DSV, DE)
Mixture DSV M2 (90% Lolium perenne, 10 % Trifolium repens) (DSV, DE)
UFA shade lawn mix (UFA seeds, CH)
Plantago lanceolata (Sativa Rheinau, from Hortus officinarum, CH)

Experimental design
2017
Trials sown on 06.04.2017, following reduced tillage of a turnip rape catch crop. Preceding crop: maize for silage. No fertilizer was added to the trial. Trials were randomised blocks with 3 replicates. Plot

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size was 7,5 m^2. Standard lupin sowing density (=100%) was 130 seeds/m^2 of **blue** lupin cv. Boruta (undetermined, non-branching) and 65 seeds/m^2 of **white** lupin Feodora. In mixtures, seed density was 80% of pure stand for lupins, 20% for cereals. Mechanical weed control with a combined hoe/curry-comb machine on 23.05.2018. Blue lupins were harvested on 21.07.2017, white lupins on 09.08.2017.

Figure 2. FiBL Lupin trials in 2017

2018

Trials sown on 25.03.2018 in Feldbach and 09.04.2018 in Mellikon. The preceding crop was vegetables in Feldbach and turnip rape catch crop after oats in Mellikon. Ploughing was done on 31.12.2017. No fertilizer was applied. Trials were randomised blocks with 2 replicates on each site. Plot size was 3,07 m^2 in Feldbach and 7,5 m^2 in Mellikon. Standard lupin sowing density (=100%) was 65 seeds/m^2. In mixtures, seed density was 80% resp. 100 % of pure stand for lupins, 40% for triticale. Mechanical weed control (hoeing) on 25.04.2018 in Feldbach and on 08.05.2018 in Mellikon (with a combined hoe/curry-comb). Harvest: 02.08.2018 (Feldbach) and 07.08.2017 (Mellikon). Statistical analysis still ongoing.

2020

In Full-Reuenthal lupins were sown on 24.03.2020 and the undersowing crops on 03.04.2020. The preceding crop in Full-Reuenthal: was sorghum.

Mechanical weed control in Full-Reuenthal was carried on 23rd April (curry-comb) and 7th May (hoeing). The lupin was harvest on 19th August. The undersowing trial was structured in a randomized block design with 3 replicates. Plot size was 4,62 m^2. The standard lupin sowing density of 65 seeds/m^2 was used in this trial.

Evaluation

Table2. Locations of traits assessed in 2017, 18 and 2020

Trait	Assessment description	Data type
Crop development and agro-ecological performance		
2017		
Field emergence lupins	Counting of plants in two randomly chosen 1m sections of two rows	Quantitative (3 reps) (plants/m^2)
Soil coverage of crop plants	Estimating on three dates for blue lupin, one date for white lupin.	Semi-quantitative (3 reps) (% soil coverage)
Total performance at maturation (only blue lupins)	Visual classification of whole plot	Semi-quantitative (3 reps) (scoring 1-9)

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Weed coverage of soil after harvest (only blue lupins)	Visual scoring	Semi-quantitative (3 reps) (scoring 1-9)
Anthracnose, mid-July (only white lupin)	Visual scoring	Semi-quantitative (3 reps) (scoring 1-9)
2018		
Field emergence lupins	Counting of plants in two randomly chosen 1m sections of two rows	Quantitative (1 site/2 reps) (plants/m^2)
Field emergence triticale	Counting of plants in two randomly chosen 1m sections of two rows	Quantitative (1 site/2 reps) (plants/m^2)
Soil coverage of crop plants	Estimating whole plot in May	Semi-quantitative (1 site, 2 reps) (% soil coverage)
Lupin development stage	Visual scoring of whole plot at three (Feldbach) resp. four dates (Mellikon)	quantitative (1 or 2 reps) (BBCH scale)
Percentage of healthy lupin plants in mid June	Counting of all plants in the four inner rows of plot (Mellikon)	quantitative (1rep) (%healthy plants per emerged plants)
Plant height at flowering	One measure per plot	quantitative (Mellikon 1 rep, Feldbach 2 reps) (cm)
Anthracnose, several dates	Repeated visual scoring of plots (several dates)	Semi-quantititative (2 sites, 1-2 reps) (scoring 1-9)
Lodging	Visual scoring of plots, end of July	Semi-quantititative (2 sites, 2 reps) (scoring 1-9)
2020		
Field emergence lupins	Counting of plants in two representative meters of rows of each plot (not at the edge of the plot), later conversion	Quantitative (3 reps) (emerged plants/m^2)
Soil coverage of lupins 19th May	Estimating lupin soil coverage in %	Quantitative (3 reps)
Soil coverage of weeds 19th May	Estimating weed soil coverage in %	Quantitative (3 reps)
Anthracnose score 25th June	Giving disease scores on a scale from 1 (=no symptoms) to 9 (=all plants dead) on whole plot level (see document: Scoring_protocol_field_antrachnose)	Quantitative (3 reps)
Development of undersowing 25th June	Scoring on a scale from 1 to 9 with 1=no plants; 9=all plants have germinated and are visible	Quantitative (1 site/3 reps) (score)
Lupin soil coverage at harvest (19th August)	Estimating lupin soil coverage in %	Quantitative (1 site/ 3 reps)
Weeds soil coverage at harvest)19th August)	Estimating weed soil coverage in %	Quantitative (3 reps)
Crop productive performance		
2017		
Lupin yield	Harvesting of whole plot, separating, and cleaning yield, H_2O correction, weighing of grains	Quantitative (3 reps) (t/ha)
Cereal yield	Harvesting of whole plot, separating, and cleaning yield, H_2O correction, weighing of grains	Quantitative (3 reps) (t/ha)
Land equivalent ratio (LER)	Calculating yields: (lupin in mixture/lupin pure stand) +(cereal in mixture/cereal pure stand)	Quantitative (3 reps) (Index: <1= pure stand is better; >1 = mixture is better)
2018		
Lupin yield	Harvesting of whole plot, separating, and cleaning yield, H_2O correction, weighing of grains	Quantitative (3 reps) (t/ha)

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Cereal yield	Triticale yielded nearly zero. Not assessed	No data
	2020	
Yield	Measured as g per plot after cleaning and measuring H_2O content with a Wile 55 measuring device. Calculated as t/ha yield with a dm content of 86%.	Quantitative (1 sites/3 reps) (t/ha)

Statistical methods

ANOVA and post-hoc test for yield comparison, LER calculation.

Participatory/multi-actor approach

Farmers and advisors as well as with feed mill owners were involved in the evaluation of the trial results. The undersowing trials in 2020 was conducted following the request by a farmer to investigate new possibilities of undersowing catch crops under white lupins in order to prevent late season weed development.

Results

2017

Total yield in mixed cropping of **blue lupins** was better than pure stand (1,4 t/ha), and it was quite similar for all cereals (ca. 2,1 t/ha). LER was >1 for oat and triticale. Under the current Swiss agricultural conditions, the economic outcome for the farmer (including all direct payments) was best for the mixture with winter triticale (120 % of the contribution margin of blue lupin pure stand).

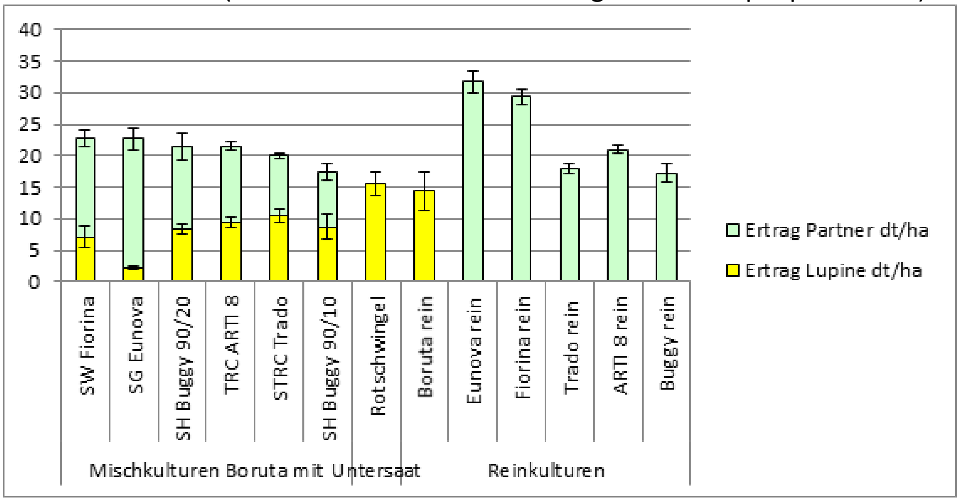

Figure 3. Yield (dt/ha) in mixed cropping of blue lupins in 2017

Pure stand of **White lupins** yielded 2,6 t/ha. Only the treatments with oats yielded more than 3 t/ha, irrespective of pure stand or mixed cropping, but the mixed treatment contained only very little lupins even though only 20% of the pure seed density had been sown. Mixed cropping rendered no improvement concerning weed suppression but a tendency of anthracnose reduction. Contribution margin was best for pure stand of lupin. The best mixed cropping treatment with winter triticale 06TC07.49 rendered 86,7% of the pure stand contribution margin.

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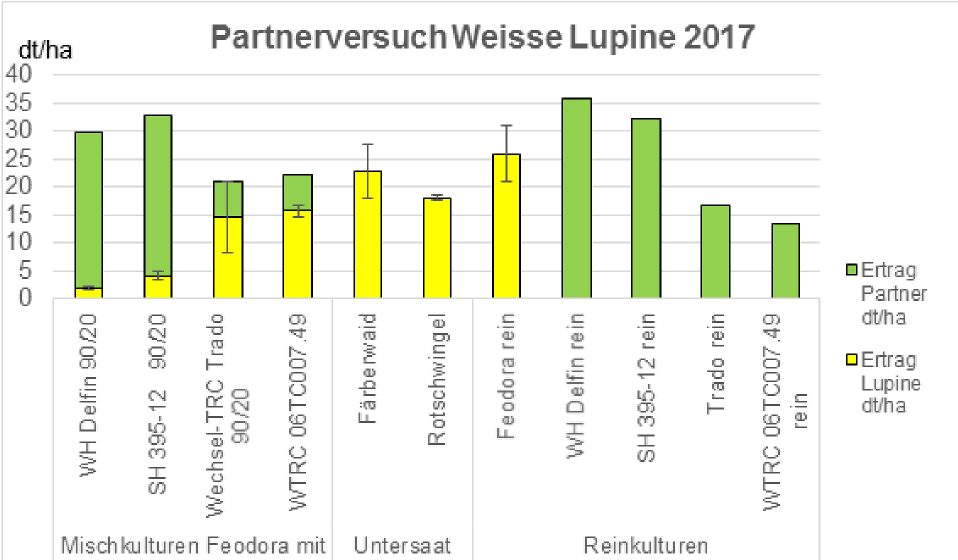

Figure 4. Yield in mixed cropping of white lupins

2018

After sowing, the Mellikon trial lacked rain for two weeks which delayed germination of the lupins, however, in May lupin field emergence was about as planned (65 plants/m^2), whereas triticale emergence was only 60-80 % of the planned density (180 plants/m2). White clover and cameline didn't grow at both sites. In Mellikon, soil coverage in May was better in both mixed treatments with triticale than in pure stand of both crops, reflecting the higher total number of sown seeds. However, unlike 2017, anthracnose scores were not better in the mixed treatments. Lupin yields in pure stands were 4,8 t/ha in Feldbach and 2,3 t/ha in Mellikon. In Feldbach, mixed cropping reduced the lupin yield to 3,9 and 3,7 t/ha in the 100/40 and 80/40 triticale mixtures, respectively. In Mellikon, yield reduction was less severe (1,9 and 2,1 t/ha, respectively). On both sites, nearly no triticale was harvested.

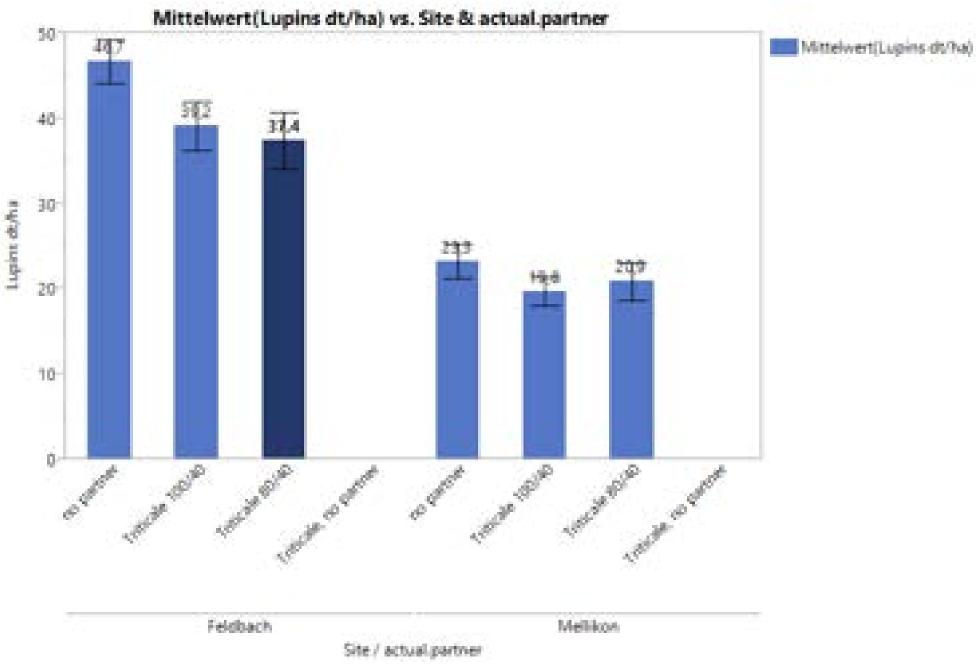

Figure 5. Yield results trial in 2018

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2020

At sowing time, the soil was already dry, and after sowing, there was no precipitation until April 28th. However, the lupins germinated and developed well, whereas hardly any germination of the undersowing crops was observed. On 25th June, the shade lawn mixture showed the best development, whereas plantago and cameline were hardly visible. After harvest, ground coverage of the shade lawn mixture was higher than that of the other undersowings, but still low (max. 7 %). However, there was a highly significant (P = 0,0027) effect of undersowing on lupin yield, with the shade lawn treatment leading to a mean lupin yield of 3,32 t/ha (compared to a mean lupin yield in pure stand of 2,69 t/ha).

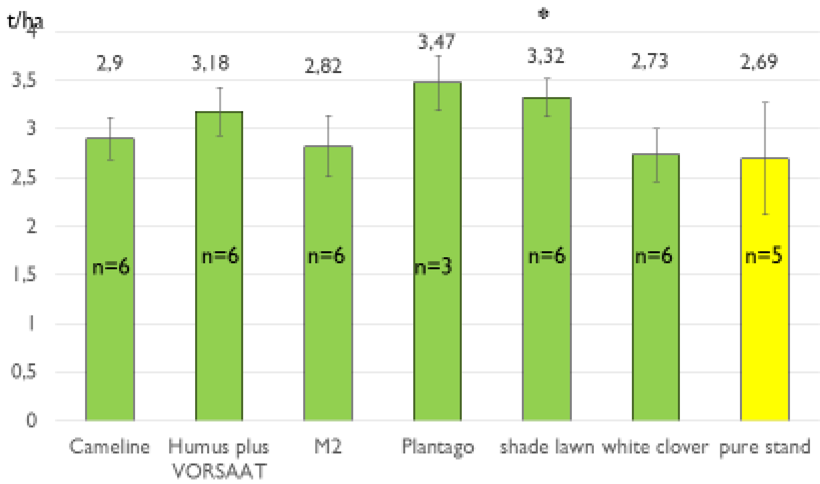

Fig.6: Lupin yield (t/ha) in the undersowing trial in Full-Reuenthal (CH) 2020.

In this trial, only the shade lawn treatment was significantly different from the pure stand treatment. Plantago was only sown in three plots and can thus not be fully compared to the other treatments. There was a clear interaction between undersowing and lupin cultivar. The effect of the undersowing was significantly higher in the cultivar Sulimo that had a significantly lower yield due to anthracnose.

Discussion

The results suggest that for the **narrow-leafed "blue" lupin species** *(Lupinus angustifolius)*, mixed cropping is a solution to weed problems in organic farming and can help to establish this additional legume crop to Swiss organic farming. Swiss organic feed mills have invested in machinery for the separation of grain legume-cereal mixtures and are ready to accept lupin mixtures and pay a higher price for them compared to the pea price as soon as enough farmers start growing lupins. Breeding should take account of the fact that better-adapted genotypes could be identified if breeding took place under mixed cropping conditions. FiBL supports introduction of blue lupin into Swiss organic agriculture with a mixed cropping regime. Small-plot field trials were run until 2017. In 2018, larger-scale experiments with farmers started.

In contrast, the performance of the **broad-leafed white lupin (*Lupinus albus)*** is mainly limited by anthracnose, and by maturation time (FiBL cultivar trials, data not shown). With the partners tested to date, no advantage of mixed cropping was seen. Weed problems mainly arise towards the end of the growing season when leaves have completely fallen down. A partner that is good in suppressing weeds in that period without disturbing harvest is still lacking. Breeding should focus on anthracnose first; mixed cropping seems less suitable for this crop in Swiss growing conditions.

In fact, to date, we have not found a good cropping partner for white lupin. Although triticale had been the best partner in the previous years, the white lupin-triticale mixture rendered no advantages

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compared to the pure stand. Under the dry conditions of 2018, white lupin outcompeted triticale as well as the weeds. Early sowing is vital for yield production and has contributed to the much higher yields in 2018. The results of the **undersowing trial** in 2020 were surprising because the undersowing crops developed poorly due to the 2020 spring drought, whereas *Chenopodium album* weeds developed very well. The trial is repeated in 2021 to produce more robust data.

Outcomes and conclusions

Main outcomes

Mixed cropping of *blue lupins (Lupinus angustifolius)* with oat or triticale is now recommended to farmers, while for *white lupins* (*Lupinus albus*) mixed cropping has no advantage under spring sowing conditions in Switzerland. Difficult to find suitable companion crops and undersowing plants for white lupin due to long vegetation period (mid-March – August/September). Strong vigor of white lupins in spring - especially under drought conditions - and early canopy coverage due to larger leaf areal compared to small leaved blue lupins results in suppression of less drought tolerant companion crops.

Main lessons learned

The advantages of mixed cropping need to be pondered against key questions:
-species related questions (e.g., in FiBL CH trials, mixed cropping *blue lupin- cereal* gave better results than mixed cropping *white lupin - cereal* and first results of *white lupin – undersowing* because white lupin is more competitive, overgrows companion and undersowing plants, thus also reduction of spread of anthracnose disease was not achieved)
-crop rotation related questions (problems we wanted to address like weed pressure might be solved more efficiently with diversity in the rotation than with diversity of mixed cropping)
-market related questions (use companion crops which not only have a good agronomic potential in mixtures, but also have a market potential and price, mixtures with cereals might cause problems if the aim is gluten free products)
- important to define objective which performance and ecosystem functions should be improved, which is many cases is not total yield of mixture

How the information can be transfer to the other situations?

Results related for example on (I) growth cycle alignment of the two companion crops, (ii) optimizing sowing density, (iii) effect of site/season specific climate on the competition between the two crops, (iv) general aspects to take into account (e.g., market, crop rotation, traces of gluten), can be useful for other experiences on mixed cropping, (v) involvement of mills who are able to separate crops after harvest is essential to upscale trials.

Publications

- References, i.e., publications from your work on LIVESEED

Paper at EUCARPIA Conference 2018 (book of abstracts), publication in bioaktuell targeting farmers, video together with ReMIX
Scientific paper planned to be submitted in autumn 2021.

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

LBI Lupin - wheat mixtures (Trial #19)

Lead: LBI - The Netherlands.
Floor van Malland - f.vanMalland@louisbolk.nl, Abco de Buck - a.debuck@louisbolk.nl

Abstract

The mixing ability of different lupin varieties with different spring wheat varieties is determined in organic field trials in 2018, 2019 and 2020. Mixtures of species are compared with species in pure stand based on yield, protein content, development of diseases, plant height, flowering and ripening, general growth of plants and plant architecture. The outcome of this research showed that for example height and number of pods of the lupin variety are important factors to perform well in intercropping conditions with wheat. The results will allow a better understanding of traits important for successful mixed cropping of species.

Introduction

Objectives

The main aim of this trial is to identify traits important for mixed cropping of spring wheat and white lupin. What are the differences in yield, diseases, plant architecture and development and quality of cereals between de cultivars in mixed and pure stand? Are commercial varieties (white lupin and spring wheat) suitable for mixed cropping and is there a difference between varieties how they adapt to mixed cropping? Can we say something about the general or specific mixing ability?

Material and Methods

Location

The trial fields (2018-2020) were located within a commercial field with wheat at an organic farm in the central polder area (Ens, The Netherlands) on a limey, light marine clay soil.

Figure 1: Field lupin-wheat trial 2018 (photo taken by Floor van Malland 30-05-2018)

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Genetic resources

Wheat varieties, wheat populations, white lupin varieties and mixtures:

Accessions only mentioned when tested in all 3 trial years.

Spring wheat:

- *Lennox (Agrifirm, The Netherlands)*
- *Arabella (Danko, Poland)*
- *Alderon (KWS, UK)*
- *Heliaro (Dottenfelderhof, Germany)*
- *Harenda (Limagrain)*
- *Population HS-2-08 (original)*
- *Population Convinto C (original)*

White Lupin:

- *Boros*
- *Butan*

Mixtures:

- *Boros & Lennox*
- *Boros & Arabella*
- *Boros & Alderon*
- *Boros & Heliaro*
- *Boros & Harenda*
- *Boros & Population HS-2-08*
- *Boros & Population Convinto C*
- *Butan & Lennox*
- *Butan & Arabella*
- *Butan & Alderon*
- *Butan & Heliaro*
- *Butan & Harenda*
- *Butan & Population HS-2-08*
- *Butan & Population Convinto C*

Creation of experimental population

Intercropping

Experimental design

Year	Sowing	Harvest
2018	10-04-2018	20-08-2018
2019	11-04-2019	03-09-2019
2020	08-04-2020	18-09-2020

The trial was sown in 2 repetitions (plot size gross 10,5 m2, net 8,25 m2).

Sowing density:

- wheat pure stand: 300 seeds/m2
- white lupin pure stand: 50 seeds/m2

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mixture of wheat and white lupins:
- wheat: 100 seeds/m2
- white lupin: 50 seeds/m2

Evaluation

List of the traits collected for lupin and wheat concerning crop development and agro-ecological performance, crop productive performance and crop quality performance.

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Germination	Counting plants	%
Disease's wheat	Yellow rust, Brow rust (1=dead heavy infected to 9=not infected) Fusarium and Septoria (counting infected plants)	Scoring
Disease's lupin	Brown spots	Scoring
Plant development lupin, architecture	Counting number of pods main stem, 1ste branch, 2nd branch etc. Counting number of branches.	Scoring
Plant development wheat	Development of spikes (1= stretching of flag leaf, 2= spike developing in flag leaf, 3=spike in top of flag leaf, 4=spike emerging, 5= spike totally emerged)	Scoring
Plant height	Measuring height	cm
Ripening plants	Ripening, state of yellowing (Yellowing of total plant 1=green, 5=yellow)	Scoring

Table 1: Crop development and agro-ecological performance traits

Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**
Yield	Harvesting per plot, dried (mixtures with lupin are separated) and cleaned	t/ha = 1.000kg/ha
Crop quality performance (organoleptic, processing, nutritional, nutraceutical, immaterial)		
Trait	**How it has been assessed**	**Type of data available**
Protein content	Laboratory Ghent University	%

Table 2: Crop productive performance and crop quality performance traits

Statistical methods

Statistical analyses: ANOVA randomised block design

Participatory/multi-actor approach

Organisation of workshops and annual field days to inform and discuss results with growers, researchers, breeders, advisors, and other stakeholders of the value chain.

Results and discussion

Main results

Figure 2 shows the average total yield (t/ha) of the different treatments in 2018, 2019 and 2020. Significant differences were found between the treatments (P<0,001, LSD=0,5347, shown in Figure 2),

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between the three years (P<0,001, LSD=0,225, 2018; 5,05 t/ha (b), 2019; 5,26 t/ha (b), 2020; 2,36 t/ha (a)) and for the treatment*year interaction (P<0,001).

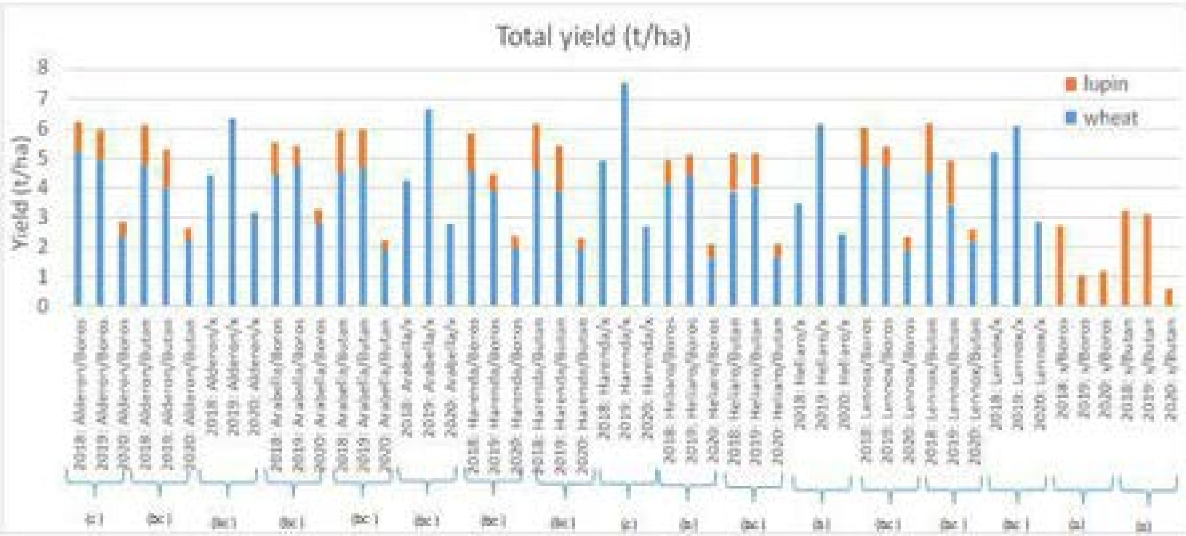

Figure 2: Average total yield (ton/ha) for the different treatments in 2018, 2019 and 2020. Treatment notation = year: wheat variety / white lupin variety.

Figure 3 shows the average plant height of wheat for the different treatments in 2018, 2019 and 2020.

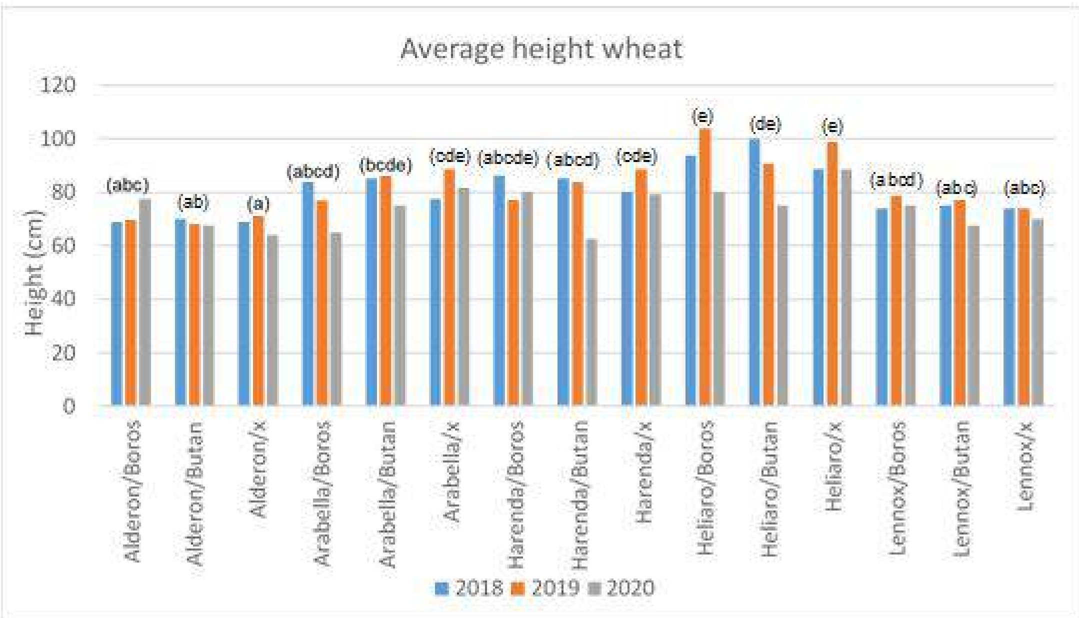

Figure 3: Average height of wheat for the different treatments in the white lupin/wheat trial conducted in 2018, 2019 and 2020 in The Netherlands. Treatment notation = wheat variety / white lupin variety.

Significant differences were found between the treatments for wheat height, using a two-way ANOVA (P<0,001, LSD=7,829 differences are shown in figure 3). Also, significant differences were found

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between the three years (P<0,001, LSD=3,501, 2018; 80,67cm (b), 2019; 82,17cm (b), 2020; 73,87cm (a)) and not for the treatment*year interaction (P=0,098).

Figure 4 shows the average plant height of white lupin for the different treatments in 2018, 2019 and 2020.

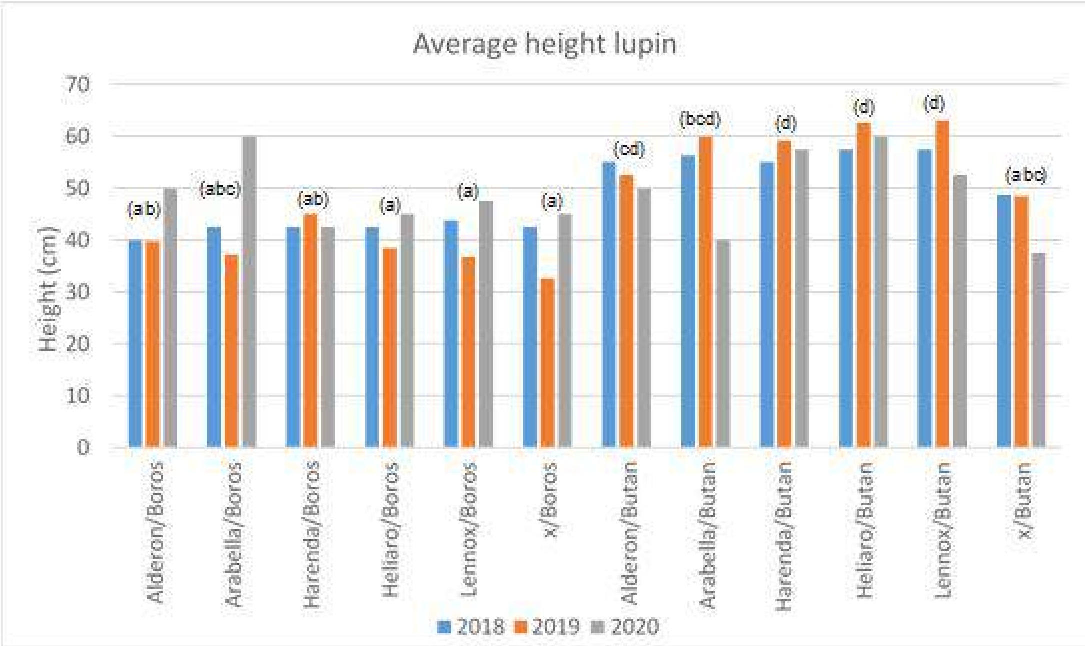

Figure 4: Average height of lupin for the different treatments in the white lupin/wheat trial conducted in 2018, 2019 and 2020 in The Netherlands. Treatment notation = wheat variety / white lupin variety, x = mono crop.

Significant differences were found between the treatments for lupin height using a two-way ANOVA (P<0,001, LSD=8,850 differences are shown in Figure 4). No significant differences were found between the three years (P=0,985) and for the treatment*year interaction (P=0,159). Compared to a pure stand crop, 'Butan' shows a significant higher crop in combination with 'Heliaro', 'Harenda' and 'Lennox'. 'Boros' does not show any significant increase in crop height when grown as a mixed crop.

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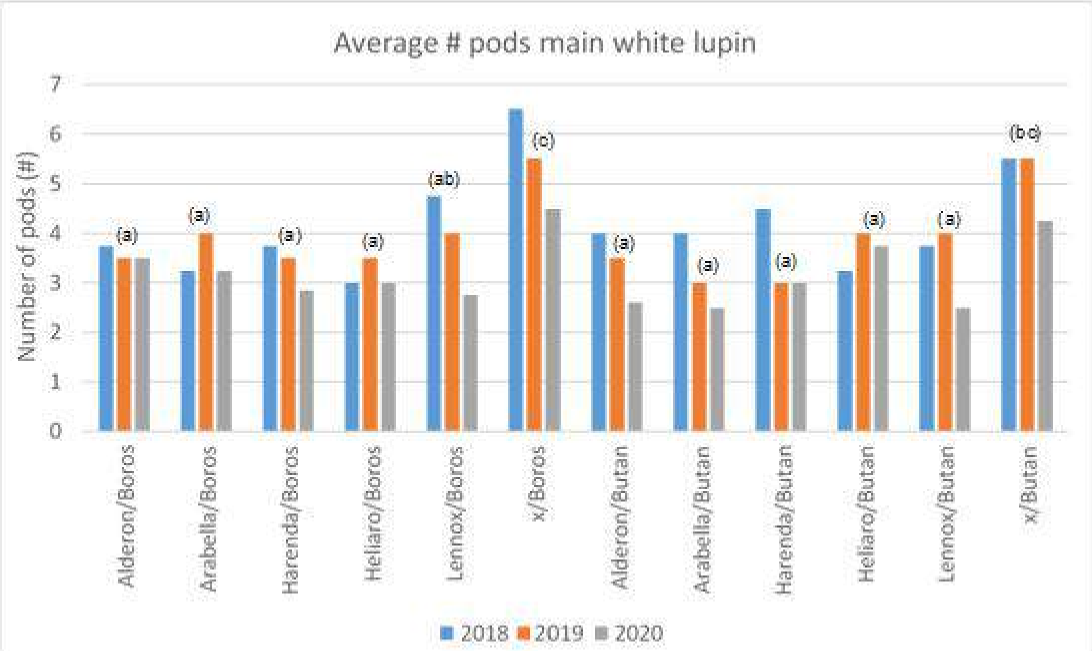

Figure 5: Average number of pods of the main stem of white lupin for the different treatments in 2018, 2019 and 2020. Treatment notation = wheat variety / white lupin variety.

Figure 5 (previous page) shows the average amounts of pods of the main stem of the white lupins for the different treatments in 2018, 2019 and 2020. Significant differences were found between the treatments for the number of pods on the main stem (P<0,001, LSD=0,7834, shown in figure 5), between the three years (P<0,001, LSD=0,3917, 2018; 4,17 (b), 2019; 3,92 (b) and 2020; 3,2 (a)), but not for the treatment*year interaction (P=0,399).

General Mixing Ability (GMA) effects

Lupin genotype GMA effect

Figure 6 shows the mean lupin yield and the mean wheat yield for the 2 lupin genotypes in the mixed cropping trials (2018-2019).

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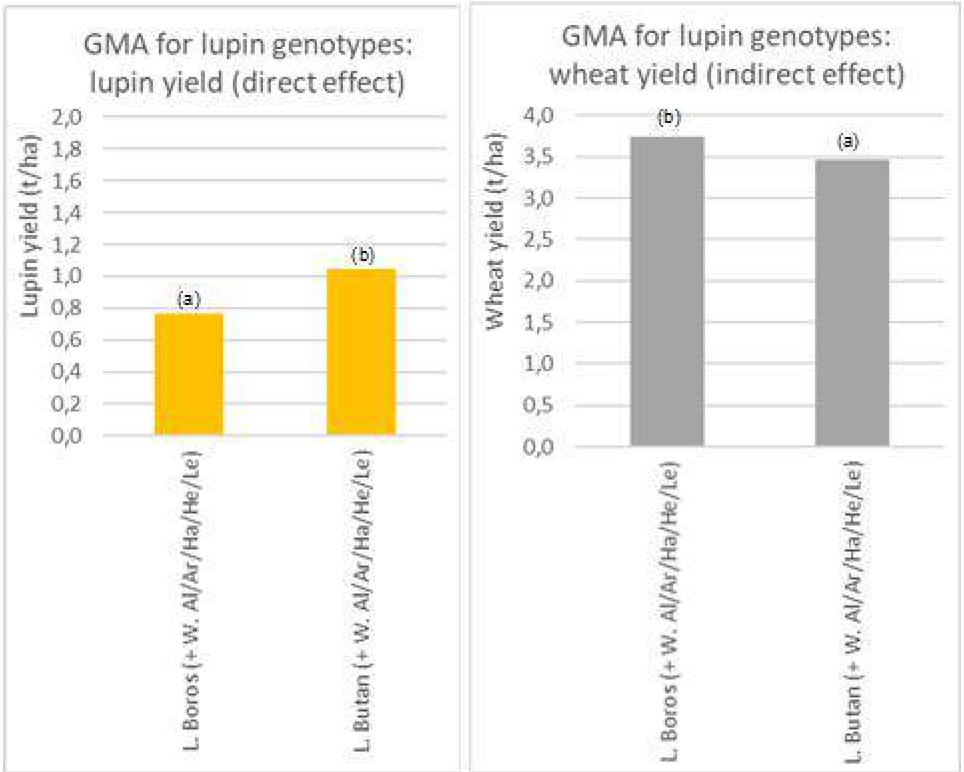

	P (treatment)	LSD (treatment)	P (year)	LSD (year)	P (treatment*year)	LSD (treatment*year)
Lupin yield	<0,001	0,0970	<0,001	0,1188	<0,001	0,1680
Wheat yield	0,025	0,2437	<0,001	0,2984	<0,001	0,4220

Figure 6: direct and indirect GMA effects of lupin genotypes determined by the average wheat yield (ton/ha) and lupin yield for the 2 different lupin genotypes in the mixed cropping white lupin/wheat trial conducted in 2018, 2019 and 2020 in The Netherlands.

Wheat genotype GMA effect

Figure 7 shows the mean lupin yield and the mean wheat yield for the 5 wheat genotypes in the mixed cropping trials (2018-2019).

	P (treatment)	LSD (treatment)	P (year)	LSD (year)	P (treatment*year)	LSD (treatment*year)
Wheat yield	0,004	0,3575	<0,001	0,2769	0,375	0,6192
Lupin yield	0,314	0,2279	<0,001	0,1765	0,706	0,3947

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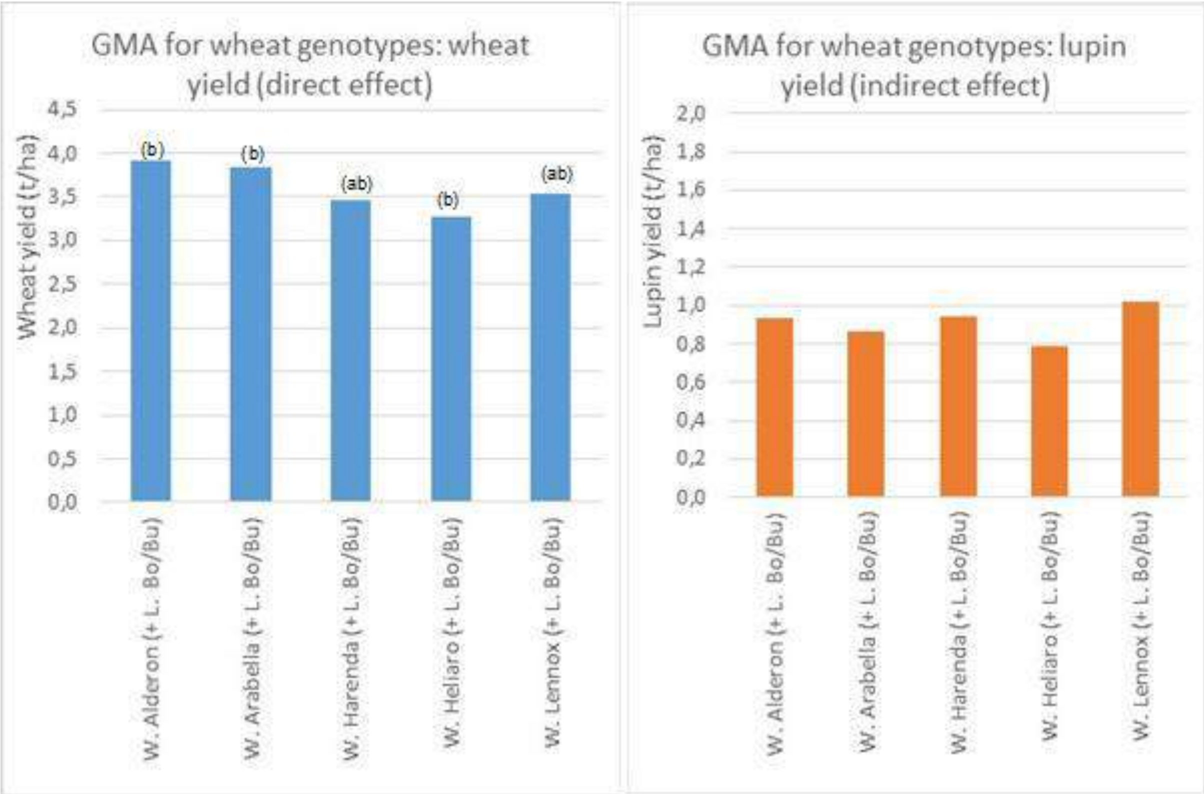
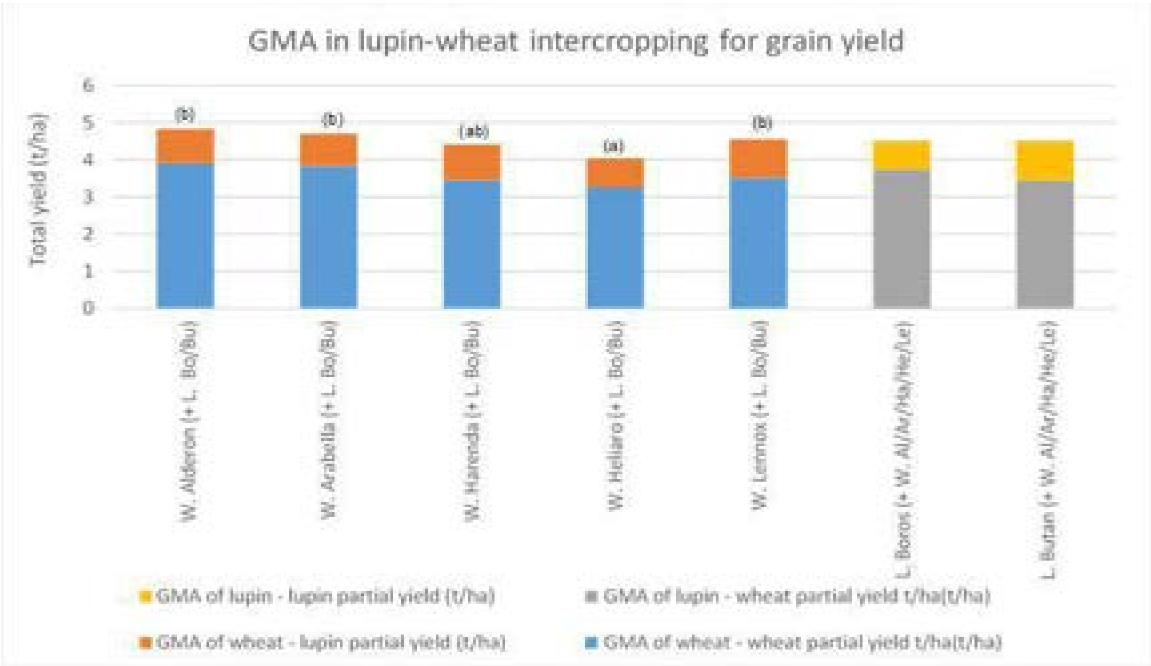


Figure 7: Direct and indirect GMA effects of wheat genotypes determined by the average wheat yield (ton/ha) and lupin yield for the 5 different wheat genotypes in the mixed cropping white lupin/wheat trial conducted in 2018, 2019 and 2020 in The Netherlands.

GMA effect for total yield

Figure 8 shows the mean total yield for the 5 wheat genotypes and the 2 lupin genotypes separately in the mixed cropping trials (2018-2019)



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	P (treatment)	LSD (treatment)	P (year)	LSD (year)	P (treatment*year)	LSD (treatment*year)
total yield wheat genotypes	<0,001	0,3615	<0,001	0,28	0,176	0,6262
total yield lupin genotypes	0,965	0,2735	<0,001	0,335	0,481	0,4737

Figure 8 GMA effect for 5 wheat genotypes and 2 lupin genotypes determined of the average total yield (ton/ha) in the mixed cropping white lupin/wheat trial conducted in 2018, 2019 and 2020 in The Netherlands.

Specific Mixing Ability (SMA)

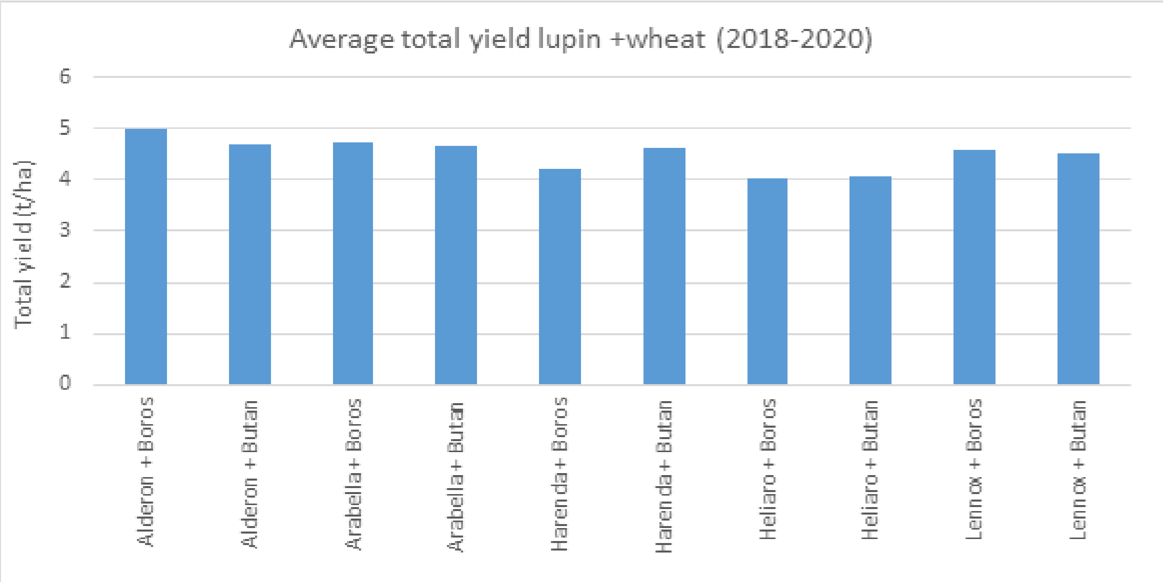

Figure 9: Average total yield (ton/ha) of the mixed treatments in the mixed cropping white lupin/wheat trial conducted in 2018, 2019 and 2020 in The Netherlands. Treatment notation = wheat variety / white lupin variety.

Table 3 shows the statistical results to determine if there is a SMA effect found for the years separately.

Table 3: Statistical results of SMA effect for total yield (t/ha) in the mixed cropping white lupin/wheat trial conducted in 2018.2019 and 2020 in the Netherlands.

	2018	2019	2020
Average total yield wheat genotype Alderon	6,15	5,62	2,76
Average total yield wheat genotype Arabella	5,70	5,65	2,74
Average total yield wheat genotype Harenda	6,00	4,91	2,32
Average total yield wheat genotype Heliaro	5,00	5,09	2,05
Average total yield wheat genotype Lennox	6,09	5,12	2,45
Average total yield lupin genotype Boros	5,70	5,25	2,57
Average total yield lupin genotype Butan	5,88	5,30	2,35
P (wheat genotype)	0,035	0,101	0,110
LSD (wheat genotype)	0,747	0,656	0,591
P (lupin genotype)	0,411	0,782	0,225
LSD (lupin genotype)	0,473	0,415	0,374

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P (wheat genotype*lupin genotype)	0,969	0,093	0,244
LSD (wheat genotype*lupin genotype)	1,057	0,928	0,836

Conclusions

Yield

- Significant differences between the treatments for total mixed yield.
- Mixed lupin/wheat treatments showed a significant higher total yield (t/ha) compared to the pure stand lupin treatments.
- Mixed lupin/wheat treatments showed no significant differences in total yield (t/ha) compared to the pre stand wheat treatments.
- LER is > 1 (in almost all treatments), proving that mixing crops is profitable.
- Wheat in mixed cropping conditions has a significant higher protein content, compared to wheat in pure stand.

Height

- The mixed lupin/wheat treatments showed no significant differences in wheat height compared to the wheat height of the pure stand wheat treatments (for the specific varieties).
- Compared to pure stand, the white lupin variety 'Butan' grew significantly higher in mixed stand with the wheat varieties 'Lennox', 'Heliaro' and 'Harenda'.

Plant architecture lupin

- The number of pods on the main stem was significantly lower in the mixed stands, compared to the pure stand lupin crop. No significant differences were found between the different mixed treatments.

General mixing ability (GMA)

- General mixing ability effect for lupin genotype is significant for lupin (direct effect) and wheat yield (indirect effect)
- General mixing ability effect for wheat genotype is only significant for wheat yield (direct), but not for lupin yield (indirect)
- General mixing ability effect for total yield is only significant for wheat genotype and not for lupin genotype.

Specific mixing ability (SMA)

- No significant SMA effect was found (no significant wheat genotype*lupin genotype interaction) for all years.

Discussion and next steps

Yield

The mixed lupin/wheat treatments showed a significant higher total yield compared to the pure stand lupin treatments. The high weed pressure probably reduced the yield of the pure stand lupin treatments. So intercropping shows to be a good solution for weed suppression when cultivating lupins. The pure stand wheat treatments showed no difference in total yield compared to the mixed lupin/wheat treatments, indicating the compensation, or 'filling' capacity of the wheat crop.

Plant height

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Observed differences in plant height for spring wheat are caused by the genetics of the different varieties. The wheat varieties did not perform differently in a mixed or pure stand condition. All wheat varieties were higher compared to the lupin plants, so the lupin did not compete with wheat in this respect. The lupin variety 'Butan' was able to grow higher and to adapt in a mixed cropping situation with certain wheat varieties but did not result in a higher lupin yield. This was not found for the lupin variety 'Boros'.

Plant architecture lupin

The number of pods on the main stem is a good predictor for yield (correlation coefficient for 'Butan': r=0,6887 and 'Boros': r=0,7851). Both varieties showed a significantly lower number of pods on the main stem in mixed stand conditions compared to pure stand. This can be caused by the height of these specific tested lupin varieties. They were too short to compete with the higher wheat. New higher lupin varieties/breeding lines show a better adaptation to mixed cropping with wheat (some were tested in the trial of 2020) and give a higher yield.

GMA and SMA effects

General mixing ability (GMA) effects were significantly present in the experiments. This indicated that specific breeding for intercropping is needed for species exposed to competition with another species in mixed cropping. No effects of SMA were found. This is comforting for breeding, meaning that each species could be bred for wide adaptation to intercropping conditions by selection with one or a few testers of the associated species.

Relevance

The transition from meat to plant-based proteins and the reduction of the use of imported soy, as a part of agro-ecology, is on the political agenda in the EU and in the Netherlands. This is one of the reasons for the interest of the agricultural and food community for the field days, workshops and magazine articles on protein crops and mixed cropping systems. As opposed to Lupin, Faba beans and peas are well-known leguminous crops in the Netherlands. The trials described are important for the development of suitable lupin cultivars (in pure stand or in mixed cropping) under the prevailing pedo-climatic conditions.

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AGROSCOPE AND NARDI - Forage mixture breeding (Trial #23a and #23b)

Lead: AGROSCOPE (Switzerland) - Christoph Grieder
Partner: NARDI Fundulea, Prof. Ion Toncea

Abstract

Cultivation of forage mixtures offers several advantages over monocultures, but forage legumes like alfalfa (*Medicago sativa* L.) are mostly bred in pure stands. Thereby, possible interactions of alfalfa with its companion grasses are not accounted for and may lead to lower selection efficiency compared to direct selection in mixed stands. In a main experiment, we assessed accession-by-cultivation system interaction when alfalfa plants are grown in pure stands or in an easily adaptable nursery system with their companion grasses. Spaced plants of 50 accessions were grown on bare soil as control treatment (CONV), in a sown sward of short growing lawn cultivars of tall fescue (*Festuca arundinacea* Schreb.) and red fescue (*F. rubra* L.) (LAWN), and in a sown sward of forage cultivars of the same species (FORA). Accession-by-cultivation system interaction variances were large for plant habitus but small for plant vigor. Phenotypic correlation coefficients (r_p) among the cultivation systems were high for plant vigor, whereby LAWN was somewhat more predictive for FORA ($r_p = 0.83$) than CONV ($r_p = 0.77$). The LAWN cultivation system might be a good compromise that allows accounting for given competition effects among species, but still allowing an easy assessment of traits in the nursery.

To be used as a genetic resource and to assess the realized genetic progress from the three experimental cultivation systems in the main experiment (CONV, FORA, LAWN), nine different experimental populations (three per cultivation system) were produced via polycross. Seeds were produced in fall 2020 at AGROS and are currently being grown at AGROS and NARDI in plots to determine realized selection gain.

Introduction

Hypothesis and research question

Extensive literature reviews summarizing current theories and experimental data of selecting forage legumes and grasses for mixture have recently been published. Data assessed in previous experiments so far has led to different conclusions about the importance of breeding/testing in mixtures. Based on experiences from cultivar testing trials in Switzerland with different forage crops, it has been concluded that a good cultivar in monoculture will also show good performance in mixture. In comparison, correlations between pure stands and mixtures with grasses for white clover (*Trifolium repens* L.) were found to be low to moderate, and some authors conclude that direct selection of white clover in mixture would be more effective than selection in pure stands. For the case of alfalfa in mixture with grasses, generally positive, moderate to strong correlations between performance in pure stand and mixture were observed but lower correlations in elite breeding material might lead to changes in ranking of genotypes, making direct selection in the mixture again promising.

Adapting alfalfa to the conditions encountered in the mixture early in the breeding process would be desired and most straightforward. To select the parental plants for good performance in mixture, the corresponding selection pressure should already be early applied in the spaced plant nursery field. Therefore, we wanted to evaluate the effect of selection of spaced alfalfa plants that are grown in

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monoculture versus mixtures within a densely sown sward of tall fescue and red fescue. Such a simple spaced plant breeding nursery setup could easily be implemented in a breeding program.

Objectives

Three cultivation systems for a spaced plant breeding nursery of alfalfa were evaluated: spaced plants of alfalfa grown on bare soil as control treatment (CONV), in a sown sward of short growing lawn cultivars of tall fescue (*Festuca arundinacea* Schreb.) and red fescue (*F. rubra* L.) (LAWN), and in a sown sward of forage cultivars of the same species (FORA). Thereby, the objectives were (1) to assess the extent of accession-by-cultivation system interaction, (2) to determine a suitable selection system for selection in mixture, and (3) to produce seeds of experimental populations that have been selected in the three systems to assess realized selection gain in later plot trials.

Material and Methods

Genetic resources

The main experiment was based on 50 different accessions of alfalfa, of which 25 are landraces, ecotypes, or old cultivars (accession 1 to 25) and 25 are currently registered cultivars (accession 26 to 50, see Table 1 below)

Table 1. List of alfalfa (Medicago sativa L.) accessions used in the experiment

Nr	Name	Type[1]	Source / Breeder	Origin
1	Grimm	LR	IPK	CAN
2	HunterRiver	LR	IPK	AUS
3	Flamande_2	LR	IPK	FRA
4	AltfraenkischeBW	LR	IPK	DEU
5	IPK_LE864	ECO	IPK	SUN
6	Taschkentskaja_721	ECO	IPK	UZB
7	Taschkentskaja_1728	ECO	IPK	UZB
8	Taschkentskaja_3192	ECO	IPK	UZB
9	CN-39465	LR	GRIN	GBR
10	Miechowska	LR	GRIN	POL
11	Alta-Sierra	LR	GRIN	CHL
12	Criolla	LR	GRIN	BOL
13	X910013	LR	GRIN	CHN
14	GRIN_262541	ECO	GRIN	BUL
15	FC-34511	NA	GRIN	NZL
16	No.527	ECO	GRIN	CAN
17	VIR-24712	ECO	GRIN	RUS
18	No.218	ECO	GRIN	RUS

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19	No.2838	ECO	GRIN	POL
20	No.16272	ECO	GRIN	CHE
21	No.16273	ECO	GRIN	CHE
22	S-225	ECO	GRIN	RUS
23	CIN-1956	ECO	GRIN	GBR
24	Bobrava	CV	GRIN	CZE
25	Tuna	CV	GRIN	SWE
26	Artemis	CV	Barenbrug	NLD
27	Sanditi	CV	Barenbrug	NLD
28	Alexis	CV	Barenbrug	NLD
29	Alpha	CV	Barenbrug	NLD
30	Bardine	CV	Barenbrug	NLD
31	Rachel	CV	Caussade	FRA
32	Excelle	CV	Caussade	FRA
33	Eride	CV	Continental	ITA
34	Gea	CV	Continental	ITA
35	Robot	CV	CRA-FLC	ITA
36	Creno	CV	DLF Trifolium	DNK
37	Power_4.2	CV	DLF Trifolium	DNK
38	Europe	CV	DLF Trifolium	DNK
39	Eugenia	CV	D'Eugenio Sementi	ITA
40	Blue-Moon	CV	Natura s.l.r.	ITA
41	Daphne	CV	Florimond Desprez	FRA
42	Timbale	CV	Gie Grass	FRA
43	Galaxie	CV	Jouffray-Drillaud	FRA
44	Felicia	CV	Jouffray-Drillaud	FRA
45	Luzelle	CV	Jouffray-Drillaud	FRA
46	SK-Rasa	CV	LLU Research Institute of Agriculture	LVA
47	Cannelle	CV	R2n	FRA
48	Fusion	CV	Schmidt-Gambazza	FRA
49	Fiesta	CV	Schmidt-Gambazza	FRA
50	Fleetwood	CV	Saatzucht Steinach	DEU

[1] LR = landrace, ECO = ecotype, CV = cultivar.

Creation of experimental population

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In the main experiment, plants of the 50 original accessions were grown to follow up the first two objectives (i.e. (1) to assess the extent of accession-by-cultivation system interaction and (2) to determine a suitable selection system for selection in mixture).

Mass selection of single plants out of the main experiment and synthesis of new populations via polycross was performed to follow up the third objective (i.e. (3) to produce seeds of experimental populations that have been selected in the three systems to assess realized selection gain in later plot trials). For each cultivation system, (i) a first polycross was done using the 25 best plants according to an index summarizing all traits assessed ("best"), (ii) as second polycross was done using 25 plants with an intermediate performance according to the same index and with plants being from the same original accessions as for the "best" polycross ("intermediate"), and (iii) a third polycross was done using unselected plants of the same original accessions as for the "best" and "intermediate" polycross that were grown from seed ("base"). Seeds of the resulting nine experimental populations (CONV_best, CONV_int, CONV_base, FORA_best, FORA_int, FORA_base, LAWN_best, LAWN_int, LAWN_base) were produced in fall 2020 at AGROS and are currently being grown at AGROS and NARDI in plots to determine realized selection gain.

Experimental design

The spaced alfalfa plants were grown in three cultivation systems: alfalfa plants on bare soil (CONV), alfalfa plants in a sward of shallow growing lawn type grasses (LAWN), and alfalfa plants in a sward of tall growing forage type grasses (FORA). For each of the 50 accessions, 30 plants were grown per cultivation system, resulting in 4500 spaced plants in total. Plants of the CONV cultivation system were grown on a conventionally managed field and plants of the LAWN and FORA cultivation systems on an adjacent organically managed field. A typical breeding nursery design was used, with ten alfalfa plants of the same accession being planted in a row. On the conventionally managed field, three blocks of 50 rows (1 row per accession) were formed, all of them receiving the CONV treatment. On the organically managed field, six blocks of 50 rows were formed, alternately receiving the LAWN and the FORA treatment. This resulted in a split-plot like design with blocks and rows representing two different strata of main-plots.

Evaluation

Traits were recorded on a single plant basis before each cut (one cut in autumn 2017, four cuts in 2018, one cut in May 2019). Plant vigour was rated on a scale from 1 = high to 9 = low. Crown and root rot caused by *Sclerotinia trifoliorum* and downy mildew caused by *Peronospora trifoliorum* were rated upon occurrence from 1 = no symptoms to 9 = highly infected. Additional traits assessed visually were plant habitus (1 = erect, 9 = prostrate), stem diameter (1 = thin, 9 = thick), and leaf-to-stem ratio (1 = high leaf proportion, 9 = low leaf proportion).

Statistical methods

Analysis of variance (ANOVA) of data from the main experiment was computed using mixed models. Thereby, variance components could be estimated. Phenotypic correlations among the cultivation systems were calculated as Pearson's correlation coefficients based on accession means per cultivation systems (n = 50). Using the extent of variance components (especially accessions-by-cultivation system interaction) and correlations among cultivation systems, the necessity for specific selection in mixtures could be determined.

Results and discussion

Variance components estimated from data of the main experiment revealed low to very low accession by cultivation system interaction variance for vigor traits (see Table 2 below). Among the disease traits,

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occurrence of sclerotinia showed low genetic control, whereas downy mildew showed strong genetic control and low accession by cultivation system interaction variance. This indicates that accessions grow (or have been rated) similarly among the cultivation systems for plant vigor. Especially for the first cut of the 2018 season, when forage grasses among the alfalfa plants were growing tall and forming tillers, a large accession by cultivation system interaction variance (Vacc-cs) was observed for habitus (0.30 relative to Verror). This value was highest among all traits assessed and indicates a different rating/growth of alfalfa plants among cultivation systems.

Table 2. Estimates of variance components due to accession (Vacc), accession-by-cultivation system interaction (Vacc-cs) and residual error (Verror). Numbers in brackets give the extent of a variance component relative to the error variance.

	Variance components					
Trait[1]	Vacc		Vacc-cs		Verror	
			Vigor traits			
vigor_2017_1	0.83	(0.46)	0.09	(0.05)	1.79	(1.00)
vigor_2018_1	0.59	(0.18)	0.06	(0.02)	3.24	(1.00)
vigor_2018_2	0.72	(0.28)	0.01	(0.00)	2.59	(1.00)
vigor_2018_3	0.62	(0.25)	0.04	(0.02)	2.49	(1.00)
vigor_2018_4	0.69	(0.30)	0.06	(0.03)	2.28	(1.00)
vigor_2019_1	0.63	(0.24)	0.07	(0.03)	2.60	(1.00)
			Disease occurrence traits			
sclerotinia_2018_1a	0.005	(0.00)	0.089	(0.04)	2.23	(1.00)
sclerotinia_2018_1b	0.018	(0.01)	0.092	(0.03)	2.86	(1.00)
sclerotinia_2019_1	0.000	(0.00)	0.006	(0.02)	0.32	(1.00)
peronospora_2018_1	0.605	(0.24)	0.000	(0.00)	2.51	(1.00)
			Morphological traits			
habitus_2018_1	0.39	(0.42)	0.28	(0.30)	0.95	(1.00)
habitus_2018_2	0.29	(0.31)	0.09	(0.10)	0.95	(1.00)
stem-diametr_2018_1	0.45	(0.19)	0.02	(0.01)	2.33	(1.00)
leaf-stem-ratio_2018_1	0.11	(0.10)	0.00	(0.00)	1.19	(1.00)
leaf-stem-ratio_2018_2	0.16	(0.11)	0.01	(0.01)	1.50	(1.00)

[1] First and second number after trait name indicate the year and growth at which data was recorded, respectively (i.e., vigor_2018_1 means vigor assessed in 2018, first cut). If a trait was assessed two times on the same growth, this is indicated by a letter following the trait (i.e., sclerotinia_2018_1a, sclerotinia_2018_1b).

The patterns observed in the variance components were also reflected in the correlations among cultivation systems. Among the vigour traits that showed rather small accession-by-cultivation system interactions, strong correlations were observed, with r_p ranging between 0.71 and 0.90 (see Table

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below). Examining each time point when vigour was assessed separately, weakest correlations were observed for the first cut of the second year (vigor_2018_1) with r_p ranging between 0.74 and 0.76. During this time, the companion grasses are forming tillers and (especially forage grasses) are growing strongest. Already in the subsequent cut (vigor_2018_2), when grasses do not form tillers and do not interfere with growth of alfalfa plants that much anymore, r_p increased to 0.83 to 0.86, indicating a higher congruency among cultivation systems. The small genetic effects observed for occurrence of sclerotinia crown and root rot are reflected in the weak to very weak correlation among cultivation systems for this trait. On the contrary, the strong genetic control but rather low interaction between accession and cultivation system for occurrence of downy mildew led to strong correlations among all cultivation systems (r_p from 0.82 to 0.86). This indicates a congruent assessment of downy mildew occurrence independent of the cultivation system. The large accession-by-cultivation system interaction observed for plant habitus in the first cut (habitus_2018_1) is reflected by the only moderate correlation between the FORA and CONV cultivation systems (r_p = 0.57). This means that plant habitus of alfalfa plants was strongly influenced by tall growing grasses in the FORA cultivation system, leading to a different ranking compared to CONV.

Table 3. Correlations among the three cultivation systems for different traits assessed on spaced alfalfa plants. Correlation coefficients are based on accession means per cultivation system (n = 50). For vigour traits, numbers in brackets give the correlation coefficients based on the 15 accessions that showed best performance on average over all vigour traits.

	r_p		
Trait[1]	**FORA-LAWN**	**FORA-CONV**	**LAWN-CONV**
	Vigor traits		
vigor_2017_1	0.90** (0.83**)	0.73** (0.44)	0.74** (0.47)
vigor_2018_1	0.76** (-0.09)	0.74** (0.01)	0.75** (0.55*)
vigor_2018_2	0.83** (0.23)	0.85** (0.52*)	0.86** (0.56*)
vigor_2018_3	0.82** (0.29)	0.78** (0.06)	0.80** (0.74**)
vigor_2018_4	0.84** (0.06)	0.79** (-0.33)	0.79** (-0.05)
vigor_2019_1	0.85** (0.24)	0.74** (0.05)	0.71** (-0.23)
Mean vigor	0.83** (0.26)	0.77** (0.13)	0.78** (0.34)
	Disease occurrence traits		
sclerotinia_2018_1a	0.16	-0.05	0.06
sclerotinia_2018_1b	0.30*	-0.02	0.17
sclerotinia_2019_1a	-0.02	-0.07	-0.20
peronospora_2018_1	0.86**	0.82**	0.86**
	Morphological traits		
habitus_2018_1	0.73**	0.57**	0.86**
habitus_2018_2	0.77**	0.79**	0.78**
stem-diameter_2018_1	0.77**	0.72**	0.78**
leaf-stem-ratio_2018_1	0.53**	0.80**	0.49**

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leaf-stem-ratio_2018_2	0.71**	0.62**	0.44**

In the main experiment, generally positive correlations were observed among cultivation systems, what might lead to the conclusion that direct selection in the mixture is not necessary. However, we also demonstrated that when working within a set of elite germplasm with reduced variation (only 15 best accessions), genotype-by-cultivation system interactions might play a more important role and selection decisions, therefore, be different among cultivation systems again.

Outcomes and conclusions

Growing forage legumes and grasses in mixture have proven positive effects of productivity already for long time. The necessity for selection in mixture and an appropriate system here for yet must be determined. Therefore, we compared the three cultivation systems of a spaced plant nursery for selection of alfalfa as described above (CONV, FORA, LAWN) in a main experiment. Following conclusions can be drawn from the results:

- We found that selection decisions for alfalfa in a nursery with undersown grasses to alfalfa in a nursery with bare soil differed depending on the trait. Interestingly differences were rather small for vigor related traits, but larger for growth type.
- No clear recommendations can be given from the nursery trial as correlations among the compared systems were rather high (low genotype-by-cultivation system interaction) in the complete set of accessions. However, we found indications that when working within a set of elite germplasm with reduced variation, selection decisions might again depend on the cultivation system (different in mixture to non-mixture) and direct selection in the mixture would be more straightforward.
- Using a nursery system with undersown lawn type grasses (LAWN) might be a good compromise: it is predictive for the system with forage type grasses (FORA) but offers easier phenotyping compared to FORA and easier trial handling (reduced weeding efforts) in comparison to the bare soil system (CONV).

How resilience was improved

No improvement of resilience of the system after current experimental activities.

How information was transferred to the other situations

- Our lessons learned from the alfalfa-grass system are basically transferable to any combination of forage legume <-> forage grass. However, still many open questions regarding an ideotype of alfalfa as forage legume for use in mixture remain.
- 9 experimental selections have been produced that are now started to be compared in plot trials to test if selection pressure was different (measurement of realized selection gain).

Publications

- Poster "Selecting Forage Legumes for Use in Mixture with Grasses" at EUCARPIA-LIVESEED Conference March 2021

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- Scientific publication “Breeding alfalfa (*Medicago sativa* L.) in mixture with grasses” based on data from the main experiment planned for June 2021 after acceptance by the LIVESEED executive board.

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

Additional results from NARDI Fundulea (Trial #23b)

2020 – the alfalfa *(Medicago sativa L.)* synthetics, clover *(Trifolium pratense L.)* and them mixture with perrenial grass species - *Lollium hybridum, Dactylis glomerata, Festuca arundinacea and Trifollium alexandrinum* after 4 anual preceding crops - *Soybean, Maize, Sunflower and Winter wheat.*

The surface cultivated in ecological system in Romania with alfalfa (Medicago sativa L.) - 11810 ha/2017, 19613 ha/2018, 21681/2019 and with mixtures of forage plants – 4597 ha/2017, 5758 ha/2018, 7720 ha/2019 is constantly growing. Given this expansion of the cultivation area of alfalfa (Medicago sativa L.) and of forage mixtures as a result of, mainly, significant financial support of these, it is somewhat natural that the area cultivated with red clover (Trifolium pratense L.) in ecological system be smaller and variable over time – 2207ha/2017, 2605 ha/2018, 2306/2019, although the red clover is another precious perennial legume, mainly in the hilly and montain areas where alfalafa is less favourable. Therefore, we wanted to evaluate the effect of the alfalfa (Medicago sativa L.) and red clover (Trifollium pratense L.) that are grown in monoculture versus mixtures with diferrrent perrenial grass species in competition with different technological, biological and climate factors.

Results and discussion

1. **Main climate characteristics in Fundulea area (2019 – 2020)**

The vegetation season, September 2019 - August 2020, was, as in each year for the last decade, atypical under one aspect or another. This season was drier and generally warmer than long-term average for each month. Even if October and November 2019 were only 5, respective, 9 mm less in precipitation than long-term average (LTA), the 42 mm deficit from September, already put soil moisture content on an unfavourable trend for crops. Also, the rain deficit for December 2019 - April 2020 exceeded 114 mm and had a stronger negative influence on all crops. Even if, the rains from May-June 2020 (summing up 112 mm together) alleviated temporary the water stress, it is increased again in July and August 2020 when precipitation was 34 and respectively 5 mm, with 37 and respectively 44 mm less than LTA. The months November 2019 -March 2020 were (each) with more than +3°C warmer than LTA. Only May 2020 was as warmer as the long-term monthly averages (LTA), all other months exceeded the LTA. That warmer weather combined with dry winds (especially in April and August 2020) increased very much the potential evapotranspiration and created several periods with heat stress conditions in addition to the water stress.

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Figure 1. Monthly average air temperature Sept 2019 - August 2020

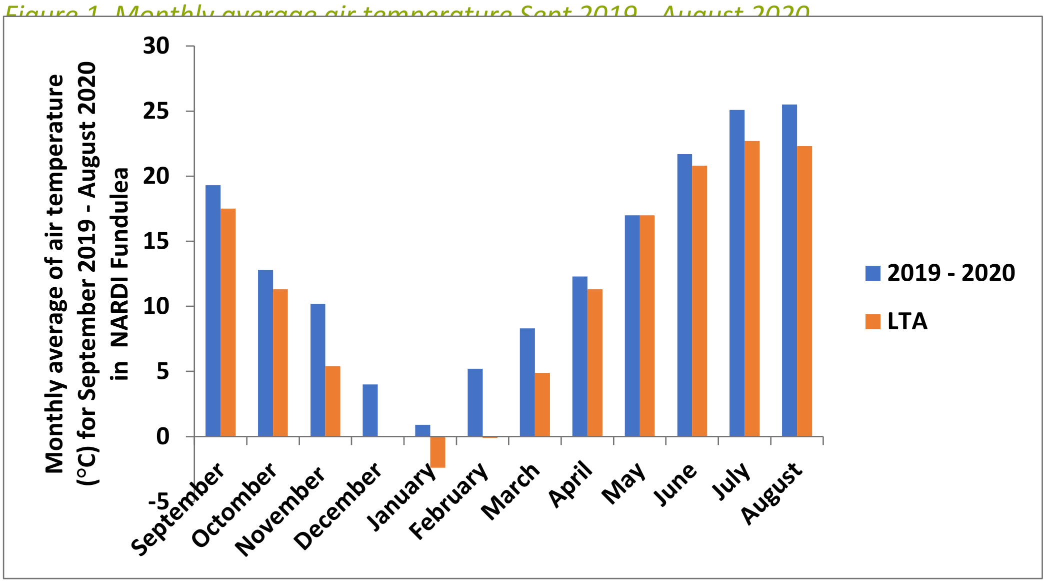
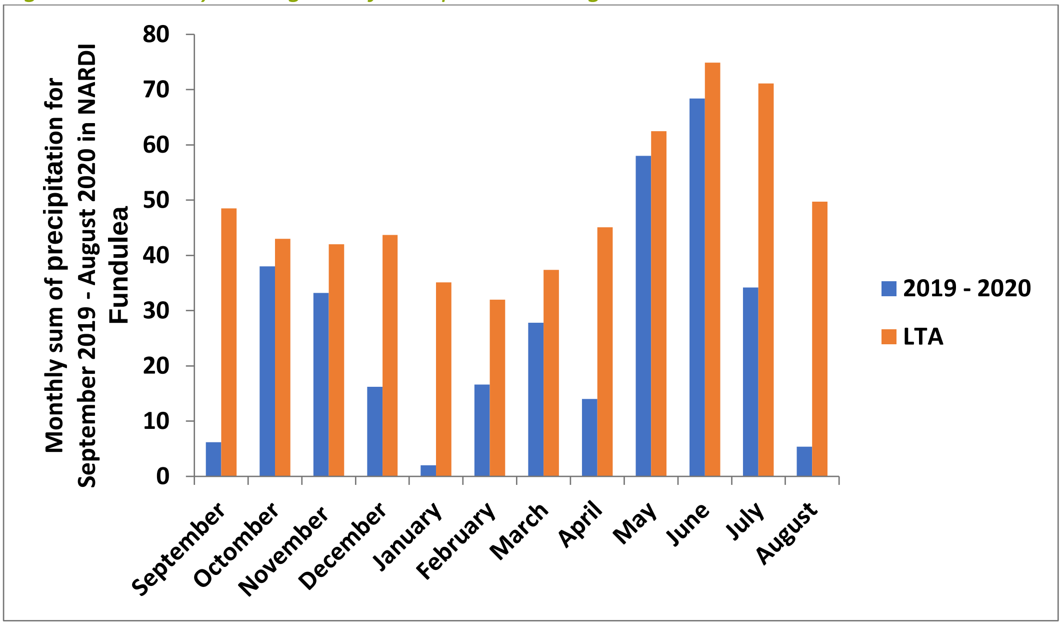


Figure 2. Monthly average rainfall Sept 2019 - August 2020



2. **Seed age and vigour of 19 alfalfa (Medicago sativa L.) synthetics and of the red clover (Trifolium pratense L.)**

Conclusion: The seed vigour of alfalfa (Medicago sativa L.) decreasing significant according to its age, but in case of two synthetic varieties *(Syn 0007 – 01 and Syn 0015 – 04)*, the seed vigour was about 83% although the seeds age is 15 - 18 years old (Figure 3).

Figure 3. Results on seed age and vigour of 19 alfalfa (Medicago sativa L.) synthetics and of the red clover (Trifolium pratense L.)

Variety		Seed age (years)	Seed germination (%)		Variety		Seed age (years)	Seed germination (%)	
No.	Name		First (EG)	Final (FG)	No.	Name		First (EG) *(4 days*	Final (FG) *(7 days)*

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			(4 days)	(7 days)					
1	Syn 0017 – 08	10,58	73	88	11	Syn 0006 – 08	10,58	55	58
2	Syn 0004 – 09	9,58	77	85	12	Syn 0012 – 05	13,58	51	57
3	Syn 0007 – 01	17,58	83	85	13	Syn 0035 – 03	15,58	43	49
4	Syn 0015 – 04	14,58	82	82	14	Syn 0065 – 94	24,58	35	44
5	Syn 0011 – 09	9,58	74	80	15	Syn 0815 – 96	22,58	36	42
6	Syn 0005 – 09	9,58	75	77	16	Red clover	2	87	87
7	Syn 0016-01-07 Comp.	11,58	58	75	17	Syn 0011 – 01 + Syn 0006 – 01 + Syn 0006 – 00 + Syn 0008 – 09;	15,83	29,25 *(31+29+29 +28)/4*	33,35 *(36+35+32+31)/4*
8	Syn 0010 – 08	10,58	64	73	18	Syn 0815 – 96 + Syn 0065 – 94 + Syn 0035 - 03	20,85	38 *(36+35+43)/3*	45 *(42+44+49)/3*
9	Syn 0016-02-07 Comp.	11,58	60	68	19	Syn 0016 – 07 + Syn 0016-02-07 Comp.	11,58	60,5 *(61+60)/2*	66,5 *(65+68)/2*
10	Syn 0016 – 07	11,58	61	65	20	Syn 0011 – 09 + Syn 0015 – 04	12,08	78 *(74 + 82)/2*	81 (80+82)/2
Correlation” Seed *age. x EG.”*: - 0,63394**					Correlation ”Seed *age x FG.”*: - 0,61916822**				

3. **Seed vigour and plant density of alfalfa synthetics at emergence, depending on preceding crops**

Figure 4. Correlation between seed vigour and plant density of alfalfa synthetics at emergence, depending on preceding crops (Fundulea, 2019).

Preceding crop	Seed vigour of alfalfa synthetics	
	Germination after 4 days *(EG)*	Germination after 7 days *(FG)*
Soybeen *(Glycine max L. Merr)*	0,654538084***	0,61928459**
Maize *(Zea mais L.)*	0,7132175***	0,652362276***
Sunflower *(Helianthus annuus)*	0,692210486***	0,630503659**
Winter wheat *(Triticum aestivum L.)*	0,7999908***	0,760399444***

Conclusion: Plant density of alfalfa *(Medicago sativa L.)* at emergence depend, exclusively, on seed vigour.

4. **The emergence density of synthetic varieties of alfalfa *(Medicago sativa L.)* and red clover *(Trifolium pratense L.)***

Conclusion: The plants density of alfalfa *(Medicago sativa L.)* at emergence depends on both the vigour of the seed and of preceding plant, but the red clover *(Trifolium pratense L.)* emergence

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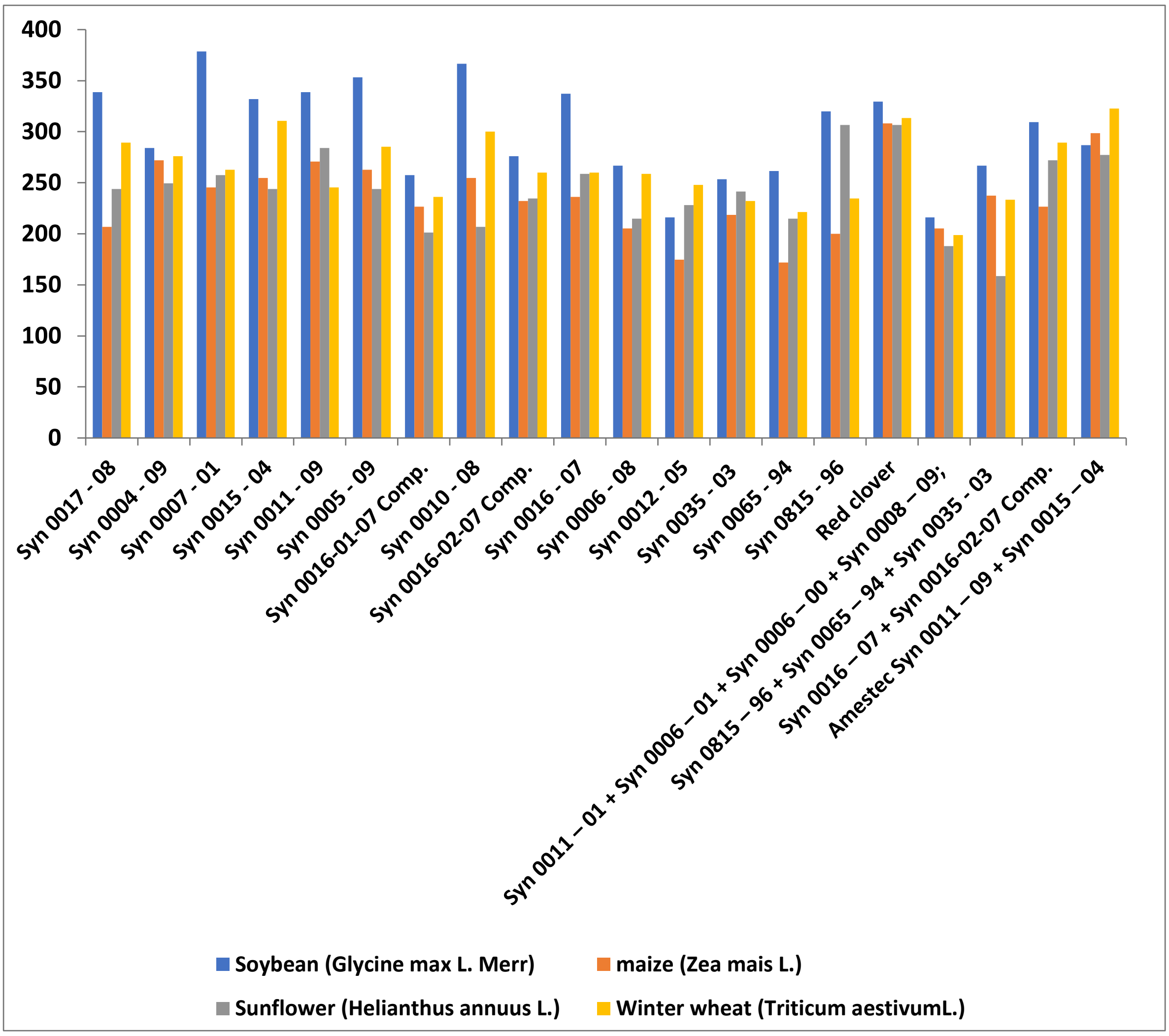
depends only on the seed vigour. The best preceding crop for alfalafa *(Medicago sativa L.)* synthetics is soybean for 17 synthetics and winter wheat for two synthetics (Figure 5).

Figure 5. The emergence density of synthetic varieties of alfalfa (Medicago sativa L.) and red clover (Trifolium pratense L.) grown organically, depending on the preceding plant (Fundulea, 07 - 10.06.2019)



5. Degree of weeding of the alfalfa synthetic varieties and red clover in ecological system

Conclusion: Weeding of alfalfa (Medicago sativa L.), red clover (Trifolium pratense L.) and of the mixture of forage plants is strong, especially in the first year of cultivation after wheat (Triticum aestivum L.), soybeans (Glycine max L. Merr) and corn (Zea mais L.). In the second year, the weeding of alfalfa (Medicago sativa L.) and fodder plant mixtures decreases very significantly, regardless of the preceding plant. This rule is also confirmed in the case of red clover (Trifolium pratense L.), especially in 2020 when the clover plots (Trifolium pratense L.) were, because of severe drought, very clean of weeds, regardless of the preceding plant (Figure 6).

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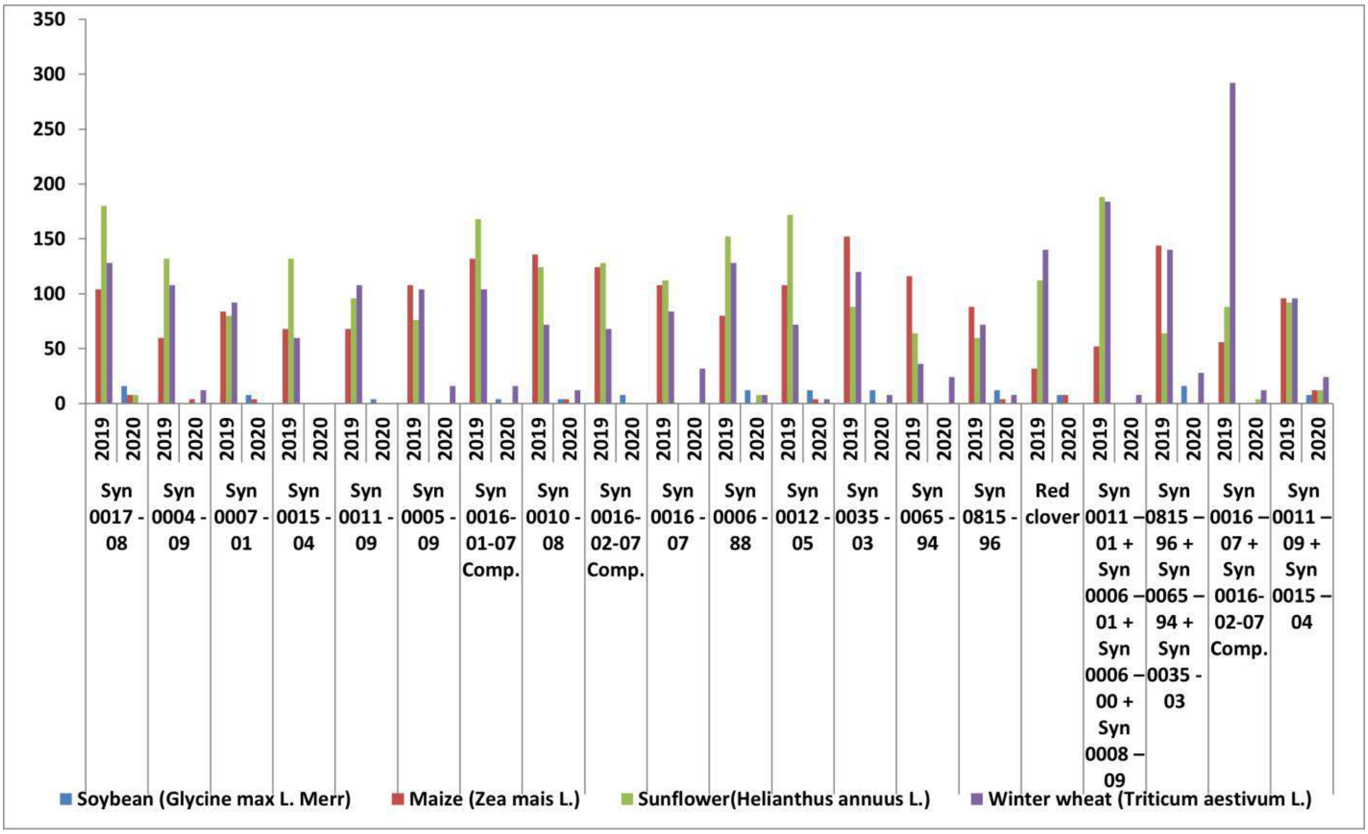
Figure 6. Degree of weeding (weeds no. / sqm) of the alfalfa (Medicago sativa L) synthetic varieties and red clover (Trifolium pratense L.) in ecological system, depending on the year of cultivation and the preceding plant (Fundulea 2019 and 2020)



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6. **Hay production of alfalfa and red clover grown organically, depending on the preceding plant and the cultivated variety of them**

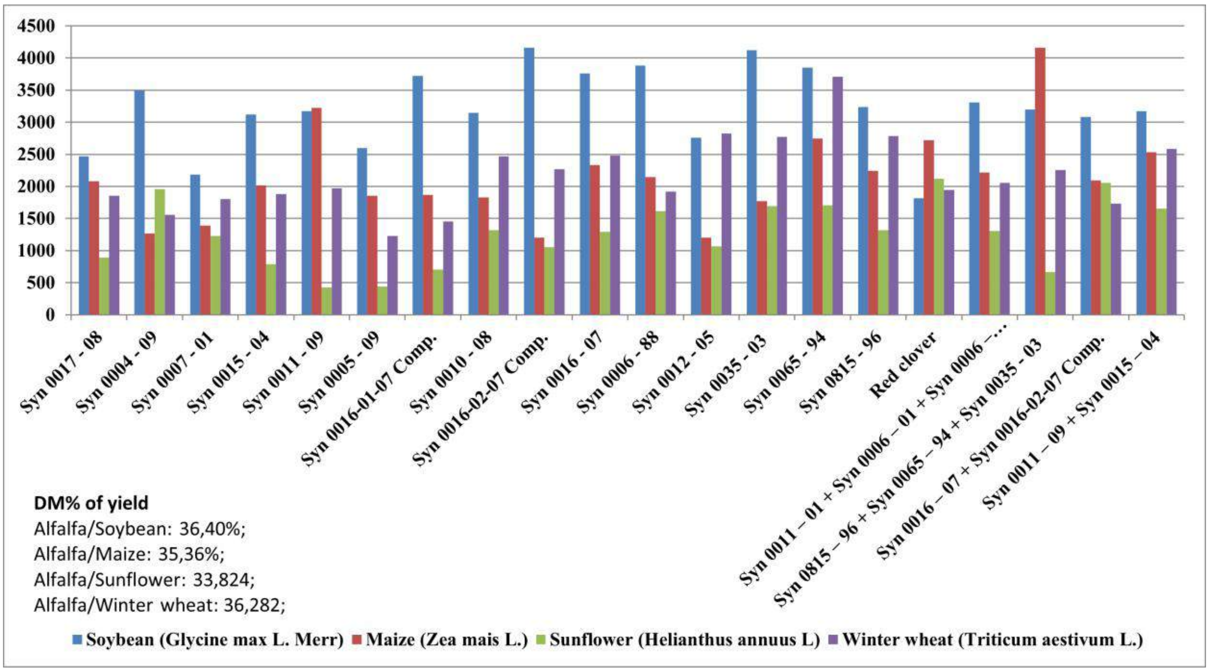

Figure 7. Hay production (kg / ha D.M) of alfalfa (Medicago sativa L.) and red clover (Trifolium pratense L) grown organically, depending on the preceding plant and the cultivated variety of them (Fundulea, 07 - 13.07. 2020)

Conclusion: The production of alfalfa (Medicago sativa L.) and red clover (Trifolium pratense L.) hay depends on the preceding plant and the cultivated variety, the highest productions being obtained after soybeans (at 16 Synthetics) and corn (at 3 Synthetics and in red clover), and the smallest, after sunflower (in 18 Synthetics), corn (in 1 Synthetic) and soybeans (in red clover) (Figure 7 previous page).

7. **Production of seeds obtained from different synthetic varieties of alfalfa grown organically, depending on the preceding plant**

Conclusion: Seed production of organically grown varieties of alfalfa (Medicago sativa L.) depends, mainly, on the preceding plant, with the highest yields being obtained after soybeans (at 14 Synthetics) and wheat (at 5 Synthetics), and the smallest after sunflower (at 12 Synthetics) and after corn (at 7 Synthetics) (Figure 8.)

Figure 8. Production of seeds obtained from different synthetic varieties of alfalfa (Medicago sativa L.) grown organically, depending on the preceding plant (Fundulea, 2020)

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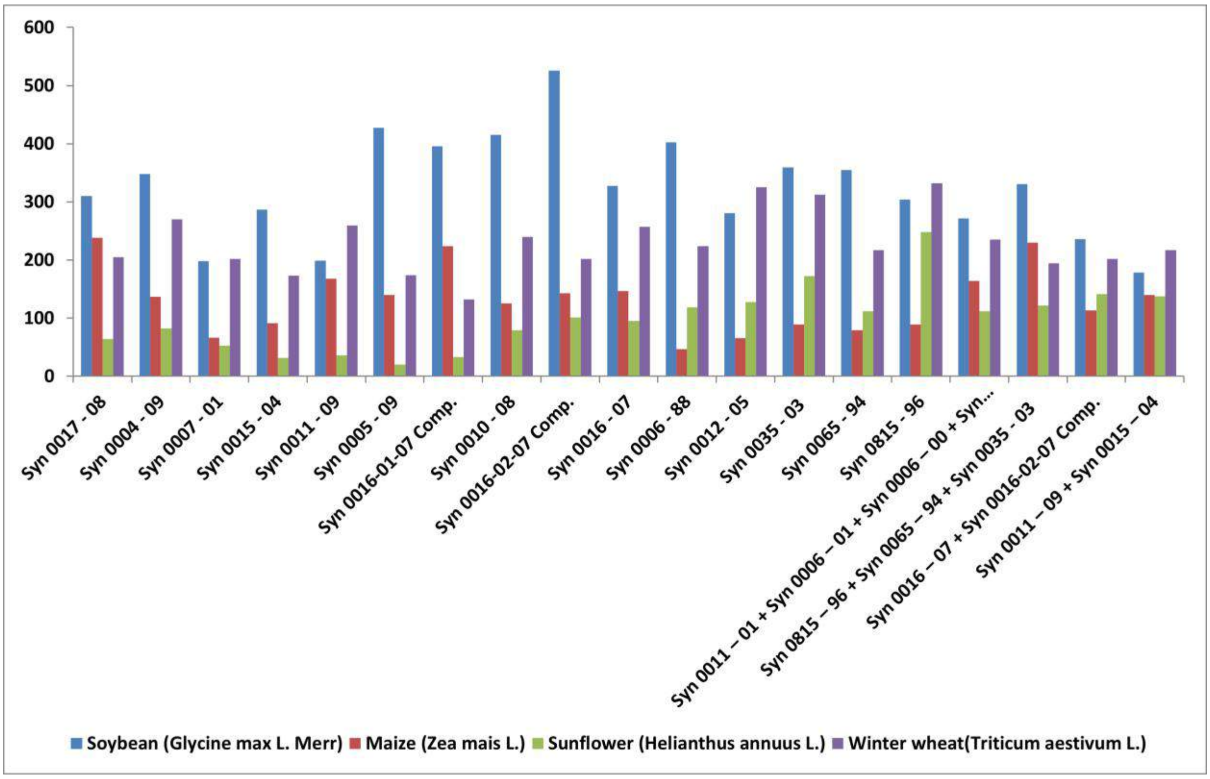

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IPC-Mixed Cropping – Maize-beans Intercropping trials 2019 (Trial #24)

Lead: IPC– Portugal
Pedro Mendes Moreira, Duarte Pintado, André Pereira – pmm@esac.pt

Abstract
Maize (*Zea mays* L.) and beans (*Phaseolus vulgaris* L.) intercropping is a traditional method of cultivation used in Portugal. This intercropping system has a renewed interest in organic agriculture and better binomial combinations are needed to provide to farmers towards reduction the use of fertilizers and erosion effect so as soil improvement.
The aim of this study intends to compare different beans populations per se or in intercropping with maize to access yield and LER.

Introduction
Intercropping is the cultivation of two or more crops at the same time on a certain location, it is still important in rural areas in the central and northern regions of Portugal, where combinations of a cereal with a grain legume are common. This method provides a better use of the land, due to nitrogen fixation, and provides also high protein food.
In SOLIBAM project trials allowed us to search the best pairs combination between maize and beans, plus beans trials information. In the past years ESAC-IPC has been collect and trial a large quantity of beans genotypes, trying to test their ability for intercropping (Ferreira, Pintado, & Mendes Moreira, 2018).
Our hypothesis search intends to obtain better combinations between and maize and beans and promote their coevolutionary process under intercropping, to increase yield and LER

Material and Methods
2019
Genetic Resources
The genotypes used in this trial were collected under FCT national project and are being studied in national and international projects until 2017 (e.g., SOLIBAM and DIVERSIFOOD projects) by ESAC-IPC.

Intercropping trial

Beans (n° accessions) name of the variety
1. 123-R-4135
2. 59-4071
3. 220-5248
4. 107-4199

Maize (n° accessions) name of the variety
1. Pg C0S0 18 (Maize Landrace)

2020

In 2020 an intercropping trial with 14 bean genotypes was planned and seeded, however, due to successive attacks by wild animals and birds it was not possible to withdraw any data. The remaining plants were harvested for seed to use in the next season trial.

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Location

The trials were managed at the organic field of IPC-ESAC in Bencanta, Coimbra Portugal (40.21709426119619; -8.44779968261719; 15 m) in 2019.

Experimental design

The trial was sowed on 15th May 2019 in a low input and organic agriculture Production Systems. No preceding crops neither fertilization since the last maize campaign were done.
The 4 beans genotypes were evaluated in a randomized complete block design at Coimbra (Figure 1), with a 60 000 stand, in plots of four lines with 2.4 length and 0,75 interrow distance.
The mechanical weed control occurred on 1st June 2019, and the manual weed control occurred during June and July. The harvest was done by hand on 16th September. Statistically analysis was done with IBM SPSS Statistics program and Excel.

Pg COSO 18											
123-R-4135	59-4071	220-5248	107-4199	123-R-4135	59-4071	107-4199	220-5248	220-5248	107-4199	123-R-4135	59-4071
Pg COSO 18											

Figure 1 - Trial design in a randomized complete block of 3 repetitions per population.

Statistical methods

ANOVA, descriptive statistic, and correlations analysis was done with IBM SPSS Statistics program and Excel.

Evaluation

The list of traits evaluated are presented below in Table 1-2.

Table 1. Crop development and agro-ecological performance traits

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Land Equivalent Ratio for Beans (LER Beans)	LER Beans = Yield Intercropping/Yield *per se*	%

Table 2. Crop productive performance traits

Crop productive performance (yield, yield components)		
Trait	**How it has been assessed**	**Type of data available**
Yield per se	Hand harvested, used grain yield content directly measured;	Quantitative (Mg/ha)
Yield Intercropping	Hand harvested, used grain yield content directly measured.	Quantitative (Mg/ha)

Results and discussion

No significant differences were registered for yield among the varieties tested in the two systems (Extreme or per se and Intercropping).

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The Yield trait (Figure 2), indicates that the values range from 1.301 Mg/ha (220-5248) to 1.87 Mg/ha (123-R-4135) in the system per se. In Intercropping system these values range from 1.241 Mg/ha and 1.671 Mg/ha (107-4199).

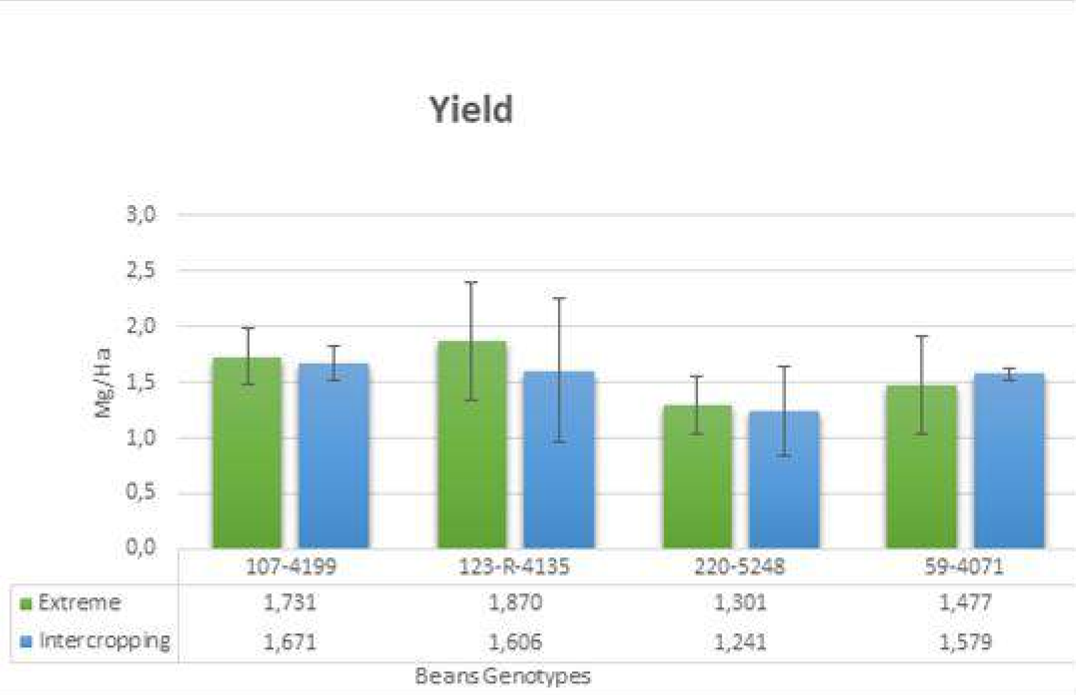

Figure 2. Land Equivalent Ratio (LER) beans: Yield Beans Intercropping/Yield Beans Extreme

The Land Equivalent Ratio (LER) Beans (Table 3) was higher than one only in the case of 59-4071.

Genotype	LER Beans	
	Mean	*SD*
123-R-4135	0,864744674 ±	0,232847402
59-4071	1,15245795 ±	0,429895648
220-5248	0,943075158 ±	0,155750669
107-4199	0,972628276 ±	0,094504896

Table 3. The Land Equivalent Ratio (LER) Beans.

No significant differences were observed for yield across systems and among genotypes, however some tendencies indicate that only '59-4071' showed more productivity in Intercropping system (1.579 Mg/ha) than per se (1.477 Mg/ha), for the other genotypes the adaptability was similar to both systems.

The results of LER, for beans, indicate a value near 1. This indicates that is probably needed to develop a co-breeding program for a more specific adaptation. The results obtained are probably the output of the farmers' modus operandi, i.e., farmers use to do both intercropping and per se systems according to their needs, but with the same seed. Conducting a multi-site trials in PPB can also provide us a more accurate comparison.

Outcomes and conclusions

The LER results indicates potential use of the analysed beans genotypes for co-breeding work initiatives in a PPB context.

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

Publications

Ferreira, E. A., Pintado, D., & Mendes Moreira, P. (2018). Maize and Beans Intercropping Under Participatory Plant Breeding. DIVERSIFOOD Final Congress: Cultivating diversity and food quality. Rennes, France.

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

INRAE - Agroforestry (Trial #25)

Lead: INRAE
Solene Lemiche and Dr. Veronique Chable, (INRAE)

Abstract

Understanding plants/microbiome interactions is a growing issue to locally adapt plants to the diversity of organic systems, while combining scientific methods and farmers' knowledge. On-farm plant breeding is a holistic approach in which plants are regarded as biological systems shaped by genetic, epigenetic, and microbiological processes. Understanding how on-farm breeding enhances these interactions could inspire organic plant breeding aiming resilient cropping systems (Duhamel & Vandenkoornhuyse, 2013).

In this study, we focused on fungal and bacterial communities of the tomato root endosphere. We sampled roots of two varieties in two farms located in the Cévennes region in South-East France, in 2019 and 2020. Both farms integrate market gardening in agroforestrial systems, with different pruning intensities of trees and display a shade less control plot as well. Among the different factors tested (tomato variety, year, site, agroforestrial modality), year and agroforestrial modality played a major role in the microbiome structuration, while variety seems to have a lower impact, despite their genetic distinction. The impact of agroforestry on tomato chemotype and yield when put in perspective of microbial results invites us to consider plant adaptation and resilience as dynamic relative concepts.

Introduction

Hypothesis and research question

Our study relies on two main hypotheses linked by a cause-and-effect relationship:
- on farm selection has an impact on the microbiome structuration
- microbiome supports plant adaptation through on-farm selection

The research question then is "are the differences in microbial communities' structuration linked to on-farm selection and do these differences support plant adaptation?".

Objectives

The first objective is to understand the main drivers of microbiome structuration of on-farm selected tomato in contrasted agro-forestrial conditions. Then, the aim is to get inspiration for organic selection to improve resilience with holistic breeding strategies relying on plant and microorganism's co-evolution.

Material and Methods

Genetic resources

In this study, we worked with populations of two tomato varieties: Rose de Berne (RDB) and Coeur de Boeuf (CDB). These populations are selected on-farm since 2016.

Creation of experimental population

(CCP, dynamic mixtures, mass selection and variations, OPV, natural selection, intercropping)

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Since 2018, tomatoes are selected and grown in the same agroforestrial modality at *les Terres du Roumassouze* farm. For both varieties, an initial population was sown in 2018 after 2 years of mass selection. Then, plants sampled in 2019 corresponded to the first generation of agroforestrial differentiated selection, and those sampled in 2020 to the second generation (Fig.1).

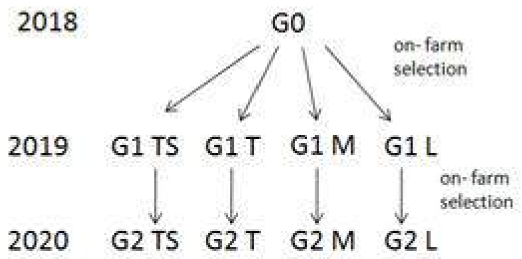

Figure 1. Selection scheme at les Terres du Roumassouze. For each variety, an initial lot (G0) is sown and selected in each agroforestrial modality (TS shade less control; T pollard; M medium pruning; L light pruning).

Experimental design

The main experimental, *les Terres du Roumassouze*, is in Vézénobres (44° 30' 11'' North, 4°08' 10'' East) in the Cévennes area in France. It consists of 11 hectares of organic agroforestrial market gardening, open field cultures and sylviculture. On the agroforestrial area, hybrid walnut (*Juglans nigra L.* x *Juglans regia L.*) planted in 1996 are interspaced with market gardening crops, with 10m space between each tree.

In 2015, 4 agroforestrial modalities are defined, corresponding to different pruning intensities: Light pruning (L), Medium pruning (M), Pollard (T) and Shade less control (TS). The volume (m^3) of wood removed from trees in each modality was 0.015 (L), 0.084 (M) and 0.3 (T). A second pruning was done in 2018 to maintain the desired shade intensities. Each modality corresponds to a 30*40m surface (1200 m^2), including cultivation beds (3 rows) and grass strips between trees. Cultivation beds are 25m long, 1m wide and separated by a 0.8m pathway. The different agroforestrial modalities are separated by a one meter large a grass strip. Cultural practices are similar between the different agroforestrial modalities. Only plants in the central row are sampled to ensure a homogenous relative distance to the trees. Different analyses are conducted on this site to picture a global understanding: genetic characterisation, chemotype analysis, yield and phenology monitoring, and microbiome reconstruction.

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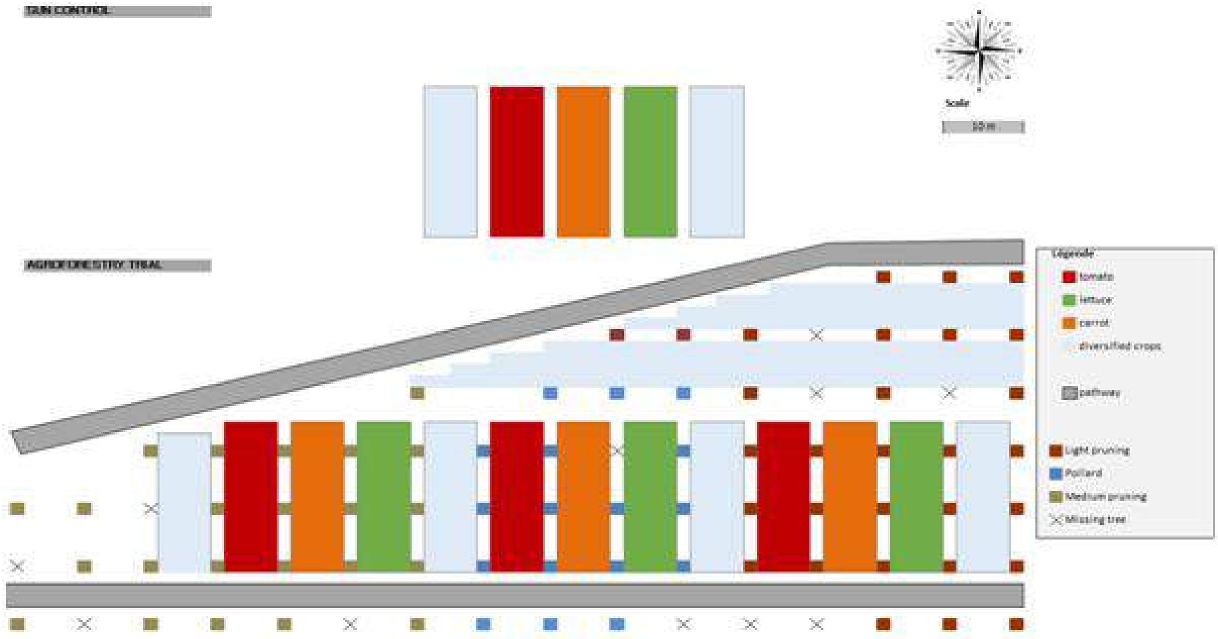

Figure 2. Experimental site at les Terres de Roumassouze

A second site located in Cazilhac, *la Ferme du Boulidou*, also in the Cévennes region is taken as a comparison point for microbial communities only. On this farm, diversified fruit trees grow in a permaculture design with market garden crops. The variety Rose de Berne is also cultivated there and selected on-farm. A control plot without trees is also established.

Evaluation

1. Genetic analysis

Molecular marking of 12 plants per combination of variety and modality was performed by VEGENOV. It targeted 8 SSR markers usually used in varietal differentiation. This genetical analysis was performed on plants of the second selection generation in 2020.

2. Chemotype analysis (INRAE PSH)

In 2019, analysis regarding physical and biological quality of fruits was conducted by INRAE UMR PSH. Harvesting was spread over 3 dates during the production period due to the heterogeneity of fruit ripening. Three fruits were harvested per individual. Each fruit was weighed and tested for firmness with the Durofel. External colouration was measured with a colorimeter (CR-5, Konica Minolta) in the CIELAB colour space on two opposite sides of the fruit. The 3 parameters measured are:

- L: lightness, which is derived from the luminance of the surface and takes values between 0 (black) and 100 (reference white).
- a: axis from green (-60) to red (+60)
- b: axis from blue (-60) to yellow (+60)

The fruits are then cut on the equatorial plane. The upper part (columella pedicel area) is divided into 4 equal quarters, the placenta and seeds are removed to keep only the pericarp. Two opposite quarters are used for measurements of percentage dry matter and refractometric index (Brix). The pericarp of the lower part is stored at -80°C, crushed in liquid nitrogen and freeze-dried before the biochemical determination of sugars (glucose, fructose, sucrose), acids (citric and malic), vitamin C and major phenolic compounds.

3. Yield and production monitoring

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140 tomato plants were monitored (over 260) for the experiment in each agroforestry treatment and control (70 of "Coeur de boeuf" (CDB) variety and 70 of "Rose de Berne" (RDB) variety). Production was monitored by AGROOF from the plantation (May) to the end of the crops (October), from 2015 to 2020. In 2020, light pruning modality was not monitored. For each tomato plant, fruits were counted, weighed, and then classified according to their physiological disorders within 4 categories related to the commercial value of tomato according to the European regulation N°543/2011 on fruits and vegetables:
Extra: high quality (no defects),
Class I: good quality (slight defects without market devaluation),
Class II: marketable quality (defects with market devaluation),
Waste: not marketable.

4. Microbiome characterisation

For each variety*agroforestrial modality combination, the roots of 12 plants were sampled, for a total of 96 samples. The fine roots (<1mm) were taken at a depth of 10 to 20cm in the immediate vicinity of the plant studied, and the weight of a sample was calibrated between 80 and 100 mg. The samples were rinsed with a 5/1000 Triton solution and then with sterile water before being dried and stored at -80°C before DNA extraction. The extraction was performed by the GENTYANE platform (UMR GDEC, INRAE) using a Sbeadex™ kit (LGC) automated on an oKtopur™ machine (LGC). The DNA assay was performed with the Hoechst 33258 dye and an Infinite® M1000 automaton (TECAN). Extracted DNA was stored at -20°C before being standardised to a concentration of 10 ng/µl. A PCR (illustra™ puReTaq Ready-To-Go™ PCR Beads, GE healthcare®) was performed with the fungal primer pair NS22B (5'-AATTAAGCAGACAAATCACT-3') and SSU0817 (5'-TTAGCATGGAATAATRRAATAGGA-3') targeting a region of approximately 550 bp of the 18S rRNA (Borneman & Hartin, 2000; Lê Van et al., 2017). The amplification protocol included 35 cycles of denaturation (95°C for 30s), hybridisation (54°C for 30s) and elongation (72°C for 1min), with an initial denaturation (95°C for 4min) and a final elongation (72°C for 7min). The 16S rRNA gene was amplified using bacterial primers 799F (5'-AACMGGATTAGATACCCKG-3') and 1223R (5'-CCATTGTAGTACGTGTGTA-3'). The conditions for this PCR consisted of an initial denaturation step at 94 °C for 4 min followed by 32 cycles of 94 °C for 30 s, 54 °C for 30 s, and 72 °C for 1 min with a final extension step at 72 °C for 10 min. The products of these first PCR were then purified (Agencourt® AMpureXP Magnetic Beads) with a robotic pipettor (Bravo-Agilent®), quantified by fluorimetry (Quant-iT PicoGreen™ dsDNA Assay Kit) and standardised to the same concentration (0.5 ng/µL) for the preparation of the sequencing library. A second PCR reaction was performed on a Smartchip instrument (Takara) to allow individual labelling of the samples (Illumina multiplexing) and the final production of the sequencing library. This library was then purified (AMpureXP Agencourt® magnetic beads) and quantified (Kapa Library Quantification Kit-Illumina®) before being sequenced with a MiSeq Illumina® instrument (Paired-end 2x300 cycles kit). The production of the libraries and the sequencing were carried out by the Environmental and Human Genomics platform (GEH, Rennes, France).

The sequencing data were processed with the FROGS (Find, Rapidly, OTU, with Galaxy Solution) pipeline (Escudié et al., 2017). The clustering method used was SWARM (Mahé et al., 2014), which generates sequence clusters (~OTUs) but with the advantage of not using an arbitrary identity threshold (which produces OTUs). To avoid the creation of artificial sequence clusters, only those containing sequences from at least 3 independent samples were kept. The fungal sequence database PHYMYCO-DB (Mahé et al., 2012) was used for BLAST taxonomic affiliation of sequence clusters in FROGS. The one used for the affiliation of bacterial sequences was SYLVA 16S. These clusters were then filtered according to the quality of the affiliation with a threshold of 95% in coverage and 95% in identity. The data were organised in a contingency table and the number of sequences per sample

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was normalised using the 'vegan' package in R. This number of sequences allowed to accurately describe the composition of the root microbiome, verified by rarefaction curves for each sample with the 'vegan' module in R. These normalised data were used for statistical analysis. The assembly of the root microbiota was studied at the community and main phyla levels.

Statistical methods

An ANOVA was performed on the genetic data to assess the genetic structure at the level of varieties and agroforestrial modalities. Then, to test the effect of tomato variety and pruning intensity on the physical and biochemical quality of the fruit, a first linear model was constructed with the variety and pruning modality as fixed factors, and the individuals were included as a random effect. For each factor, a PCA is used to visualise the distribution of individuals according to their modality. The variables for which the interaction of the factors is significant are not considered in order not to bias the analysis, as well as those for which the tested factor has no effect. A second linear model to test the effect of variety and pruning intensity on microbiota structuring (Shannon index, Simpson index and equitability) is constructed in a similar way. As they were two sampling campaigns, year is also integrated as factor. For this model, the sampling design. For this model, the sampling design allows the sub-plot to be integrated as a random effect paired to the individuals. Finally, a Poisson model is used to test the effect of the variety, pruning modality and year on the number of microorganism species (species richness) (Tab. 2). For each model, outliers are removed and when necessary, and variables are log-transformed to ensure the normality of the residuals. The significance of each factor in the model is determined by an ANOVA. Interactions between factors are not considered. To represent the composition of the microbiota according to the variety or pruning modality effect, a PLS-DA regression on the abundance (number of sequences) of each cluster per individual is performed. Models are written in table 1. All the statistical analysis are performed with R.

Variables X	Model	Distribution	Model writing
Fruit quality	lme	Gaussian	lme(X ~ VARIETY*MODALITY, random = ~ 1 | INDIVIDUAL)
Diversity indexes	lme	Gaussian	lme(X ~ VARIETY*MODALITY*YEAR, random = ~ 1 | SUB-PLOT/INDIVIDUAL)
Specific richness	glm	Poisson	glm(X ~ VARIETY*MODALITY*YEAR, family="poisson")

Table 1.: Models used for statistical analyses

Participatory/multi-actor approach

This study was performed in collaboration with different research teams:

- INRAE UMR PSH for the bio-physical and chemotype analysis
- AGROOF for phenology and yield monitoring (not included in this report)
- CNRS UMR ECOBIO regarding molecular ecology and microbiome analysis

Plants were sampled on-farm in organic production conditions, and yearly workshops are organised at la *Ferme du Roumassouze.*

Results

On-farm selection maintains a genetic structure at the variety level but does not create structuration at the level of agroforestrial modalities.

The ANOVA results show a significant genetic structure (p= 9.999e-05) at the level of variety (Fig. 3). On the other hand, differentiated selection between agroforestrial modalities does not induces a new structuration for the tested markers (p=0.19) (Fig. 4).

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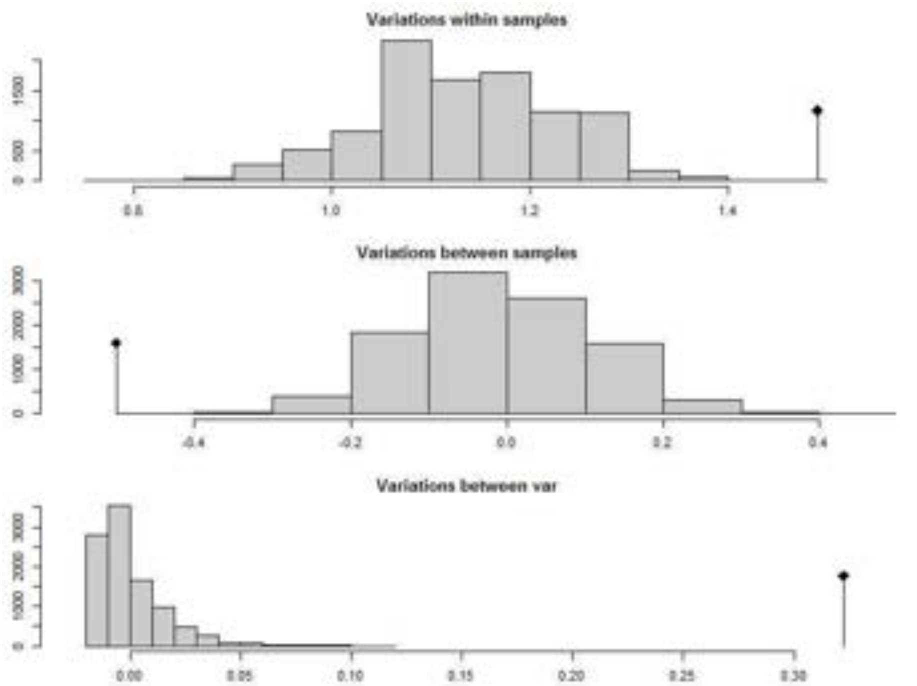

Figure 3. ANOVA results showing the variation within samples, between samples and between varieties for 8 SSR markers. Quasi-gaussian pattern reveals no structuration within and between samples, while it can be observed at the level of varieties. This variation is significant (p= 9.999e-05).

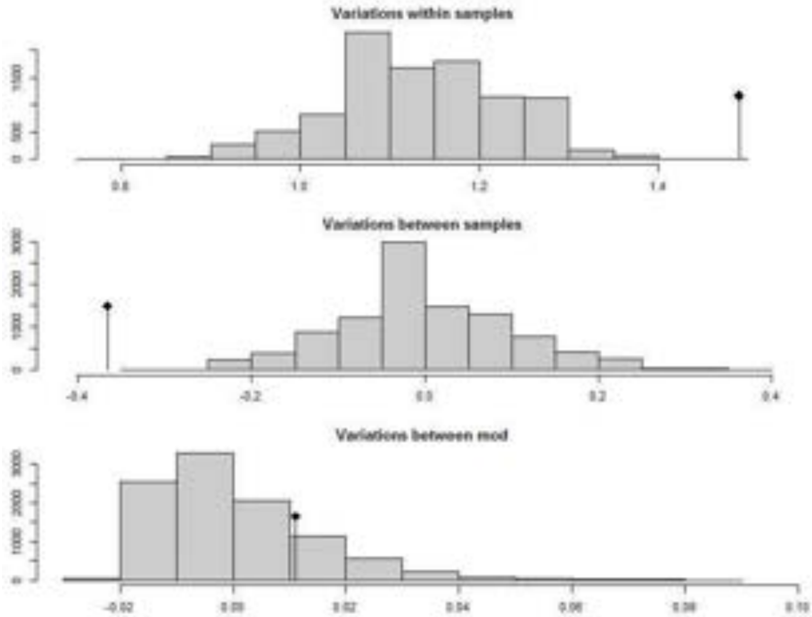

Figure 4: ANOVA results showing the variation within samples, between samples and between agroforestrial modalities for 8 SSR markers. No significant structuration is observed, even at the level of agroforestrial modalities (p=0.19).

The chemotype is impacted by both the variety and agroforestrial modality

ANOVA results show a significant effect of variety and pruning intensity for most of the variables tested. The interaction of these different factors is also significant for a limited number of variables (Table 2). For each factor, a PCA is used to visualise the distribution of individuals according to their modality. The PCA on the pruning factor highlights a differentiation of biochemical profile with a

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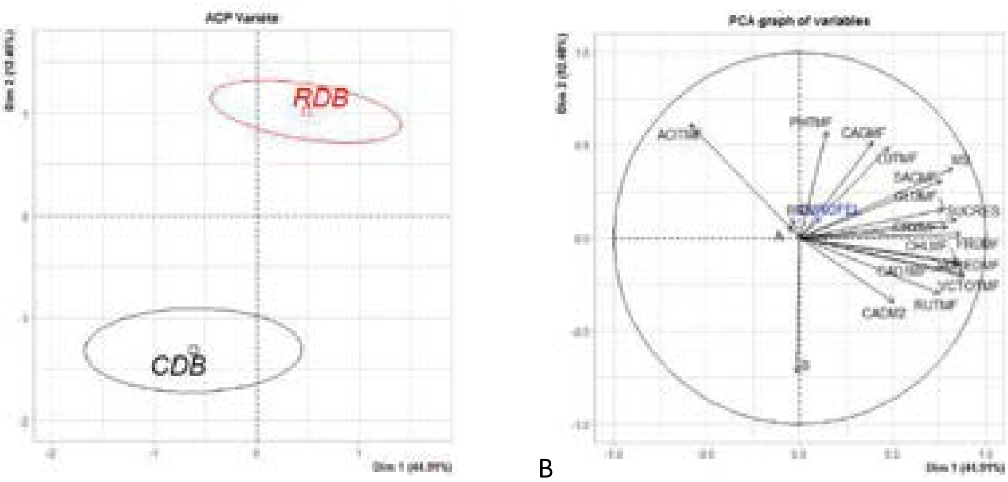
gradient according to the openness of the environment (Fig. 5). The PCA on the variety factor illustrates the divergence of biochemical profiles between the two populations (Fig. 6).

Variables	Variety		Modality		Varity*Modality	
	F-value	p-value	F-value	p-value	F-value	p-value
Dry matter	30,24	**3,814E-08**	52,22	**2,69E-11**	3,48	0,32
Weight	168,27	**< 2,2E-16**	32,89	**3,39E-07**	13,63	**3,45E-03**
Sugars	18,22	**1,97E-05**	118,49	**< 2,2E-16**	2,50	0,48
Glucose	18,06	**2,14E-05**	75,16	**3,35E-16**	3,30	0,35
Fructose	14,69	**1,27E-04**	154,89	**< 2,2E-16**	3,49	0,32
Saccharose	9,33	**2,25E-03**	40,08	**1,02E-08**	3,70	0,30
Acids	0,06	0,81	186,33	**< 2,2E-16**	9,18	**0,03**
Citric acid	1,07	0,30	159,25	**< 2,2E-16**	7,55	0,06
Malic acid	14,88	**1,14E-04**	144,85	**< 2,2E-16**	28,68	**2,62E-06**
Vitamine C (total)	32,53	**1,17E-08**	335,58	**< 2,2E-16**	2,10	0,55
Vitamine C (reduced)	31,49	**2,01E-08**	303,68	**< 2,2E-16**	3,93	0,27
Vitamine C (oxydated)	1,99	0,16	26,66	**6,95E-06**	7,84	**0,05**
Phenolic compounds	139,33	**< 2E-16**	216,41	**< 2,2E-16**	14,35	**2,47E-03**
Caffeic acid glucoside	65,97	**4,58E-16**	47,22	**3,12E-10**	3,41	0,33
Chlorogenic acid	28,70	**8,45E-08**	260,58	**< 2,2E-16**	2,18	0,54
Caffeic acid derivatives 1	4,61	**0,03**	95,29	**< 2,2E-16**	0,35	0,95
Caffeic acid derivatives 2	8,77	**3,07E-03**	22,05	**6,38E-05**	1,97	0,58
Rutin	5,13	**0,02**	125,13	**< 2,2E-16**	7,77	0,05
Naringenin chalcone	1848,93	**< 2,2E-16**	39,89	**1,12E-08**	18,74	**3,09E-04**
Cryptochlorogenic acid	19,94	**7,98E-06**	223,69	**< 2,2E-16**	1,85	0,60
Kaempferol rutinoside	13,05	**3,03E-04**	17,42	**5,79E-04**	18,15	**4,10E-04**
Brix	13,53	**2,35E-04**	11,08	**0,01**	1,01	0,80
L	18,23	**1,96E-05**	12,81	**0,01**	13,80	**3,20E-03**
A	5,07	**0,02**	4,62	0,20	4,84	0,18
B	554,16	**< 2,2E-16**	31,68	**6,11E-07**	5,15	0,16
Durofel	1,95	0,16	0,32	0,96	3,82	0,28
Carotenoids	8,79	**3,02E-03**	3,54	0,32	10,09	**0,02**
Lutein	31,01	**2,57E-08**	11,91	**7,70E-03**	5,28	0,15
Lycopene	3,59	0,06	3,49	0,32	11,80	**8,09E-03**
Beta-carotene	28,34	**1,02E-07**	4,42	0,22	7,94	**0,05**
Phytoene	28,30	**1,04E-07**	6,38	0,09	5,13	0,16

*Table 2.: ANOVA results showing the effect of tomato variety, agroforestrial modality and combination variety*agroforestry on physical and biochemical variables.*



A B

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Figure 5. PCA ordination of biochemical and physical variables depending on tomato variety (A) and graph of variables (B). CDB Coeur de Boeuf variety; RDB Rose de Berne variety.

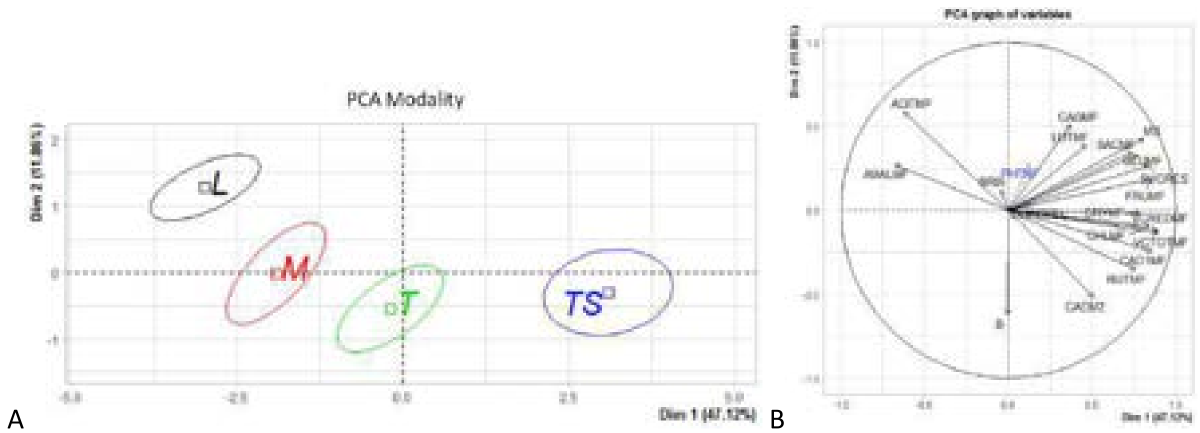
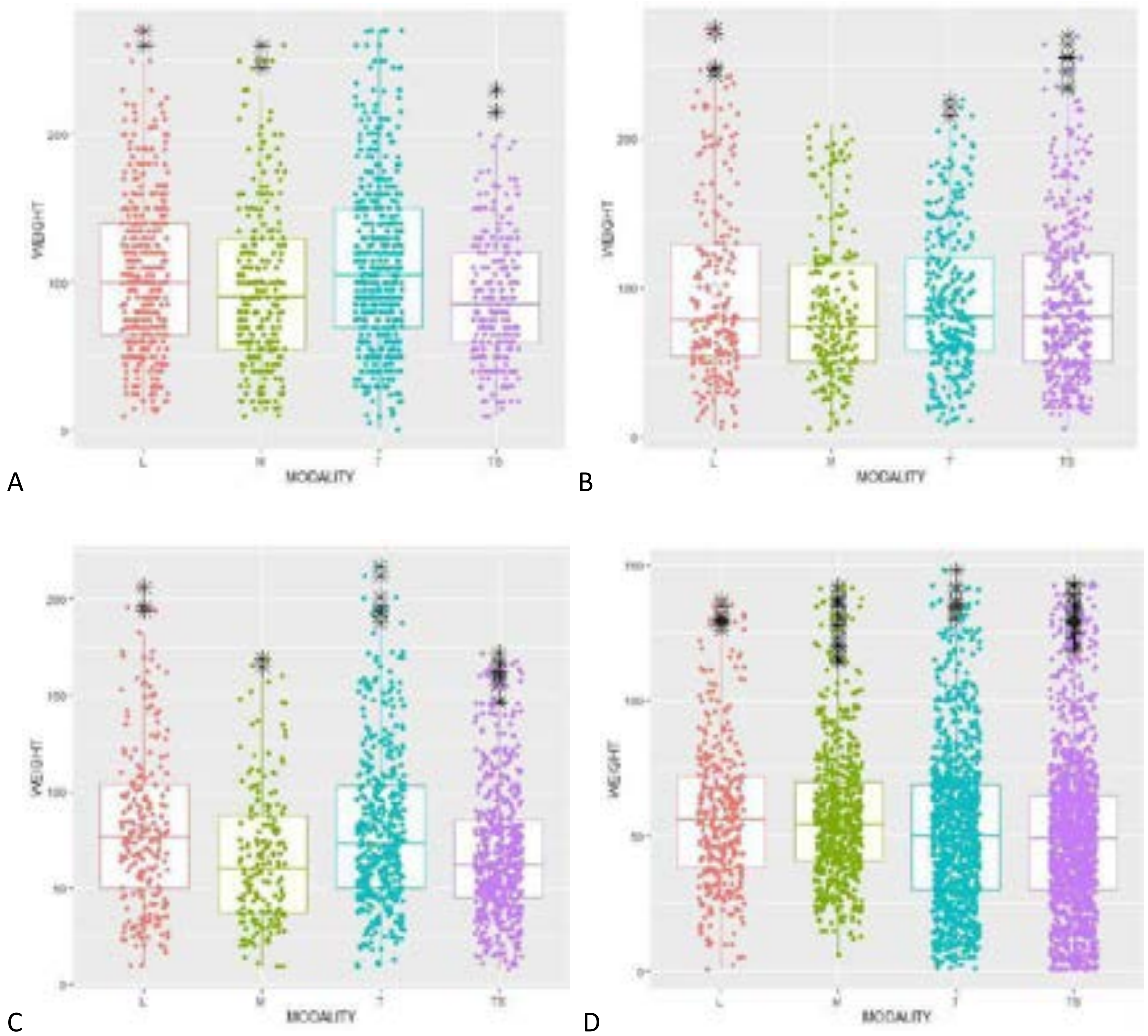


Figure 6. PCA ordination of biochemical and physical variables depending on agroforestrial modality (A) and graph of variables (B). L Light pruning; M medium pruning ; T pollard ; TS sun control.

Agroforestry impacts raw and marketable yield



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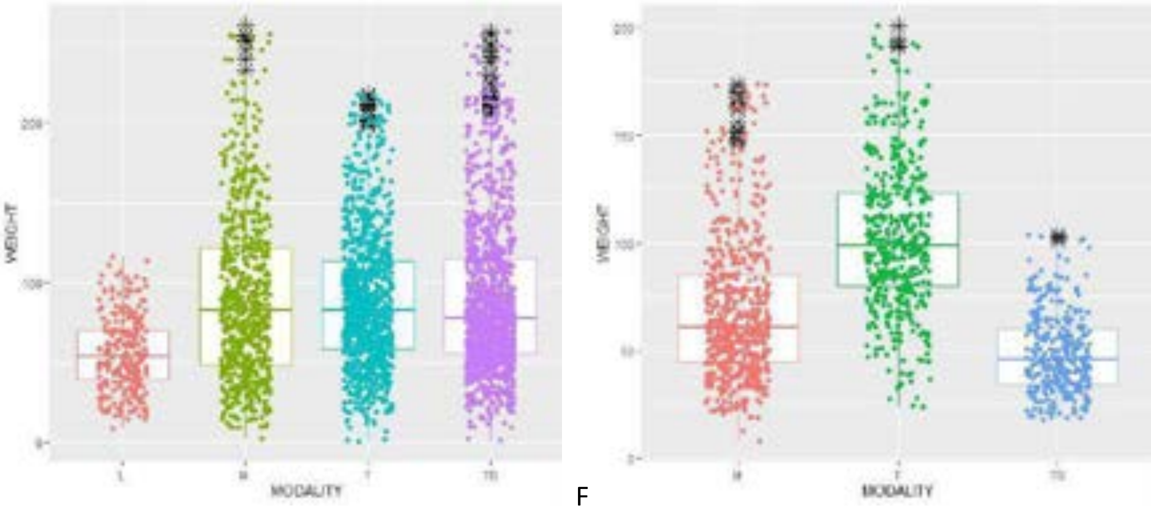

E F

Figure 7. Average fruit weight depending on agroforestrial modality in 2025 (A), 2016 (B), 2017 (C), 2018 (D), 2019 (E) and 2020 (F). L Light pruning; M medium pruning; T pollard; TS sun control.

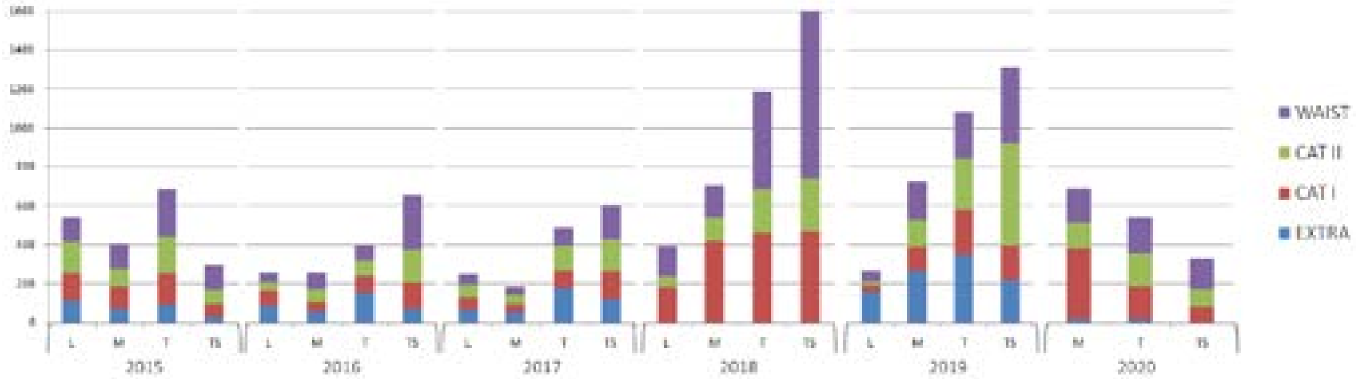

Figure 8. Tomato quality depending on agroforestrial modality for each monitoring year. L Light pruning; M medium pruning; T pollard; TS sun control.

Characterisation of fungal and bacterial communities

At the community scale, 568479 sequences were distributed in 1317 clusters for bacteria. At the γ-diversity scale (total microbiota diversity), the tomato root endospheric bacterial community was mainly composed by Proteobacteria (811 sequence-clusters, 68% of the sequences) and the Bacteroidota (312 sequence-clusters, 18% of the sequences) phyla (Figure 9). Actinobacteria, Firmicutes and Verrucomicobiota were less represented with a richness of 73, 31 and 29 sequence-clusters respectively (Figure 6).

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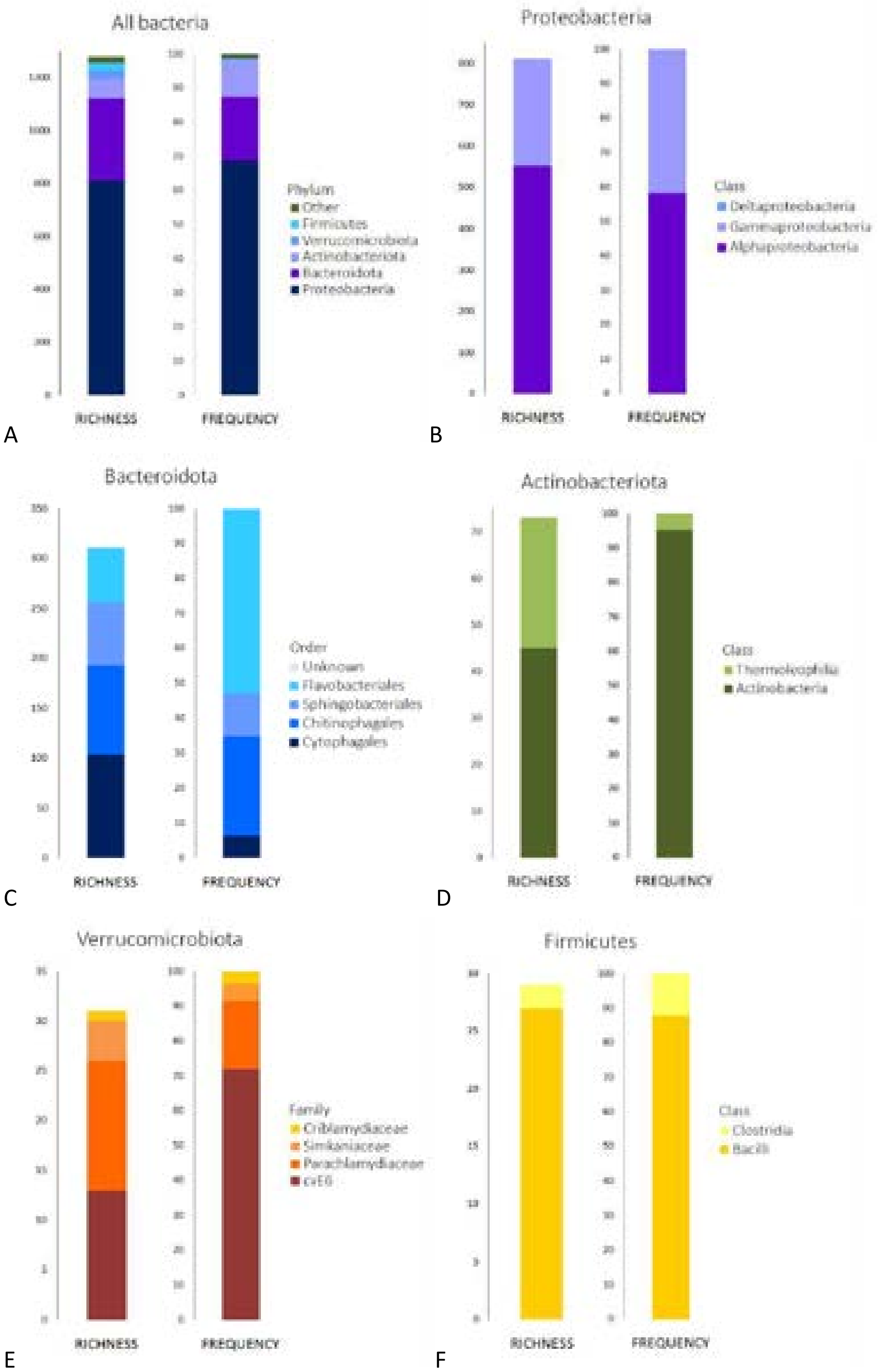

Figure 9. Sequence-clusters richness and relative abundance of the tomato root endospheric bacterial microbiota (γ-diversity) and within the most represented phyla.

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Considering fungi, 5727061 sequences were distributed in 278 clusters. The tomato endospheric microbiota was composed by 85.0% of Ascomycota (125 sequence-clusters), 8.4% of Basidiomycota (56 sequence-clusters), 2.4% of Chytridiomycota (53 sequence-clusters), 2.2% of Glomeromycota (15 sequence-clusters) and 1.4% of Zygomycota (24 sequence-clusters) (Figure 10).

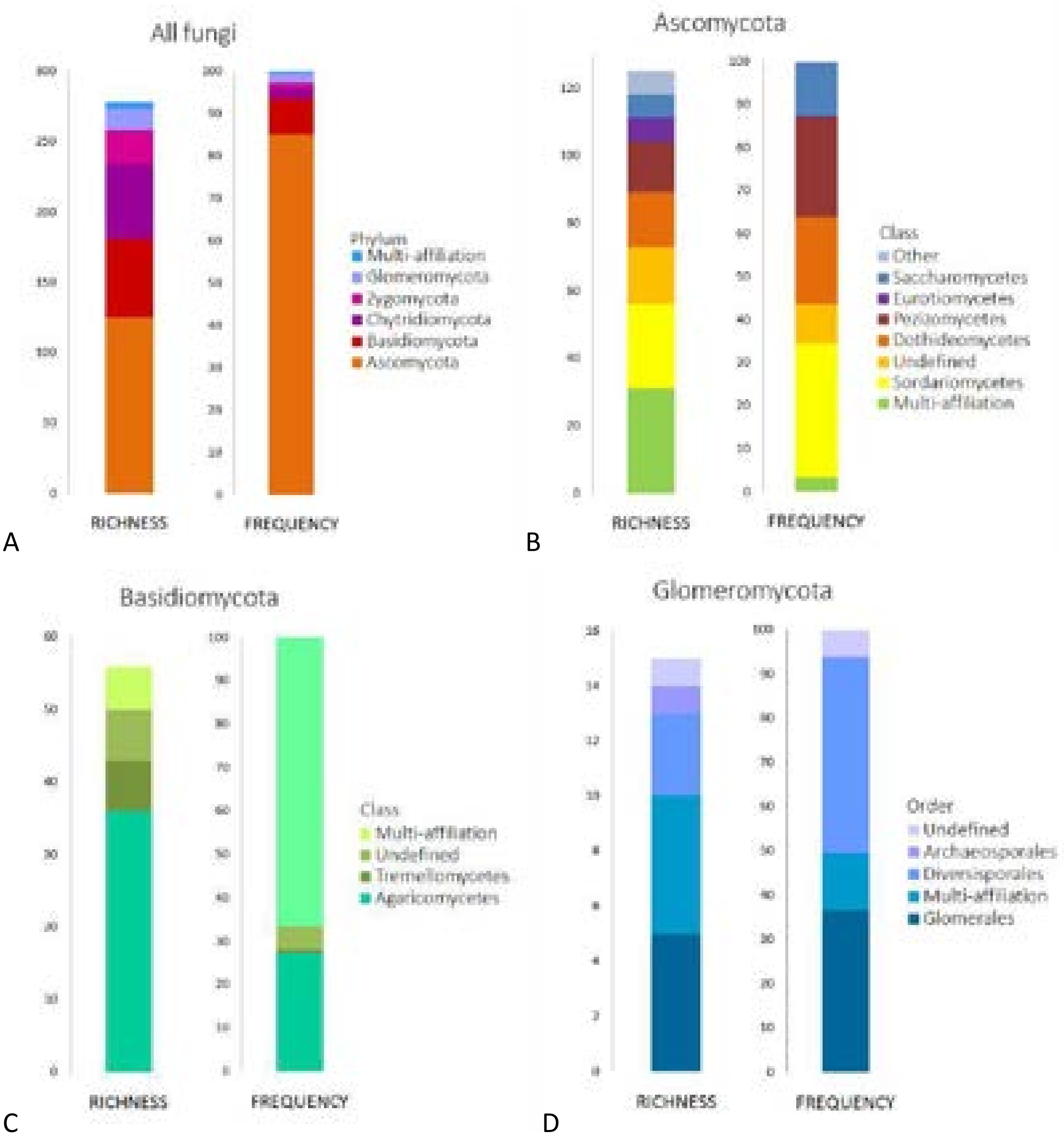

Figure 10. Sequence-clusters richness and relative abundance of the tomato root endospheric fungal microbiota (γ-diversity) and within the main phyla.

Year and agroforestrial modality have a strong impact on fungal and bacterial microbiome composition, but variety has a lower effect

ANOVA results show a predominance of year and agroforestrial modality on both bacterial (table 3) and fungal (table 4) microbiota. On the other hand, variety only has a significant effect on Firmicutes' Shannon index (table 3), fungal community specific richness as well as Ascomycota and Zygomycota specific richness (table 4).

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Variable		Year		Farm		Agroforestrial modality		Variety	
		F	p-value	F	p-value	F	p-value	F	p-value
	Specific richness	0.4359	0.510001	0.9548	0.32981	3.3383	**0.006692 ****	0.2608	0.610242
	Shannon index	24.7945	**1.571e-06 *****	0.0022	0.96285	2.8841	**0.01588 ***	0.0007	0.97842
	Pielou's equitability	35.9339	**1.211e-08 *****	0.2873	0.59261	1.9721	0.08524 .	0.0200	0.88760
Community	Simpson index	6.6151	**0.01098 ***	0.0008	0.977026	1.6852	0.14074	1.3927	0.23961
	Specific richness	17.9039	**3.812e-05 *****	0.7998	0.37236	0.9476	0.4518	0.2305	0.6318
	Shannon index	20.3717	**1.195e-05 *****	0.4392	0.5084	1.6182	0.1578	0.3684	0.5447
	Pielou's equitability	18.4052	**3.696e-05 *****	0.2769	0.59967	1.4123	0.2249	0.1598	0.6901
Acidobacteria	Simpson index	1.2075	0.27340	0.2687	0.6049	0.3107	0.90603	0.0300	0.86267
	Specific richness	5.9914	**0.015404 ***	0.9240	0.33771	1.4605	0.205374	0.2341	0.629097
	Shannon index	1.3729	0.242968	0.3730	0.5421570	2.9516	**0.013980 ***	0.0485	0.825898
	Pielou's equitability	9.0106	**0.0030938 ****	0.0213	0.8840252	4.3917	**0.0008764 ****	0.4263	0.5147030
Actinobacteria	Simpson index	0.6207	0.41192	0.7430	0.38986	2.9615	**0.01372 ***	0.0323	0.85753
	Specific richness	49.9925	**3.954e-11 *****	9.5243	**0.002348 ****	10.9491	**3.904e-09 *****	0.0948	0.75858
	Shannon index	3.2854	0.07168 .	3.9941	**0.04717 ***	9.3735	**6.78e-08 *****	0.0997	0.75253
	Pielou's equitability	1.2463	0.2659	0.8735	0.35123	5.8966	**4.763e-05 *****	0.1663	0.6839
Bacteroidota	Simpson index	0.1167	0.7331	1.6605	0.1992	5.8297	**5.418e-05 *****	0.9968	0.3195
	Specific richness	35.7055	**1.334e-08 *****	0.6789	0.411054	10.8686	**4.509e-09 *****	3.1779	0.07645 .
	Shannon index	27.2058	**5.328e-07 *****	0.1460	0.70287	5.8034	**5.700e-05 *****	4.9556	**0.02734 ***
	Pielou's equitability	0.9811	0.32359	5.2189	**0.02370 ***	1.8711	0.10299	3.6221	0.05903 .
Firmicutes	Simpson index	8.8171	**0.003421 ****	0.1746	0.6765719	2.3181	**0.045613 ***	0.5183	0.472549
	Specific richness	3.8993	**0.049945 ***	6.2996	**0.01296 ***	3.2480	**0.007954 ****	0.1039	0.747585
	Shannon index	55.4869	**4.705e-12 *****	0.1809	0.67109	1.2047	0.30908	0.0532	0.81788
	Pielou's equitability	65.7108	**1.035e-13 *****	0.4037	0.5260	0.6230	0.68242	0.0302	0.86223
Proteobacteria	Simpson index	28.6002	**2.873e-07 *****	0.0271	0.8694	0.8511	0.5155	0.1278	0.7211
	Specific richness	116.4313	**< 2.2e-16 *****	17.6966	**4.081e-05 *****	8.0754	**7.541e-07 *****	2.6814	0.10340
	Shannon index	84.7341	**< 2.2e-16 *****	13.5646	**0.0003046 *****	7.4961	**2.244e-06 *****	1.3648	0.24437
	Pielou's equitability	3.2720	0.07275 .	3.1242	0.07925 .	2.4206	**0.03898 ***	0.5067	0.47782
Verrucomicrobiota	Simpson index	23.1327	**3.343e-06 *****	3.6142	0.05888 .	3.1789	**0.009075 ****	1.6591	0.199500

Table 3. ANOVA results for bacterial community and main phyla structuration characterised by diversity indexes, depending on year, farm, agroforestrial modality and tomato variety.

Variable		Year		Farm		Agroforestrial modality		Variety	
		F	p-value	F	p-value	F	p-value	F	p-value
Community	Specific richness	585.0295	**< 2.2e-16 *****	2.0564	0.1532741	8.4073	**3.937e-07 *****	11.7541	**0.0007607 ****
	Shannon index	0.7281	0.3947	0.9630	0.3277	1.7355	0.1290	0.0648	0.7994
	Pielou's equitability	54.5152	**6.46e-12 *****	0.4962	0.4821	0.6926	0.6297	0.1462	0.7027
	Simpson index	0.0240	0.8770291	0.0759	0.7832	0.5287	0.7543747	0.2022	0.6534981
Ascomycota	Specific richness	799.6651	**< 2.2e-16 *****	0.6646	0.415997	2.6211	**0.0259703 ***	11.2447	**0.0009833 ****
	Shannon index	0.8802	0.3495	0.0334	0.8553	0.4684	0.7994	0.0044	0.9470
	Pielou's equitability	48.6902	**6.309e-11 *****	0.0163	0.8985	0.8204	0.536671	0.5515	0.458710
	Simpson index	0.6582	0.418307	0.4475	0.5044	0.8863	0.491552	0.0006	0.980057
Basidiomycota	Specific richness	166.2795	**< 2.2e-16 *****	6.6564	**0.01066 ***	6.7313	**9.419e-06 *****	2.7321	0.10018
	Shannon index	0.0014	0.9701110	5.9530	**0.01565 ***	5.1450	**0.0002012 *****	0.0016	0.9686109
	Pielou's equitability	30.2988	**1.363e-07 *****	4.7318	**0.03091 ***	4.2238	**0.001213 ****	0.2154	0.643198
	Simpson index	1.1949	0.2758716	7.9816	**0.00525 ****	4.4992	**0.0007055 *****	0.0844	0.7717091
Glomeromycota	Specific richness	8.5429	**0.003938 ****	18.4563	**2.819e-05 *****	21.5421	**< 2.2e-16 *****	2.2179	0.138264
	Shannon index	11.1460	**0.001034 ****	36.9470	**6.898e-09 *****	17.8638	**3.138e-14 *****	0.0603	0.806295
	Pielou's equitability	46.1628	**2.166e-10 *****	16.4508	**7.629e-05 *****	4.0362	**0.001796 ****	2.7286	0.100576
	Simpson index	1.1949	0.2758716	40.4139	**1.592e-09 *****	4.4992	**0.0007055 *****	0.0844	0.7717091
Zygomycota	Specific richness	142.1083	**< 2.2e-16 *****	0.5286	0.46812	2.0436	0.0749324 .	5.9875	**0.0154196 ***
	Shannon index	0.3869	0.534748	0.6073	0.4368172	1.6567	0.147647	1.8100	0.180290
	Pielou's equitability	71.7006	**1.916e-14 *****	0.3819	0.537449	1.5282	0.18418	0.0001	0.99185
	Simpson index	2.1166	0.14754	1.0331	0.310777	2.2581	0.05082 .	0.4279	0.51392
Chytridiomycota	Specific richness	8.5429	**0.003938 ****	18.4563	**2.819e-05 *****	21.5421	**< 2.2e-16 *****	2.2179	0.138264
	Shannon index	11.1460	**0.001034 ****	36.9470	**6.898e-09 *****	17.8638	**3.138e-14 *****	0.0603	0.806295
	Pielou's equitability	46.1628	**2.166e-10 *****	16.4508	**7.629e-05 *****	4.0362	**0.001796 ****	2.7286	0.100576
	Simpson index	17.6343	**4.299e-05 *****	40.4139	**1.592e-09 *****	14.8188	**4.518e-12 *****	0.0000	0.998523

Table 4. ANOVA results for fungal community and main phyla structuration characterised by diversity indexes, depending on year, farm, agroforestrial modality and tomato variety.

When looking at diversity indexes depending on agroforestrial modalities, distinct patterns between bacterial and fungal communities appear. Bacteria tend to be more diversified in term of sequence-clusters, with a more equitable repartition following the light gradient in a linear relation (Figure 11); on the other hand, fungi tend to react to "extreme" environments (Light pruning, Sun Control TS and S) (Figure 12).

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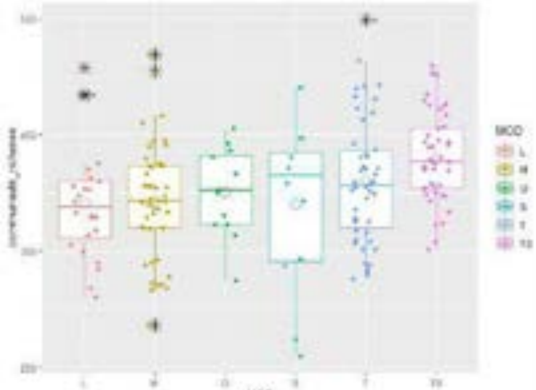

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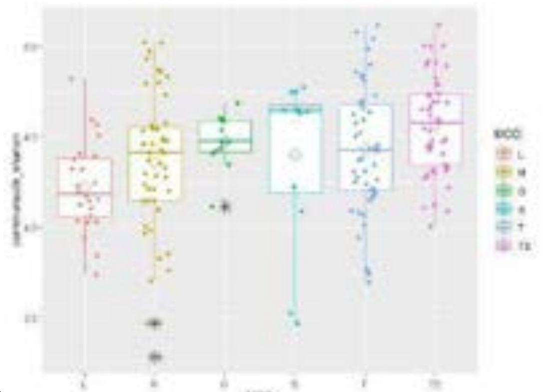

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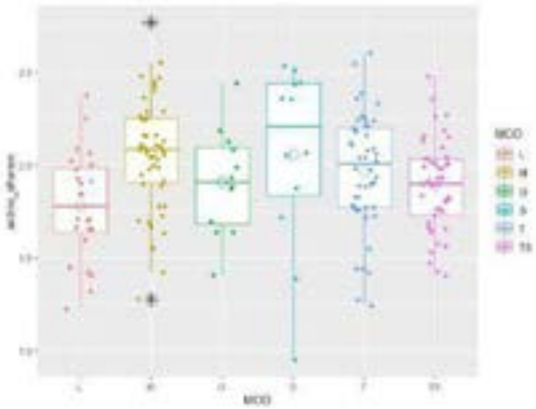

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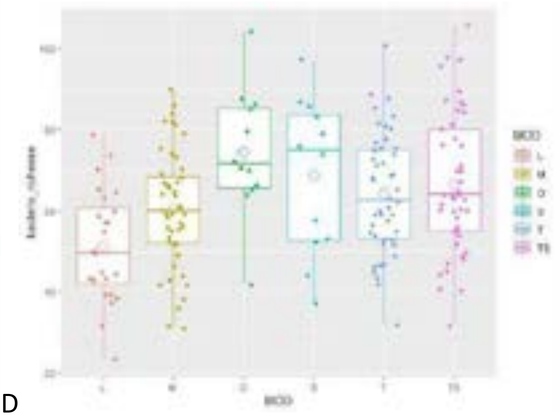

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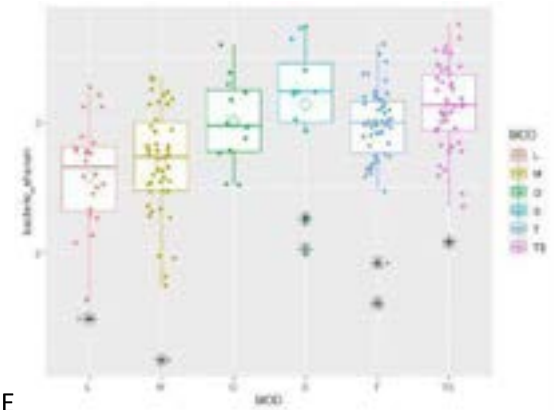

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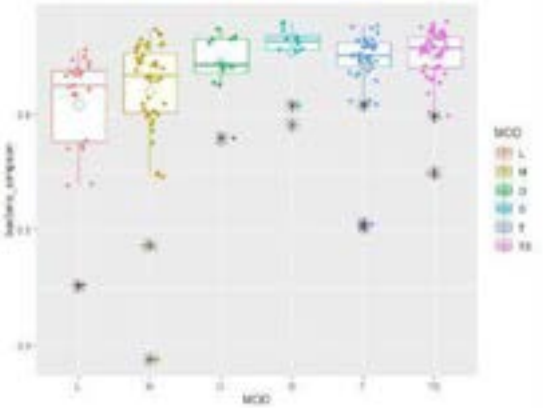

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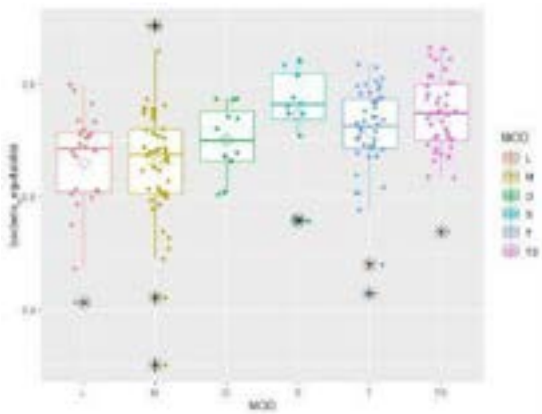

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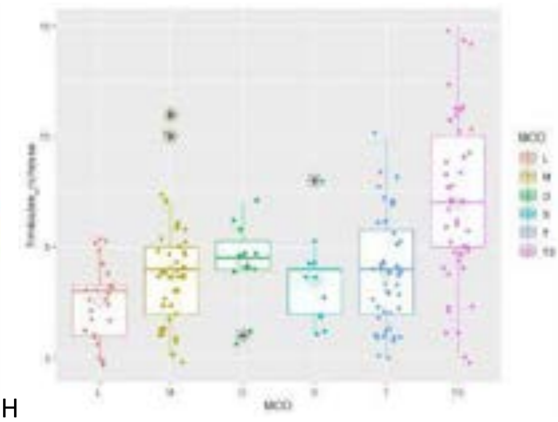

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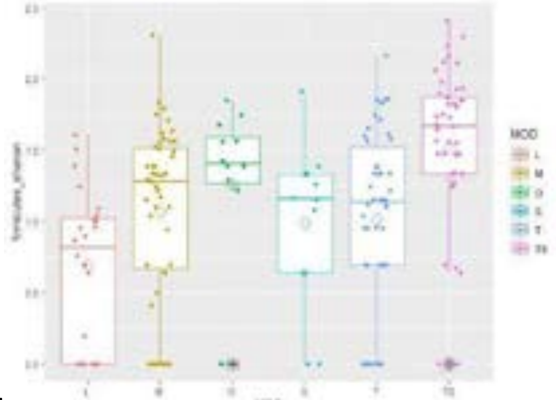

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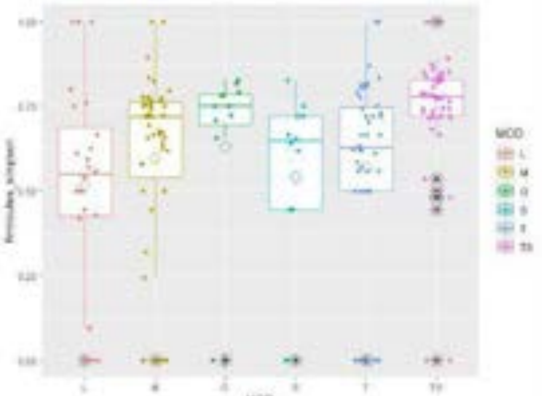

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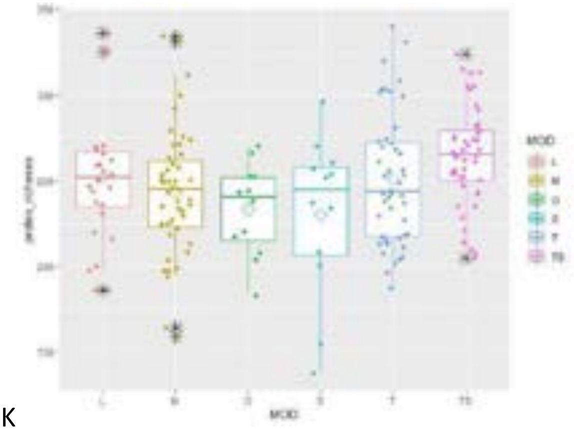

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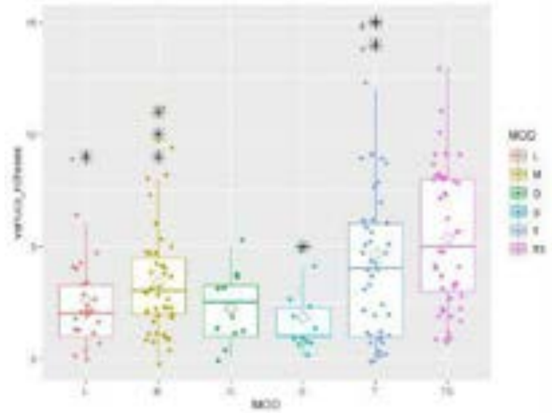

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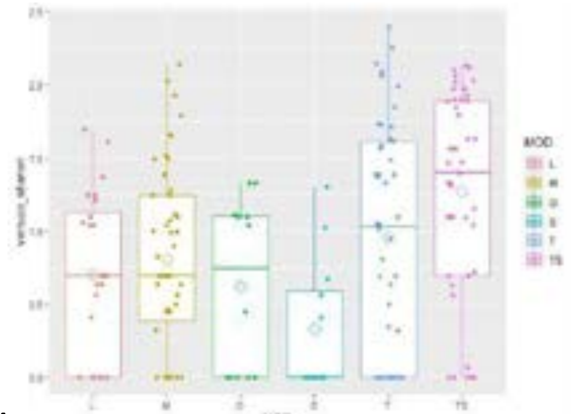

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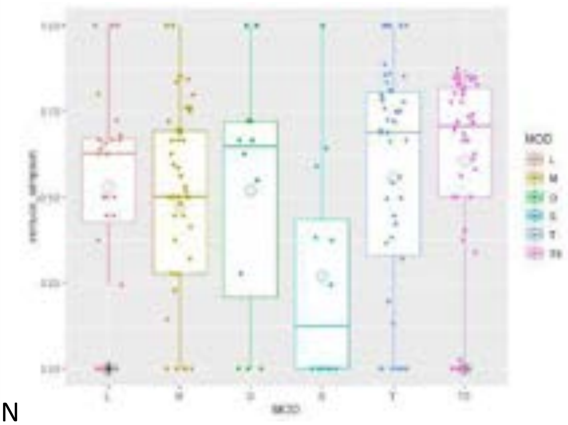

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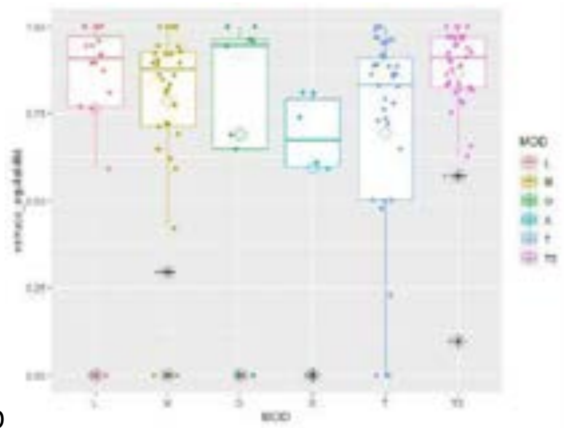

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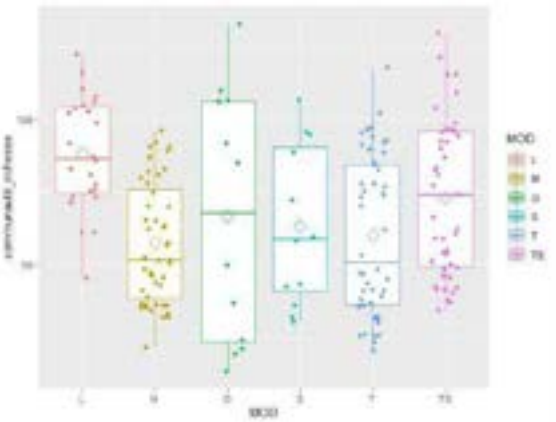
Figure 11, Diversity indexes of bacterial communities for which a significant effect of agroforestrial modality is observed. L Light pruning; M medium pruning; T pollard; TS sun control; O shade modality at Boulidou farm; S sun control at Boulidou farm. A Community specific richness; B Shannon index of community; C Shannon index of actinobacteria; D Bacteroidota specific richness; E Bacteroidota Shannon index; F Bacteroidota Simpson index; G Bacteroidota equitability; H Firmicutes specific richness; I Firmicutes Shannon index; J Firmicutes Simpson index; K Proteobacteria specific richness; L Verrucomicrobiota specific richness; M Verrucomicrobita Shannon index; N Verrucomicrobitota Simpson index; O Verrucomicrobiota equitability.



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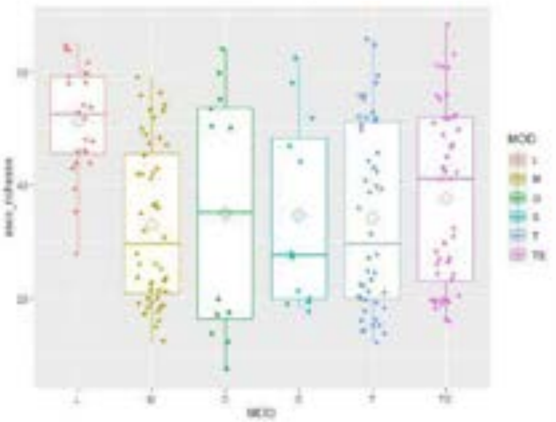

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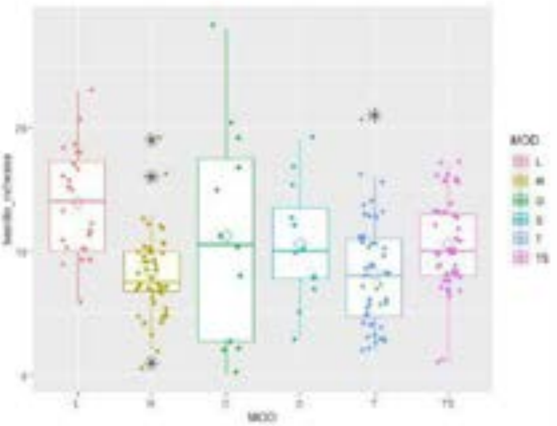

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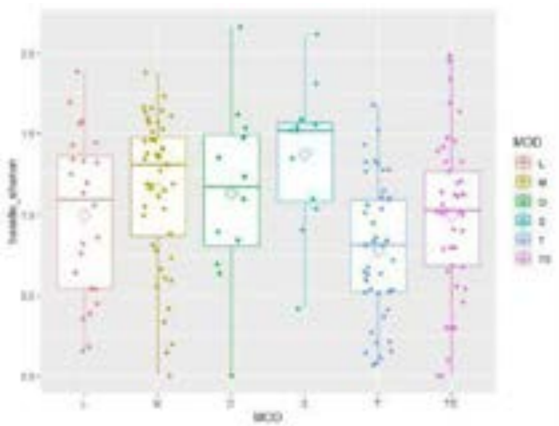

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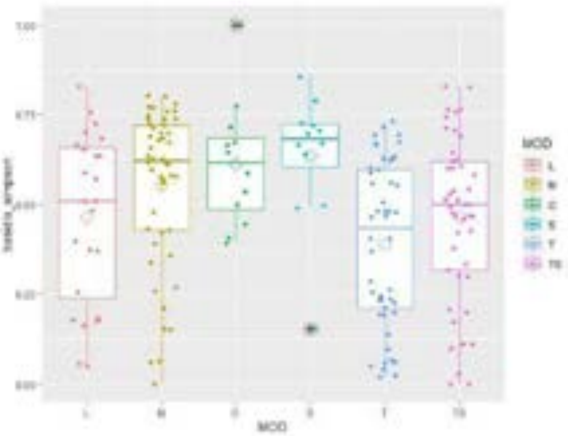

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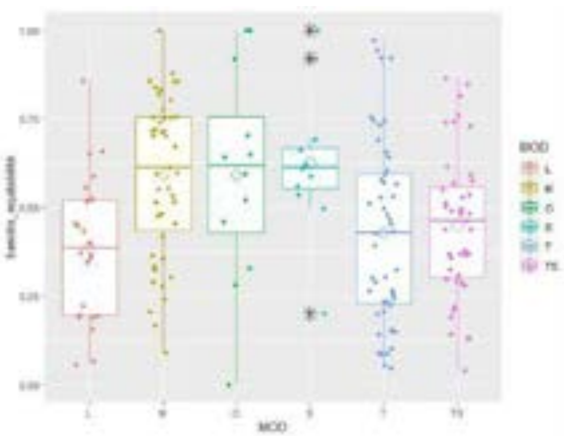

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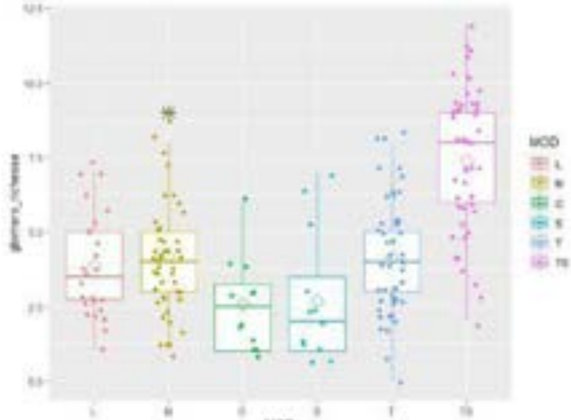

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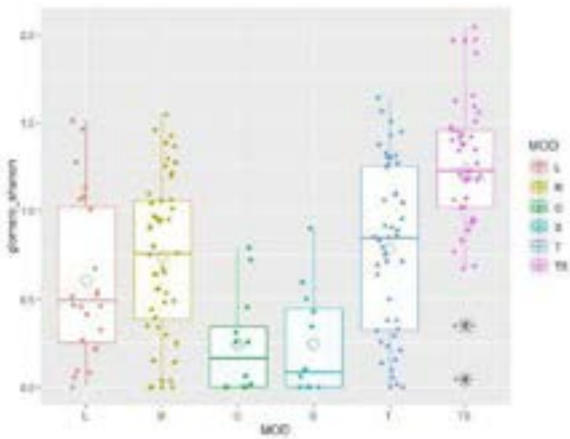

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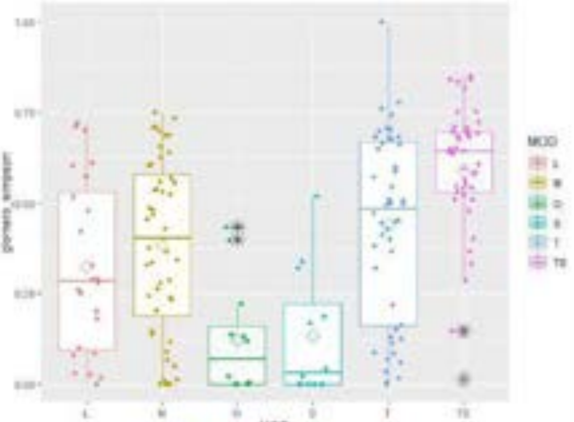

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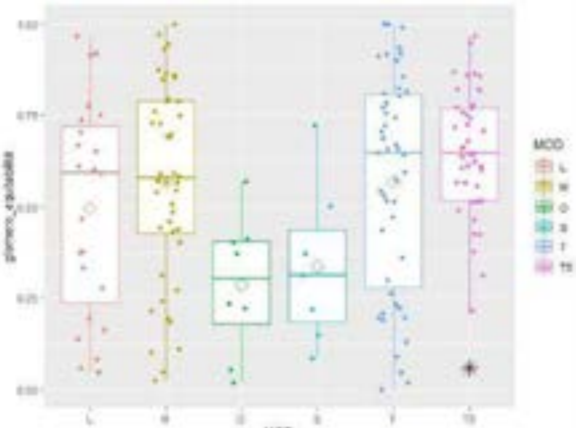

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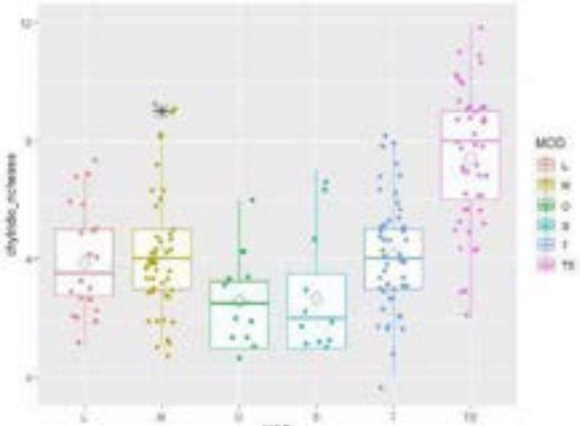

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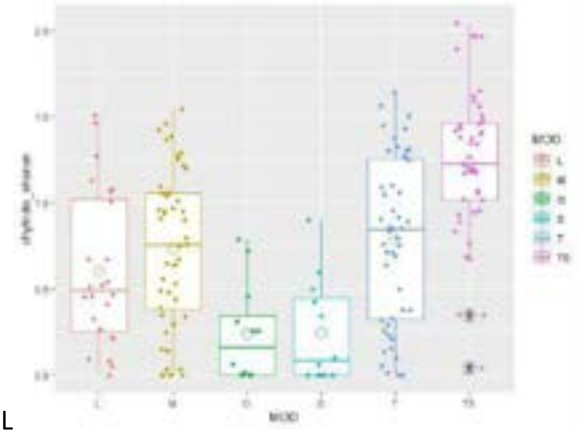

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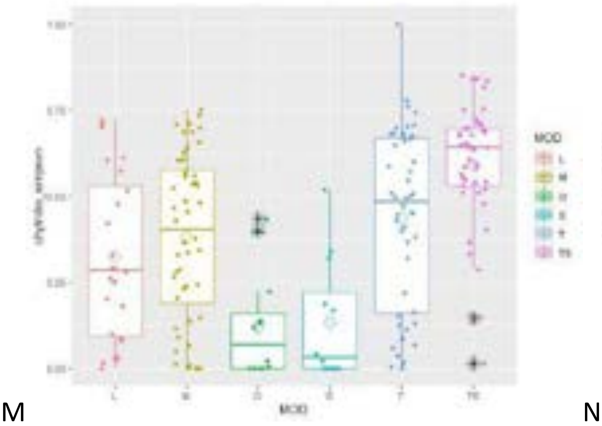

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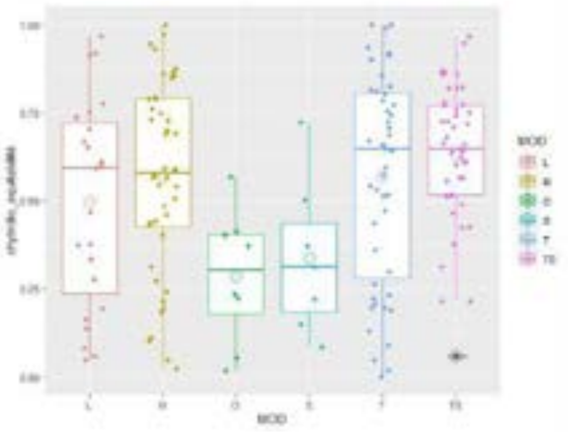

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Figure 12. Diversity indexes of fungal communities for which a significant effect of agroforestrial modality is observed. L Light pruning; M medium pruning; T pollard; TS sun control; O shade modality at Boulidou farm; S sun control at Boulidou farm. A Community specific richness; B Ascomycota Shannon index; C Basidiomycota specific richness; D Basidiomycota Shannon index; E Basidiomycota Simpson index; F Basidiomycota equitability; G Glomeromycota specific richness; H Glomeromycota Shannon index; I Glomeromycota Simpson index; J Glomeromycota equitability; K Chytridiomycota specific richness; L Chytridiomycota Shannon index; M Chytridiomycota Simpson index; N Chytridiomycota equitability.

Most of sequence clusters are shared among agroforestrial modalities, but their structuration varies between them

Venn diagram showing shared and unique sequence-clusters between agroforestrial modalities display that clusters are mainly shared between at least 4 modalities. They are very few unique sequence clusters (Fig. 13 & 14).

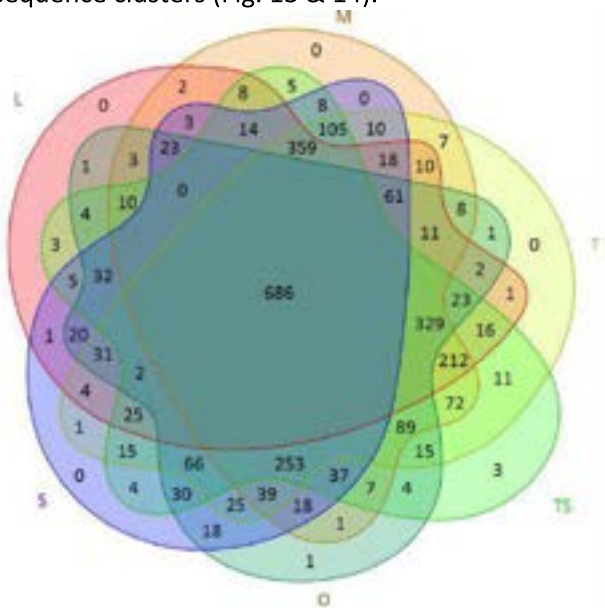

Figure 13. Venn diagram showing bacterial sequence-clusters repartition. L Light pruning; M medium pruning; T pollard; TS sun control; O shade modality at Boulidou farm; S sun control at Boulidou farm.

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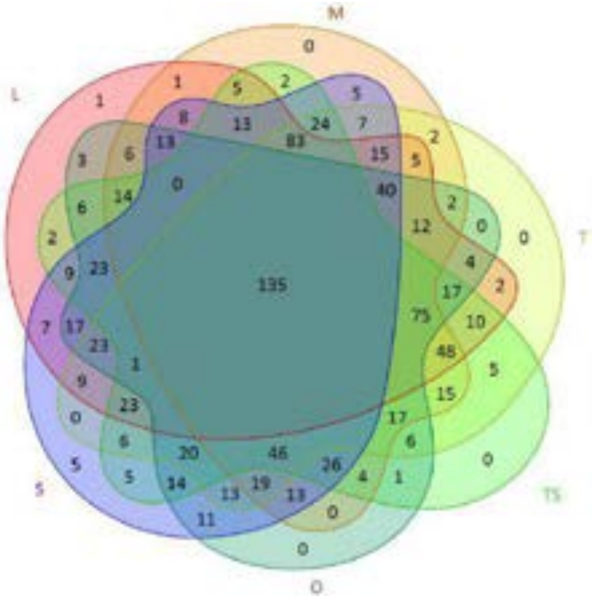
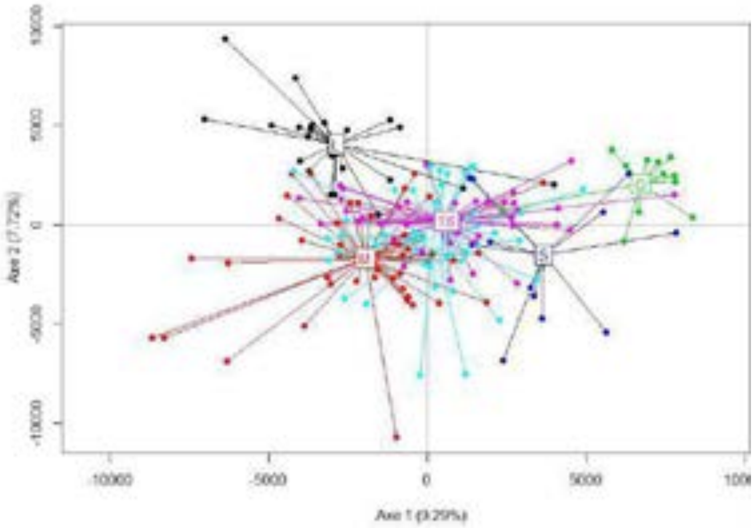


Figure 14. Venn diagram showing fungal sequence-clusters repartition. L Light pruning; M medium pruning; T pollard; TS sun control; O shade modality at Boulidou farm; S sun control at Boulidou farm.

Partial Least Squares Discriminant Analysis (PLS-DA) ordination of the bacterial and fungal communities depending on agroforestrial modalities represent these contrasted colonization patterns (Figure 15). Bacteria tend to segregate accordingly to the light gradient, with a sensitivity to the sampling site, while fungi tend to differentiate among extreme environments (deep shade L or sun control TS and S).



A

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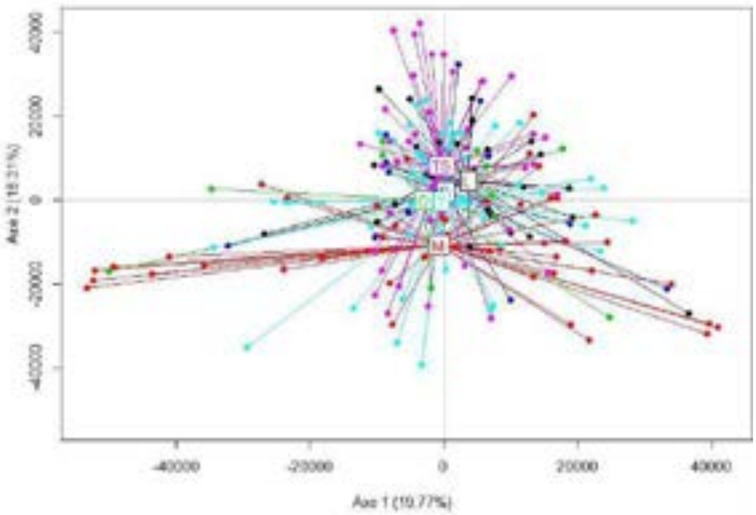

B

Figure 15. Partial Least Squares Discriminant Analysis (PLS-DA) ordination of the bacterial (A) and fungal (B) community in Light pruning (L), medium pruning (M), pollard (T), sun control (TS at Roumassouze and S at Boulidou farms), and agroforestrial modality at Boulidou (O) on the total community (1317 sequence-clusters for bacteria and 278 for fungi).

Discussion, outcomes, and conclusion

ANOVA and chemotype analysis confirm that on-farm selection maintains the specificity of both populations, which allows us to assess that we are working with two distinct varieties. Regarding the influence of agroforestry on genotype, molecular marking on 8 SSR only might not be enough to observe a genetic structuration if there is one. On the other hand, we have clear evidence of the effect of agroforestry on biochemical and physical traits of tomato, with a direct impact of light gradient on the considered traits. However, these analyses only concern fruits from 2019; it would have been interesting to consider these traits for several sampling years to study chemotype variations through on-farm selection.

Speaking of on-farm selection, we cannot directly assess its effect on microbiome structuration, as we do not have a "non-selected" control. The experiment being on-farm, having such a population could impair the farmer's work. However, the very weak effect of variety on both bacterial and fungal communities is surprising in regard of literature (Bergna et al., 2018; Bulgarelli et al., 2012; Bulgarelli et al., 2015; Kwak et al., 2018; Lebeis et al., 2015; Lundberg et al., 2012 ; Peiffer et al., 2013 ; Rasche et al., 2006) despite their difference. This result could be an indication of co-evolution between plants and micro-organisms being supported by on-farm selection, with possible epigenetic regulations as well (Vannier et al., 2015). Indeed, on-farm selection makes the most of 3 types of heredity: genetic, epigenetic and microbiotic. In our study, the genetic component seems to be balanced at least by the microbiotic one, as the epigenetic regulation has not been explored.

Our results are in accordance with the holobiont hypothesis framework (Margulis, L., 1981; Vandenkoornhuyse et al., 2015) associated to the hologenome theory (Zilber-Rosenberg, I., & Rosenberg, E., 2008). These paradigm shifts in the plants' conception are necessary in organic plant breeding aiming at resilience, in the way that they contribute to a holistic, system-based approach.

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On-farm selection, rather unconsciously, already makes the most of these interactions, as it considers plants phenotype as an expression of genetic, epigenetic and microbiotic adaptation to a given terroir.

The results of this study, even with some partial data from one year to another, give a first idea the drivers of plants and microbiome co-evolution through on-farm selection in an agroforestrial context. These results could inspire organic breeding to integrate different levels of heredity when willing to select for resilient cropping systems and are a first step toward a more holistic agriculture.

Publications

Lemichez, S. (2020). *Comprendre les performances et l'adaptation de deux variétés-populations de tomate (S. lycopersicum) dans un système agroforestier via l'étude du microbiome racinaire dans le cadre d'une recherche participative* (Doctoral dissertation, INRAE, AGROCAMPUS OUEST).

Lemichez, S., & Chable, V. (2021). *Drivers of endospheric root microbiota assembly of on-farm selected tomato in agroforestry.* BREEDING AND SEED SECTOR INNOVATIONS FOR ORGANIC FOOD SYSTEMS, 86.

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IPC-Agroforestry trials 2019-2020 (Trial #26)

Lead: IPC Portugal
Pedro Mendes Moreira, Duarte Pintado, Rosa Guilherme – pmm@esac.pt

Abstract

Syntropic Agriculture is a Succession Agroforestry System (SAF). AF it is distinguished by the cultivation, in the same place, with a diversity of species, that co-operate in a synergic and synchronized way, optimizing their productive cycles, creating rational income and positive energetic balance within the system. It requires an equilibrium between crops with ecological and economic function.

In order continue to promote Syntropic Agriculture in Portugal, a field trial was installed at ESAC-IPC, two meetings engaging 50 interested people in the topic and a master's thesis was accomplished. From the species used, maize CCP was included in co-breeding, so as two beans. The harvest seeds from the 2019-2020 trials were collected to be used in the next season in 2020.

Introduction

Portugal has a long tradition in Agroforestry Systems (e.g., Enforcado, Montado). The needed to find sustainable solutions for the recuperation of burned areas in Portugal, recuperation of uncultivated soils and provide economic benefits, led us to search new approaches and to have the opportunity to disseminate them using IPC-ESAC for practice and dissemination. The agroforestry adapted was the Syntropic Agroforestry that was developed by Ernst Götsch. Syntropic Agriculture is not a recipe, but a practice in which natural processes are translated into farming interventions in their form, function, and dynamics. Based on the interaction among species to explore synergies, and space above and below soil, creating a process of regeneration that generates, high productive agricultural areas with higher independence on inputs and irrigation and contribute for the ecosystem services (e.g., formation of soil, regulation of microclimate and favouring water cycles).

Aiming to promote and adapt this system to Portuguese reality, in particular, in Centro Region, an experimental trial field was installed in ESAC- IPC, in the certified area of Organic Agriculture, within the scope of a master thesis of the Organic Master Science Course.

The team involved in the conception project were Rosa Guilherme, Walter Sandes and Pedro Mendes Moreira. The implementation of trial included Daniela Santos, Isabel Dinis, Duarte Pintado, farmers, and other stakeholders involved in Agroforestry. A development of a maize CCP and beans in co-breeding for Agroforestry Intercropping Systems, did start also.

A relationship between D3.3 regarding de analyses of microbioma in this system, compared with organic in low-input and conventional was also tested.

Material and Methods

Genetic resources

In 2019 syntropic trials were established. The trial included several types of vegetables, fruit trees, trees for biomass and aromatic plants.

From the species used we had focus on maize CCP ‘SinPre’, and two bean genotypes (‘Catarino’ and ‘Vermelho’).

The syntropic systems included 16 perennial species including trees for biomass, fruit trees and shrubs. The horticulture included 11 species plus maize and beans.

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In 2020 the maize CCP 'Sinpre' was under selection again with one maize genotype ('Feijão Frade').

Location

The trials were managed at the organic field of IPC-ESAC in Bencanta, Coimbra Portugal (40.21709426119619; -8.44779968261719; 15 m) in 2019.

Experimental design

The trial was installed in early April 2019 with both yield and pedagogic purposes.

Two workshops were held in total, on 9 and 15 April with a total of 50 participants that contributed for the trial's installation. The organization of the workshops was a partial fulfilment of the Master Thesis of Walter Sandes.

The trial comprehends a total area of 400 m^2 (long-term) in organic agriculture ESAC-IPC's field. No preceding crops neither fertilization since the last campaign were done. The Figures bellow illustrate the phases of the trial implementation (Figure 1), the design of the plots (Figure 2, p91) and a view of the trial (Figure 3, p92).

Figure 1. Phases of the implementation of the Agroforestry System in Syntropic (Sandes, 2019)

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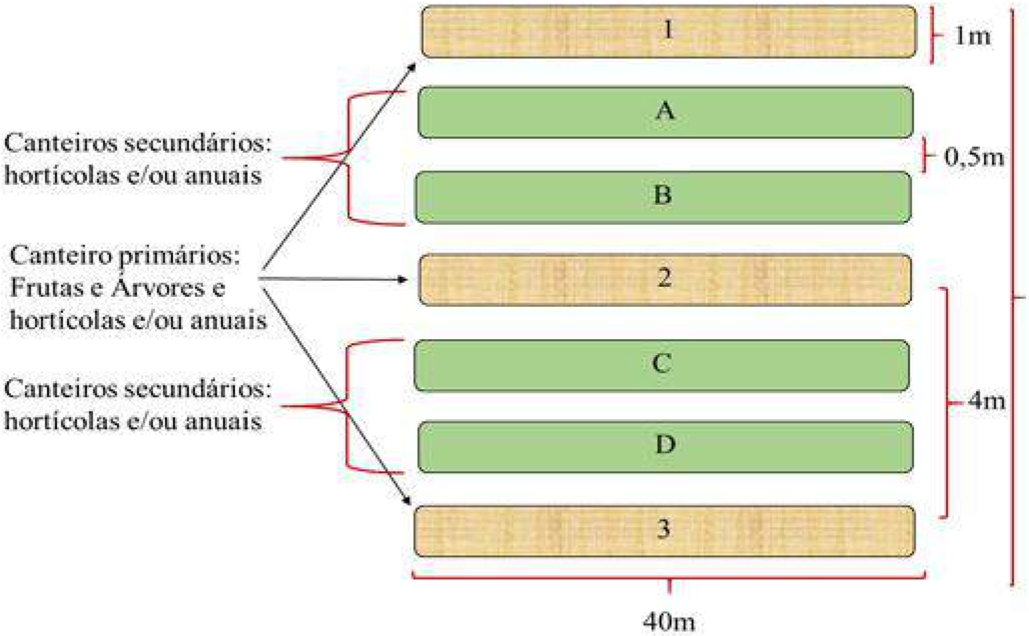

Figure 2. Distribution, dimension, and types of plots used. The numbers (1, 2, 3) refers to Primary plots (the plots where the trees will be implemented); the letters (A, B, C, D) refers to Secondary plots (the plots associated plus with horticulture and annual plants)

Statistical methods

Some qualitative data were observed, the germplasm is under selection.

Evaluation

Table 1. List of the traits collected for concerning Crop development and agro-ecological performance

Crop development and agro-ecological performance (phenology, weeds, diseases, …)		
Trait	**How it has been assessed**	**Type of data available**
Germination Ratio	Counting the number of plants	%

How resilience was improved

During the syntropic system implementation, it was possible to start the co-breeding trial with maize and beans. From this trial it was possible to select seed for 2020.

In 2020 selection continued and 2 kg of seed from the maize CCP 'Sinpre', and 1 Kg of beans adapted to the SAF was stored and sowed for the next season.

Participatory/multi-actor approach

The syntropic agroforestry system gave the opportunity to join 50 participants, that are followers of this study since its beginning, to visit and disseminate the agroforestry systems and their importance.

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Figure 3. View of the Syntropic field trial at IPC-ESAC.

Results and discussion

During the campaign it was possible to observe some synergies and development of the crops installed in the trial.

One of the major problems observed derived from the use of manure, that was not yet mature and brought us some problems for maize and beans crops that are sensitive. The other problem derived from the irrigation system that improve the conditions for fungal diseases in tomato and potato, so as the weeds development. The large quantity of mulch before the sowing could have a direct influence preventing the sun to heat the soil. These events affected maize and beans; however, it was possible to harvest seed for the year 2020. In 2020 was possible to continue with mass selection and 2 kg of maize and 1 Kg of beans were selected. Weed pressure it was very intensive.

The trial implementation strategy allowed the participation of 50 participants in two sessions; from the several sectors such as workers from some representative enterprises, producers, students, professors, and other interested parties. These 50 participants had the opportunity to help in the installation process and learn by doing.

Seeds were obtained from the co-breeding selection for next season.

Outcomes and conclusions

In 2021, a maize trial was sown in syntropic agriculture system in ESAC-IPC, with seeds coming from the CCP worked in this system in 2019, to continue the work of selection and adaptation of the CCP 'Sinpre' to this sustainable and ecological agroforest system.

Publications

Sandes W., Guilherme R., Dinis I., Mendes-Moreira P. (2019) Agricultura Sintrópica na região Centro de Portugal: implementação de um campo de demonstração. IX Congresso da APDEA e o III Encontro Lusófono em Economia, Sociologia, Ambiente e Desenvolvimento Rural (ESADR 2019). Lisboa e em Oeiras, entre 15 e 18 de outubro de 2019.

LIVESEED is funded by the European Union's Horizon 2020 under grant agreement No 727230 and by the Swiss State Secretariat for Education, Research and Innovation (SERI) under contract number 17.00090.

Walter Assunção da Silva Sandes (2019) Implantação da Agricultura Sintrópica na Escola Superior Agrária de Coimbra. Relatório de Projeto apresentado à Escola Superior Agrária de Coimbra para cumprimento dos requisitos necessários à obtenção do grau de mestre em Agricultura Biológica

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Annex II. Results of the survey on multi-actor approach

The survey "Evaluation form for participatory and multi-actor approaches in organic breeding" was developed by IPC and intended to ask project partners about their involvement in multi-actor breeding approaches. The results were obtained from 9 institutions and analysed by IPC.

Question 1: What triggered the setup of the participatory or multi-actor breeding approach involving farmers, processors, bakers, retailers, or consumers? What were the species and major breeding goal?

(Describe concisely what participatory/ multi-actor approaches you have been using so far. Who took the initiative? How did other actors in the chain been involve in decision making and evaluation regarding the materials used, traits measured and setups of the trials? (100 words max.)

Institute	Species	Breeding goal
FIBL-CH	White Lupin (Lupinus albus)	resistance to anthracnose, low alkaloid, earliness
AREI	Spring barley	feed and/or food use, adapted for organic environments
IPC	Maize and Beans	Maize for human uses (e.g., maize bread) and beans for intercropping
ITAB	Soft bread wheat	comparison between CCP and Dynamic Populations
ITAB	Rivet Wheat, spelt, oat	Creating diversified and oriented populations according to farmers' needs
UBIOS	winter wheat	The farmers work on winter wheat and there is a specific objective for each CCP created: CCP1 - for on-farm baking and poor lands, CCP2 - for on-farm baking and good lands and CCP3 for long-chain market and good lands.
GZPK	Summer pea	intercropping with cereals, high protein contents and stable yields
LBI	Wheat trials	To boost local value chains of organic wheat for baking Edwin Nuijten, Edith Lammerts van Bueren c.s. started to select suitable germplasm for the Dutch conditions. Farmers, bakers, and breeders were involved; the researchers determined the setup and traits to assess. This work was continued within LIVESEED and half-way the project taken over by Floor van Malland and Abco de Buck. First own experience with participatory approaches, involving, engaging, and demonstrating and inform about the trial results, mainly during the yearly field day and winter evaluation
LBI	Lupin trials	Same setup to promote local food protein production with an 'unknown' species for the NL. Both trails were conducted in collaboration with farmers at farmers sites to find the most suitable material under their conditions
CULTIVARI	Winter pea for mixed cropping with winter triticale	Main goal is to developed winter pea for mixed cropping with winter triticale. Developing of criteria for breeding ended up after these first trials in relatively tall normal leaf types of pea with good winter hardiness. For total yield of pea and triticale more emphasis must be given to the type of triticale to allow high pea yield without losing to much of triticale yield.
AREI	Spring pea yield potential	protein content. For the growing under organically managed conditions: leaf type and length of plant in relation to weed suppressive ability

Table 1: Species and main breeding goals at partners' PBB/MA trials

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Question 2: How many years of experience do you have in participatory of multi-actor breeding before LIVESEED?

The 50% of the partners have 5 to 10 years' experience in PPB or multi-actor breeding before LIVESEED. LIVESEED promoted 10% of the experiences and enhanced the 30% of the early projects (1 to 4 years)

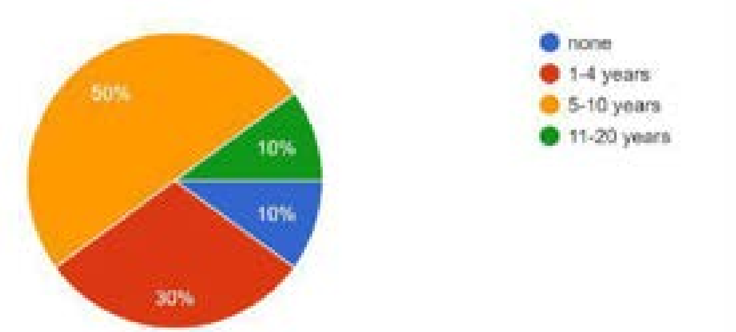

Figure 1. Experience in participatory or multi-actor breeding before LIVESEED

Question 3: What types of funding was used for PBB or multi-actor breeding?
(Indicate all options: licence, royalties, or voluntary contributions on seed; direct seed sale; charity foundations; public funding; organic growers association; processors (millers, bakers, food industry); retailors; consumers; local community; society in general (private donations, crowd funding; Other)

All the initiatives have public funding, the other three initiatives represent each 30% of the funding (licence, royalties or voluntary contributions on seed, charity foundations, organic growers' association).

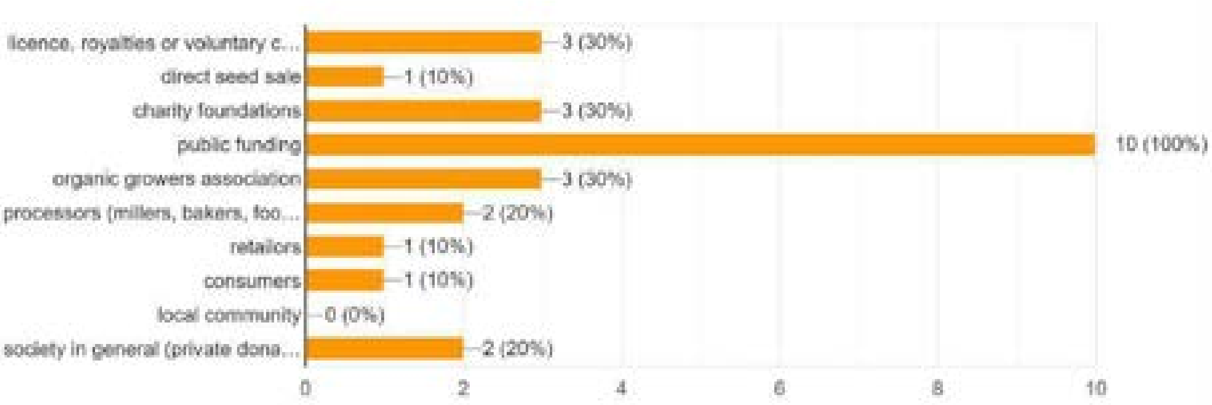

Figure 2. Type of Funding used by partners for PBB/Multi-actor breeding activity

Question 4: What were the main funding types of funding was used for PBB or multi-actor breeding? (Select one option)

When we consider the main funding source, we obtain 66.7% for public funding and 22.2% for charity foundations and 11.1% for organic growers' associations.

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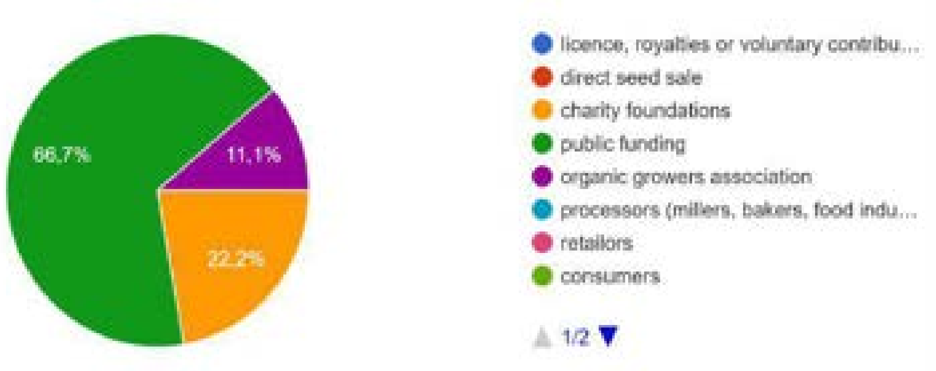

Figure 3. Type of main funding used by partners for PBB/Multi-actor breeding activity

Question 5: What is your institution's status?

The institutional status is distributed by private non-profit organization (60%), public non-profit organization (30%) and private commercial enterprise (10%).

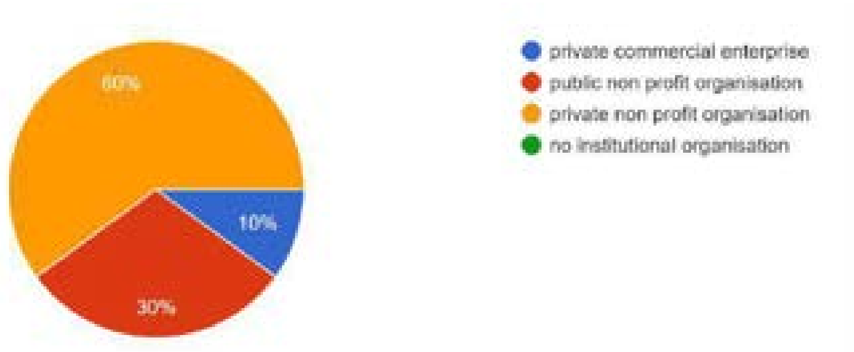

Figure 4. Institutional status of partners

Question 6: How many people are involved in your PBB/Multi-actor breeding activities?

The number of persons involved in breeding is for 90% of the cases 3 to 15 people.

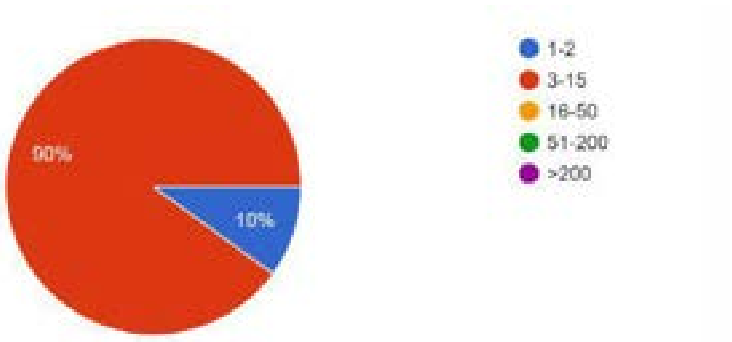

Figure 5. Number of persons involved in breeding at partners

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Question 7: What is the geographical scale for distribution of the cultivars from PBB/Multi-actor breeding?

Geographical scale for distribution of cultivars correspond to 70% at local and national level.

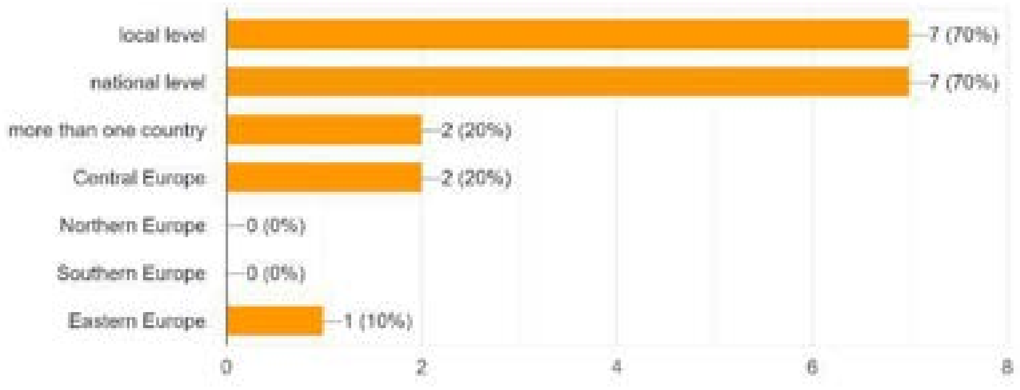

Figure 6. Geographical scale for distribution of cultivars at partners for PBB/MA breeding

Question 8: What is your selection environment? (Please click all that apply)

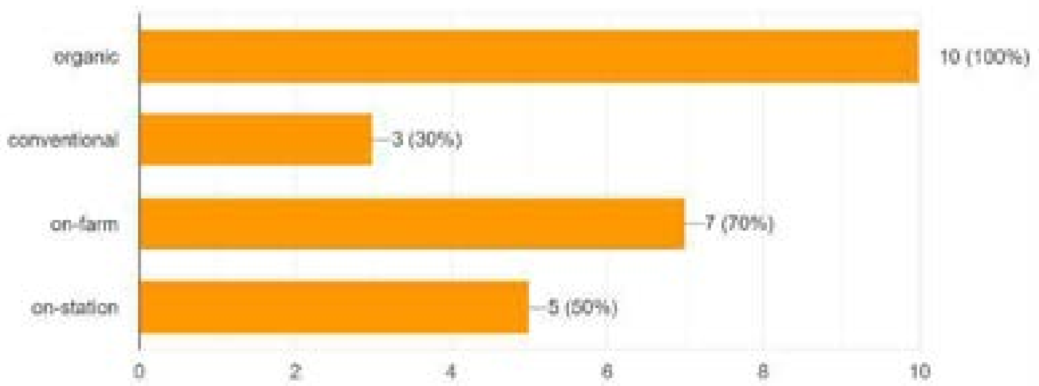

Figure 7. Selection environment of the partners PBB/MA trials

Question 9: What is the number of genotypes tested per year?

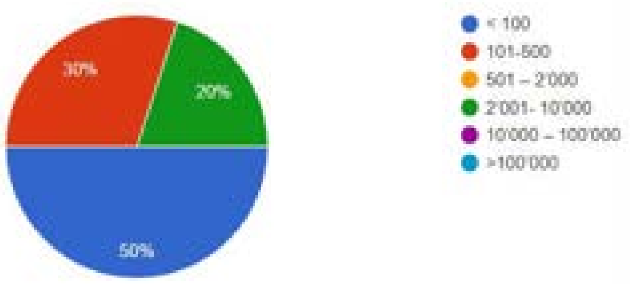

Figure 8. Number of genotypes tested per year at partners in PBB/MA breeding

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The number of genotypes tested per year represent 50% with less than 100 genotypes, and 20% of the institutions have 2001 to 10000 genotypes.

Question 10: What other factors than genotypes are looked at (e.g., different cropping systems, stress environments)?

The 80% of the factors other than genotypes include cropping system followed by stress environment with 40%.

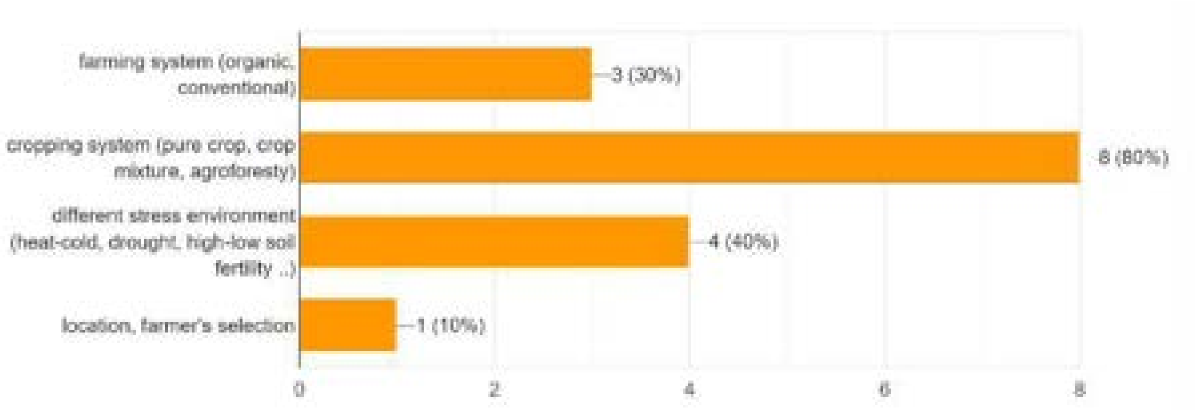

Figure 9. Factors other than genotypes researched at partners in PBB/MA breeding

Question 11: What types of testing your activity include?
(Select small trial plot (1- 20 m2); strip or pilot field trials (100 – 1000m2), nutritional profiling, sensory testing, processing trials, or consumer testing)

The testing included prominently 80% for small trial pot but also nutritional profiling.

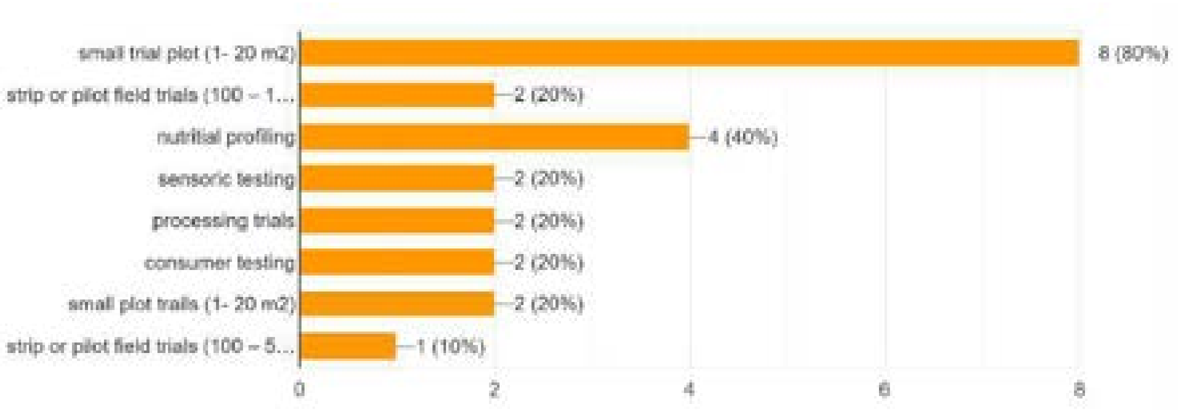

Figure 10 Types of testing at partners PBB/MA breeding

Question 12: If your activity takes place on-farm, what is the role of farmers?
(Select all that applies: collect and maintain germplasm and breeding material conduct crosses; selection of best genotypes on their own; participate in evaluation and selection process together with breeder/researcher; define breeding goals; provide organic field for selecting genotypes

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under organic production; conduct pilot or demo trials of candidate cultivars under organic production; participate in processing and direct sale; participate in on-farm post-release cultivar testing; involved in all aspects from breeding, selection, production, processing and sale; Other)

Figure 11. Indication of the task of the farmers when on-farm in conducted

The task of the farmers in on-farm breeding activities were mainly to "provide organic field for selecting genotypes under organic production" (87.5%) and to participate in the evaluation and selection process together with breeders/researchers (75%).

Question 13: If the trials are on-farm, what are the incentives used for farmers (e.g., they get paid, they get provided seed, completely voluntary, they use the results, etc)?
(Select from the options: Highly motivated to develop own cultivars for short value chain (from seed to plate); Interested to collaborate with other farmers; interested to collaborate with breeders/researchers to get access to seed of improved cultivars; interested to get involved in seed multiplication / seed business; financial reimbursement; pioneer/ early adopter farmer to drive transition of the sector; Other)

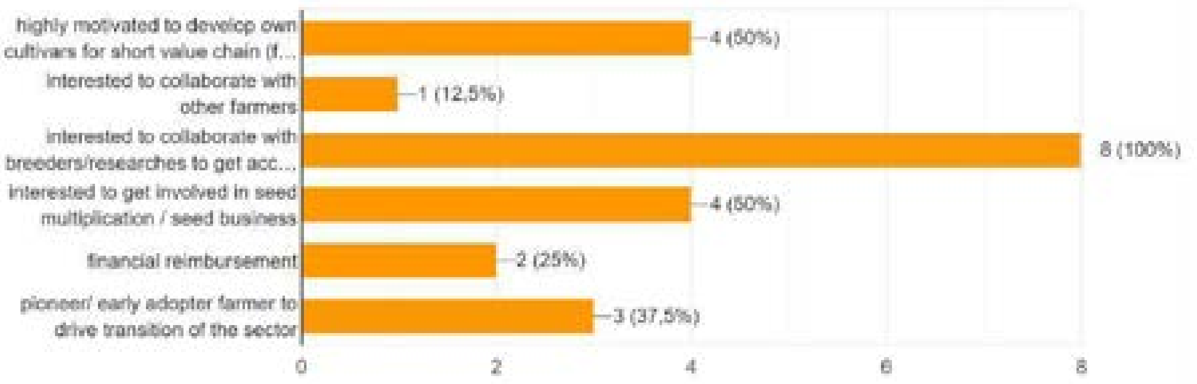

Figure 12. Incentives used for farmers to take part in on-farm trials

The incentive for farmers when trials are on the field is 100% interested to collaborate with breeders/researchers to get access to seed of improved cultivars, followed in 50% by "highly motivated to develop own cultivars for short value chain (from seed to plate)" and interested to get involved in seed multiplication/seed business.

Question 14: What do the main breeding goal address?

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(Select max 6: pest resistance; disease resistance; virus resistance; seed/soil borne diseases; weed competition; lodging tolerance; vigour; drought / flooding tolerance; drought / heat tolerance; cold tolerance; earliness; genetic diversity; yield stability; nutritional quality; organoleptic quality (taste, texture, appearance); processing quality; shelf life / storability; Other)

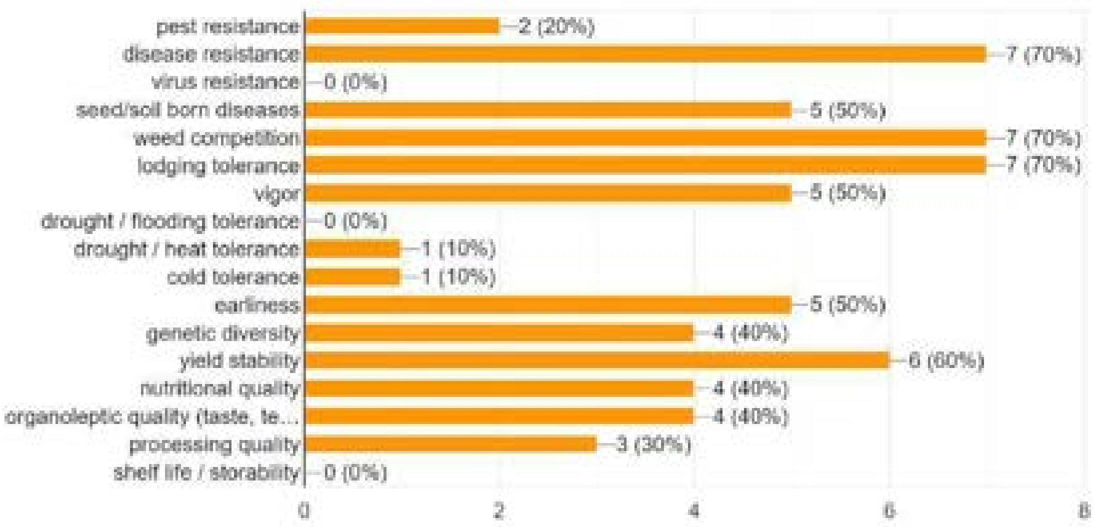

Figure 13 Main breeding goals selected by the partners

Main breeding goals selected by the partners with a maximum of 70% of were mainly related with disease resistance, weed competition and lodging tolerance.

Question 15: What is you approach for breeding for diversity?
(Please select from: breeding of neglected crop species; increase genetic diversity within species (e.g., selection for local adaptation, special usage, wide crosses); breeding of genetic diversity within cultivars (i.e., Organic Heterogeneous material like CCP, open pollinated populations); breeding for cultivar mixtures / multi-lines mixtures; breeding for species mixtures; breeding for agroforestry / permaculture / complex species mixtures; Other)

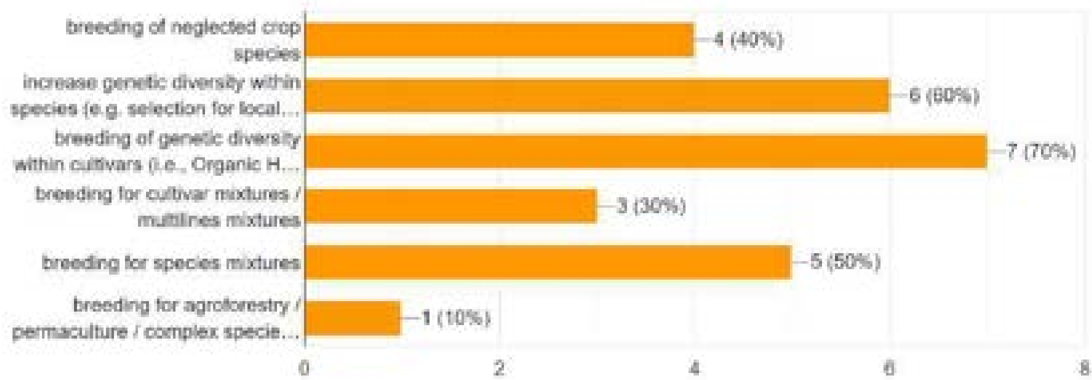

Figure 14. Approach for breeding for diversity at partners

The approach for breeding for diversity followed by the respondents were 70% breeding of genetic diversity within cultivars (i.e., Organic Heterogeneous material like CCP, open pollinated populations)

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followed by increase genetic diversity within species (e.g., selection for local adaptation, special usage, wide crosses) with 60%.

Question 16-17: Who is involved in the multi-actor breeding?

For the question “Who is involved in the multi-actor breeding”, researchers had the higher score for all the questions except for seed production, therefor they play a key role. Farmers are in most cases also involved in many of the steps, in particular defining the breeding goals, in selection and in cultivar trials. Field visits and workshops are the main platforms where all stakeholders engage with each other and the trials. Depending on the set-up, public breeders are also included in most of the steps. Millers and bakers are mostly involved in the definition of breeding goals and in processing quality. Interestingly, dissemination and facilitation mostly fall under the responsibility of the researchers.

	Researcher	Public breeder	Private breeder	Seed company	Farmer	Advisor	Miller	Baker	Other processors	Retailer	Chief / cook	Students/volunteers	NGO	Public authorities
Definition of breeding goals	10	1	1	1	9	3	4	1	0	0	0	0	1	0
Collection of germplasm	9	1	0	0	0	0	0	0	0	0	0	0	0	0
Conduction of crosses	7	1	0	0	1	0	0	0	0	0	0	0	0	0
Selection for agronomic performance of early generations	6	1	0	0	2	0	0	0	0	0	0	0	0	0
Selection of late generations	6	1	1	1	4	1	0	0	0	0	0	0	0	0
Resistance screening to pest and diseases	7	1	1	0	3	0	0	0	0	0	0	1	0	0
Resistance screening to abiotic stress	5	1	0	0	2	0	0	0	0	0	0	1	0	0
Nutritional quality analysis	7	1	0	0	0	0	1	0	0	0	0	0	0	0
Processing quality	4	0	0	0	0	0	4	4	0	0	0	0	0	0
Organoleptic quality	4	1	0	0	2	1	1	0	0	0	0	2	0	0
Maintenance breeding	5	2	1	0	3	0	0	0	0	0	0	0	0	0
Consumer studies	5	0	0	1	0	0	1	1	0	0	0	0	0	0
Statistical analysis	9	1	0	0	0	0	0	0	0	0	0	2	0	0
Final selection	6	1	1	1	4	0	2	1	0	0	0	0	0	0
Registration	4	2	1	0	1	0	0	0	0	0	0	0	0	0
Cultivar trials	8	1	1	1	6	0	0	0	0	0	0	1	0	0
Seed production	3	1	0	2	5	0	0	0	0	0	0	0	0	0
Field visits	9	3	3	4	8	6	4	2	2	3	1	3	2	3
Workshops	8	2	3	3	8	5	4	3	2	1	1	2	1	2
Dissemination	8	3	2	1	2	3	1	2	0	1	0	2	1	0
Facilitation of multi-actor processes	9	1	0	0	0	2	0	0	0	0	0	0	0	0

Table 2 Involvement of actors in different aspects of breeding

Question 18: What is the capacity building and perspective (expected impact of the LIVESEED project)?

(Select all that applies: Collaboration with other breeding initiative has improved to some extent; Collaboration with other breeding initiative has improved to considerable extent; Knowledge of involved actors has improved to some extent; Knowledge of involved actors has improved to considerable extent; Efficiency of breeding project has improved to some extent; Efficiency of

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breeding project has improved to considerable extent; Future of the breeding project after LIVESEED is secured to some extent; Future of the breeding project after LIVESEED is secured by involved actors)

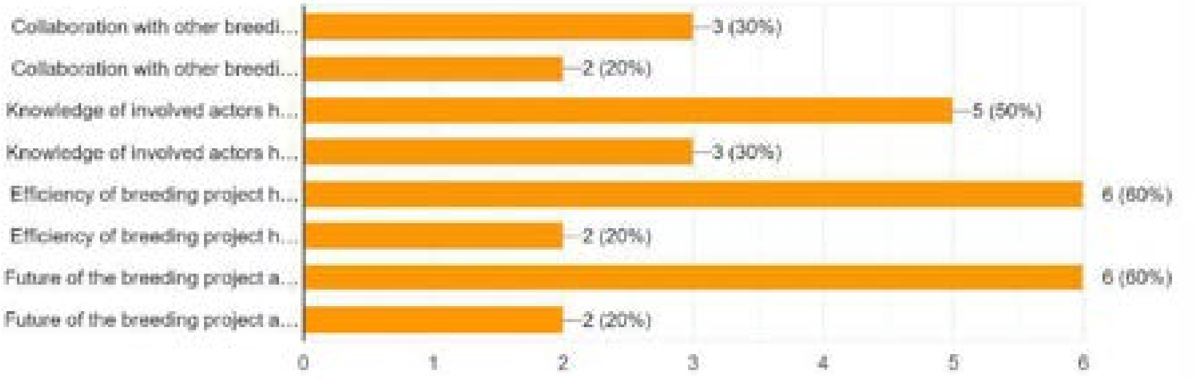

Figure 15: Capacity building results and perspective of partners of the LIVESEED trials

Question 19: Key Results

(Please select from Screening / Selection tools (e.g., for resistance, organoleptic quality); Methodological tools (e.g., efficient design for breeding for mixtures); Statistical tools for data analysis (e.g., model to determine mixing ability); Innovative Cultivars available to farmers after LIVESEED is finalized)

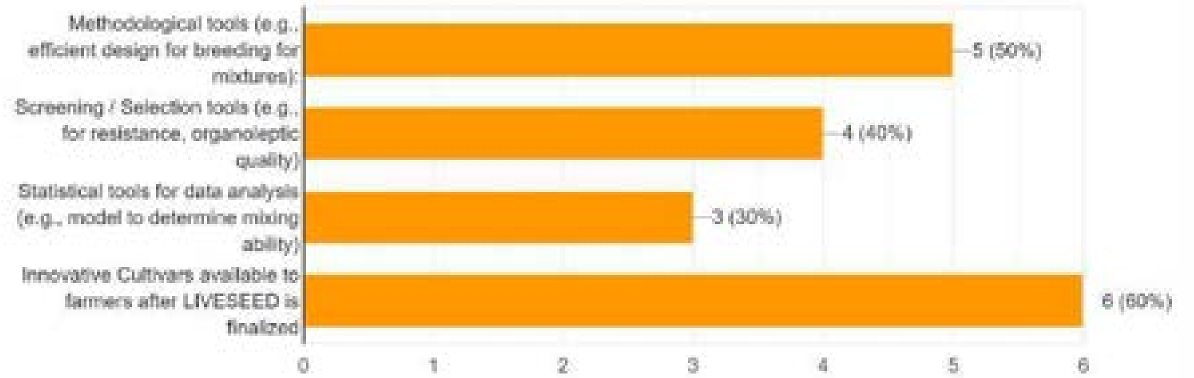

Figure 16: Summary of Key Results expected from LIVESEED at partners

Question 20: Key Results - Methodological tools (e.g., efficient design for breeding for mixtures):

(Please select list details in bullet points with short description)

Institute	Methodological tools
FIBL-CH	Methodology for artificial inoculation of lupin in the field
IPC	Promote CCPs and their selection in the following year to promote
GZPK	Summer barley was shown as a suitable mixing partner for summer peas with similar ripening times. It was more suitable than summer triticale.
LBI	Efficient design for cultivar trials on crop mixtures between and within species
LBI	The design didn't become better than expected before, but the demand on how to go on related to select also for the mixture-partner triticale shined up.
CULTIVARI	We had simple experimental design. After getting acquainted with the results and experience of other colleagues, the methodology would be changed in further studies
AREI	Methodology for artificial inoculation of lupin in the field

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Question 21: Key Results - Screening / Selection tools (e.g., for resistance, organoleptic quality)
(Please select list details in bullet points with short description)

Institute	Screening tool
FIBL-CH	Screening tool for anthracnose resistance at seedling stage, qPCR to quantify Colletotrichum in the seed
IPC	Selection for maize in fresh - several varieties were cold stored and tested later for organoleptic quality; PPB trials were done and farmers, and other actors did evaluate the varieties performance in the field; Maize bread tests are being done using the several varieties available
ITAB	Results to help famers choose between CCP and PopDyns for cereals
LBI	Tools for weed competition, lodging resistance, baking ability
LBI	It was not part of the project to develop new tools.
AREI	Screening tool for anthracnose resistance at seedling stage, qPCR to quantify Colletotrichum in the seed

Question 22: Key Results - Statistical tools for data analysis (e.g., model to determine mixing ability)
(Please select list details in bullet points with short description)

Institute	Statistical tools
IPC	Determine the best statistical tools in several workshops Determine the best traits for selection using MARS, RF, and CART
LBI	Not enough trials to develop a more precise model for selection without doing selection itself. Only the needed morphological type could be described better.
CULTIVARI	I have improved my knowledge in data analysis. In the future I am going to work on pea mixing ability in breeding program for organic as well as for conventional farming

Question 23: Key Results - Innovative Cultivars available to farmers after LIVESEED is finalized
(List species, number of cultivars, special characteristics and expected date of release)

Institute	Breeding goal
FIBL-CH	3-4 advanced breeding lines of white lupin with improved anthracnose resistance and low alkaloid content
IPC	Maize (5, adapted to human consumption as green maize or maize for maize bread, 2022) and Beans (3, different colours and taste, 2023)
ITAB	DOP populations
UBIOS	CCP and some "lines" from the different crosses of the CCP (about 47) are available after LIVESEED
GZPK	First crossings were conducted 2019 and 2020, therefore, it will take time for cultivars to be available. However, important traits were included in the breeding program.
LBI	A new two-line winter pea variety shall be released, if national seed office will agree in special testing also mixed cropping (pea-triticale) and mixed line (Normal/Tendril-2-Line-Mixture).
AREI	3-4 advanced breeding lines of white lupin with improved anthracnose resistance and low alkaloid content

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