USER MANUAL
Crop cultivar mixtures to enhance insect biocontrol

The overall objective of EcoStack is to develop and support ecologically, economically and socially sustainable crop production via enhancement of ecosystem services provision and protection of functional biodiversity.

Practice principles

The agroecological practice of cultivar mixtures, an intraspecific diversification of the cropping system, increases the genetic diversity of the crop and can provide functional diversity that limits the spread of fungal diseases and pests. The mix of cultivars includes varieties with different abilities in terms of disease and pest resistance, weed competition, and can provide an interesting alternative to stabilize yields. It is important to understand the mechanisms behind the effects of cultivar mixtures, as herbivore response to each mixture component must be considered.

Research context

EcoStack researchers investigated the impact of cultivar mixtures on natural enemies of cereal aphids and analysed whether cultivar mixtures can contribute to crop resilience against/resistance to aphids. Two-way combinations of different cereal cultivars were tested in laboratory experiments. The variety pairs with lower rate of aphid setting on the host plant were then tested in field experiments between 2019 and 2021 in different countries belonging to different pedoclimatic regions (Finland, Sweden, UK, Spain, Serbia, Bulgaria and Bosnia and Herzegovina) to investigate the impact on crop infestation by aphid.

Main results

- Plant volatile organic compounds (VOCs) play an important role in plant-plant interactions. A reduction in aphid infestations was observed in field trials, but the effects on aphid abundance were specific to the cultivar mixture.
- The effects of cultivar mixture are highly dependent on climatic factors, as abiotic conditions can weaken plants, impact their physiology and therefore influence the effect of cultivar mixtures on pest infestation.
- It became apparent that aphid suppressive mixtures are highly cultivar specific and it is necessary to find suitable combinations for different countries, climatic conditions and newly bred cultivars.

Information on crop production protocol

To be successful, we propose some guidance on desirable associations of cultivars and on optimized diversification strategies.

Machinery and cultivars example

No specific machinery is needed to implement this practice as it is a mixture of two or more cultivars of the same crop, the same seed drill can be used to sow the crop in fields.

Nevertheless, work organization for sowing is different, it would need to take some time before sowing to mix cultivars seeds together. For large quantities of seeds, space is needed to mix the seeds and store them for a few weeks until sowing.

Different cereal cultivars were used in the EcoStack experiments, Table 1 provides an overview for the respective countries.

Table 1 : EcoStack fields experiments specifications, bolded text indicates desirable cultivar combination for mixture per country, from EcoStack 2023

Country	Crop	Cultivar 1	Cultivar 2
Sweden	Barley	**Salome**	**Fairytale**
Sweden	Barley	**Fairytale**	**Anakin**
Sweden	Barley	Salome	Anakin
Finland	Barley	**Alvari**	**Vertti**
Finland	Barley	Alvari	Toria
Finland	Barley	Toria	Vertti
Bosnia and Herzegovina	Barley	**Matej**	**Jaran**
Bosnia and Herzegovina	Barley	Matej	Scarlet
Bosnia and Herzegovina	Barley	Jaran	Scarlet
Storbritannien	Wheat	Claire	Hereward
Storbritannien	Wheat	Claire	Xi19
Storbritannien	Wheat	Hereward	Xi19
Spanien	Wheat	**Florence Aurora**	**Forment**
Spanien	Wheat	Florence Aurora	Montcada
Spanien	Wheat	Montcada	Forment
Serbia and Bosnia and Herzeg.	Wheat	**NS40S**	**NS Rani otkos**
Serbia and Bosnien och Herceg.	Wheat	Simonida	NS40S
Serbia and Bosnien och Herceg.	Wheat	Simonida	NS Rani otkos
Bulgaria	Wheat	**Apolon**	**Lazarka**
Bulgaria	Wheat	Enola	Apolon
Bulgaria	Wheat	Enola	Lazarka

Crop management

Cereal sowing

Harvest

Mixing seeds of cultivars

Picture 1 : Cultivar mixtures in barley field in France. (Photo ISARA 2021)

Points to pay attention to

Different cultivars can have different growing season length, resulting in late or early harvesting.
At harvest, all cultivars must be mature to avoid yield loss or quality deterioration.

Mixing different cultivars must match the required outlet/sales quality. For example, cultivar mixtures for malting barley must exclude feed barley cultivars to ensure sales in the malting barley market.

Overview of the practice

Researchers at the Julius Kuehn Institute (Germany) gathered information to systematically identify the socioeconomic impacts and potential costs and benefits of EcoStack strategies. The description of expected impacts is based on a literature and data review, as well as interviews with EcoStack researchers about their field trials and expected outcomes. Here are results for the measure “cultivar mixtures”:

++ At the field scale, cultivar mixtures are expected to have strong positive effects on crop production, especially on weed and diseases control. At farm scale, better pest control can reduce pesticide use.

\+ Cultivar mixtures can have slightly positive effects on other crop performance parameters such as yield stability under biotic and abiotic stresses and insect pest control. Reducing pesticide use through better pest management can have a positive impact, on farmer health and overall pesticide costs at the farm level and provide better water protection at the ecosystem scale.

= The organization of work is modified by the use of cultivar mixtures. Time is needed for mixing the cultivars before sowing, but this might be compensated by reducing the time for weed or disease control. In addition, no specific investment in machinery is needed to implement this practice on farms.

– Some aspect can be slightly negatively affected by using cultivar mixtures. The seed costs may become more expensive due to the use of multiple varieties and the marketing of mixtures might be limited.

⛔ No strong negative effects are expected from this practice.

When taking into account the field, farm and regional scale to evaluate cultivar mixtures practices, the overall ranking is mostly positive due to the expected positive impact on pests control, no need for investment in machinery and fewer changes in work organization.

Reference : EcoStack Deliverable *WP4.1 “Guidance on crop cultivar mixture to enhance insect biocontrol”, 2023*

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EcoStack

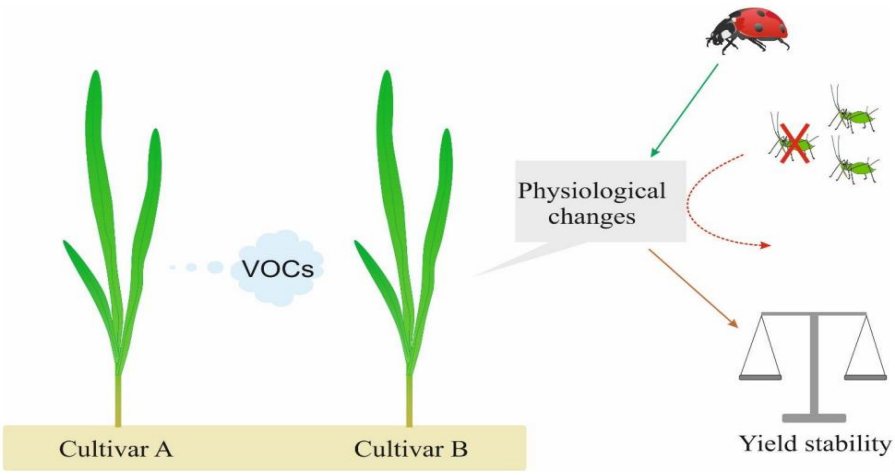
Crop cultivar mixtures to enhance insect biocontrol: implementation, monitoring and improvement

The overall objective of EcoStack is to develop and support ecologically, economically and socially sustainable crop production via enhancement of ecosystem services provision and protection of functional biodiversity.

Context

High crop yields are a principal objective of the current and conventional agriculture through hybridization and selection of plants. But simplified cropping systems are associated with negative impact on environment and biodiversity. The agroecological practice of cultivar mixtures, an intraspecific diversification of the system, increase the genetic diversity of the crop and can provide functional diversity that limits fungal disease and pest colonisation. The mix of cultivars include varieties with different abilities in terms of weed competition, disease and pest resistance, and can represent an interesting alternative to stabilize yields.

But knowledge of the processes underlying differential pest responses is required to better understand the consequences on yields and agrobiodiversity. Ecostack research was focused on the interaction of the plants with each other that effect aphid settling. It was important to grasp the mechanisms behind effects of cultivar mixtures because of the necessity of measuring herbivore response to individual mixture components. Little research has been done so far on the effects of intraspecific diversity on arthropod pests and natural enemies especially on wheat pest control. One of the mechanisms that can contribute to reduce the presence of pest insects in cultivar mixtures is the induced resistance mediated by plant volatiles. The protection is based on the stimulation of defence mechanisms and metabolic changes that prime neighbouring plants against insect attack.



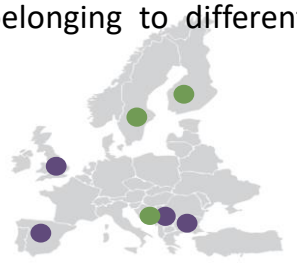
Aphids are the main food source of ladybird beetles. Most consuming are the larvae of ladybird beetles, further lacewing and hoverfly larvae are effective predators of aphids.

Summary diagram (Dimitrije Markovic (2023)

Approach and field studies

Field studies investigated the impact of cultivar mixtures on arthropod pest and natural enemies of cereal aphids in analysing whether cultivars mixture can be used as cultivation method that contributes in the field to resilience against aphids and may affect occurrence of aphid natural enemies such as ladybirds. Small field plot trials with cultivar mixtures in barley and wheat were conducted from 2019-2021 in different European countries (Finland, Sweden, UK, Spain, Serbia, Bulgaria, Bosnia and Herzegovina) with locations belonging to different pedoclimatic regions.

In each country, three regional relevant cultivars were selected. Two-way cultivar combinations that prevent aphid settling on cereal plants were identified in laboratory experiments. Based on this screening, cultivar mixtures were investigated regarding their impact on cereal aphid Abundance in the field trials.



with barley
with winter wheat

Country	Year	Crop	Cultivar 1	Cultivar 2	Cultivar 3
Sweden	2019	Barley	Salome	Fairytale	Anakin
Sweden	2020	Barley	Salome	Fairytale	Anakin
Sweden	2021	Barley	Salome	Fairytale	Anakin
Finland	2020	Barley	Alvari	Toria	Vertti
Finland	2021	Barley	Alvari	Toria	Vertti
UK	2019-20	Wheat	Claire	Hereward	Xi19
UK	2020-21	Wheat	Claire	Hereward	Xi19
Spain	2019-20	Wheat	Florence Aurora	Montcada	Forment
Spain	2020-21	Wheat	Florence Aurora	Montcada	Forment
Serbia	2019-20	Wheat	Simonida	NS40S	NS Rani otkos
Serbia	2019-20	Wheat	Simonida	NS40S	NS Rani otkos
Bulgaria	2019-20	Wheat	Enola	Apolon	Lazarka
Bulgaria	2020-21	Wheat	Enola	Apolon	Lazarka
Bosnia and Herzeg.	2019	Barley	Matej	Jaran	Scarlet
Bosnia and Herzeg.	2019-20	Wheat	Simonida	NS40S	NS Rani otkos
Bosnia and Herzeg.	2021	Barley	Matej	Jaran	Scarlet

Table 1: Small scale field experiments specifications per country and study year (from the report D4,1 Velemir and al, EcoStack, 2023)

In Spain, the selection of non-cereal species (under-sowing species) was based on farmers local knowledge and functional traits of species. The selected cultivars were exposed to the volatiles of each other in pairwise combinations, followed by assessment of aphid colonisation of the plants after receiving VOCs from the other cultivar.

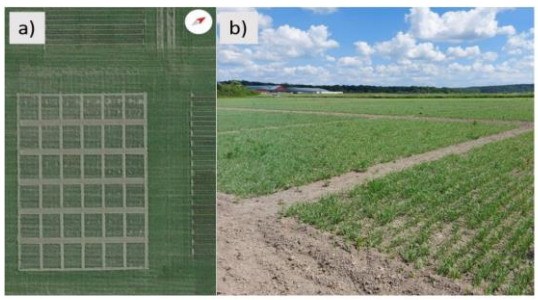


Figure 1: Field side in Sweden (from the report D4.1 Velemir and al, ECOSTACK, 2023)
a) Field plot design from the air, b) barley plants growing on the experimental field.

Key findings

1- In the field, the effect on aphids varies with great differences between the years. Most countries experienced high and dry temperatures.

2- Specific combinations were able to reduce aphid infestations in field trials but the effects on aphid abundance are very cultivar mixture specific.

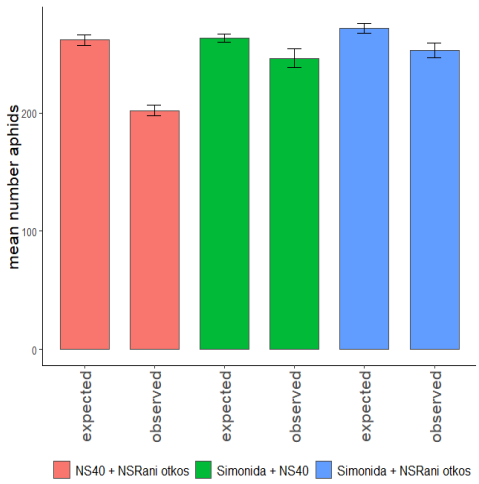
3- An impact of the climatic factors might be antagonistic to the positive effects of cultivar mixtures on pest suppression. As high and dry temperature during the field studies may weaken the plants and impact their physiology.

4- Laboratory and field studies revealed a high specificity of mixture effects, highlighting the importance of finding the right cultivar combinations for mixtures instead of simply increasing general genetic diversity to promote aphid resistance.

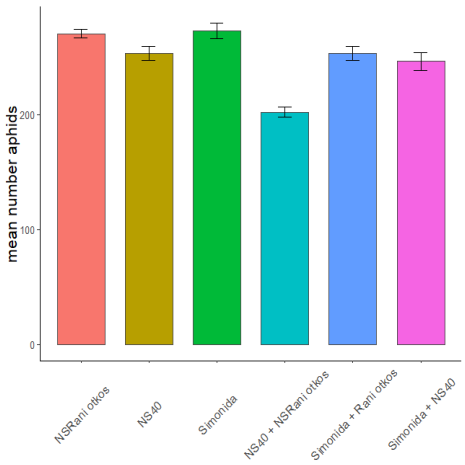
5- Plant volatile organic compounds (VOCs) play an important role in plant-plant interactions in the cultivar mixtures.

6- Volatile mediated interactions between plants can reduce the host acceptance (settling) of aphids on cereal plants and thus increase the plant resistance. The impact on aphids is clearly cultivar and mixture specific.

Example from winter wheat in Bosnia and Herzegovina to illustrate the effect of the mixture as well as highlighting the specificity of the mixture effect:



(from the report D4,1, 2023)



We found a significant reduction in NS40 + NS Rani otkos.

The graph shows the number of aphids as well as the expected number (as expected from the number of aphids in the monocultures) compared to the actually observed number of aphids in the field.

Conclusion

Lessons learnt and perspectives

- In the field, significant reduction in aphids infestation in cultivar combinations of barley or winter wheat have been identified for some locations. However, also indifferent results were found.
- The aphid attacks was significantly reduced when their abundance was high. However, this phenomenon was not observed in all tested cultivar combinations.
- Some combinations with barley was shown to reduce diseases abundance in some years. In some combinations, we found significant increase in the yield.
- It became apparent that aphid suppressive mixtures are highly cultivar specific and it is necessary to find suitable combinations for different countries, climate conditions and new bred cultivars.
- Cultivar mixtures showed the potential to impact the physiology of plants grown in the mixture and might be beneficial for growers.
- Volatile organic compounds are important mediators of mutualistic interactions between plants and their physical and biological surroundings.
- Investigation of plant VOCs from different cultivars could facilitate the identification of beneficial mixtures based on plant volatiles.
- The disruptive effect of a non-host plant on aphids could be increased by using different spatial arrangement and increasing the non-host plant biomass.

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