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Reducing symptoms of sugar beet yellows with companion plants

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The National Research and Innovation Programme (PNRI) aims to identify operational solutions for managing sugar beet yellows. Several trials have been set up in a network of farmers' plots in northern France to assess the value of companion plants. Three species were tested: spring oat, spring barley and spring faba bean. Populations of Myzus persicae aphids, yellows symptoms and yield losses due to competition between companion plants and sugar beet were assessed. Preliminary results are presented in this article for the three variables studied. A Bayesian statistical model was used to estimate the efficiency of companion plants on aphids when used alone or in combination with an aphicide treatment (flonicamide-based Teppeki®). Companion plants can be used to reduce aphid populations on sugar beet, but they are less effective than an aphicide treatment. Their value in combination with an aphicide treatment has not been demonstrated, but this does not rule out the possibility that they could have value in cases where aphicide protection is insufficient. The efficiency of companion plants on vellows symptoms and yield losses due to competition were calculated for each trial and then evaluated graphically. A reduction in yellows symptoms was observed for all three species, but the results varied widely between trials. Finally, a loss of yield was observed, and this was all the greater the later the companion plants were destroyed. The success of this lever depends on a reduction in the number of green aphids, a reduction in the yellows symptoms and the absence of competition with sugar beet. An adjustment of the technical itinerary is envisaged for future PNRI trials.

Keywords: Competition, Sugar beet, Companion plant, Myzus persicae, Yellow leaf spot

Introduction

Since September 2018, the ban on neonicotinoids (NNI) in seed treatments has posed challenges for several agricultural sectors in managing pests insect. In 2020, the sugar beet industry experienced a decline in yields in part due to yellow disease, a viral disease transmitted by the green peach aphid, *Myzus persicae* (Audran, 2020). This aphid can transmit several viruses responsible for yellow disease : Beet Chlorosis Virus (BChV, polerovirus), Beet Mild Yellowing Virus (BMYV, polerovirus), Beet Yellows Virus (BYV, closterovirus) and Beet Mosaic Virus (BtMV, potyvirus) (Hossain *et al.*, 2021). In response to this situation, a National Research and Innovation Plan (PNRI) was deployed in 2020 to identify operational solutions for sustainable yellow disease management.

The chemical alternatives to NNI used so far are foliar aphicide treatments (Teppeki® based on flonicamide and Movento® based on spirotetramat, the latter being subject to a request for exemption each year). However, these treatments did not provide sufficient control of aphid populations across the whole of the sugar beet regions in 2020. Moreover, these active substances are not sustainable solutions either: there is a risk of resistance developing, and the spirotetramat-based product will be withdrawn from



the market. Alternative solutions are therefore being tested as part of the PNRI, and the use of companion plants during the sensitive stages of the sugar beet cycle is among the most promising.

Companion plants are species grown with the main crop that are not harvested, and which can confer benefits to the main crop. Many studies document their value for weed management (Verret *et al.*, 2017) but they can also be used to manage pests insect and contribute to soil fertility and structure improvement (Ben-Issa *et al.*, 2017; Liebman and Dyck, 1993). In the case of sugar beet, companion plants could limit the presence of aphids on the crop by trapping them, altering crop recognition (chromatic disruption) or attracting their natural enemies (Ben-Issa *et al.*, 2017). For example, Heathcote (1974) showed a reduction in *Myzus persicae* populations in sugar beet in the presence of companion plants (mustard and barley). The companion plants have allowed to mask the sugar beet crop, preventing aphid colonisation upon landing. Other successful cases have been shown for aphids in other crops, for example on white cabbage with marigold (France; Jankowska *et al.*, 2009) and on tobacco with garlic (China ; Lai et *al.*, 2011), but fewer references exist for Western European field crops.

While companion plants have the potential to reduce aphid populations and therefore yellows symptoms on sugar beet, they can also compete with the main crop and lead to significant yield losses (Liebman and Dyck, 1993). Therefore, a number of parameters need to be anticipated in the management of companion plants. The chosen species must easily integrate into the sugar beet's technical management (e.g. sugar beet herbicide), be suitable for the climatic conditions of the northern regions of France. Grasses are interesting species, as they are less sensitive to frost and their management is compatible with sugar beet management. Legumes could be an interesting alternative in areas heavily infested by grass weeds, and would be less competitive than grasses due to their ability to fix atmospheric nitrogen (Verret *et al.*, 2017). The type of sowing in the fields, seeding density and timing of companion plant destruction are also parameters to be considered in the crop management. Too high seeding density or too late destruction of companion plant could lead to a significant yield loss.

Objectives

The first objective of this study is to evaluate the effect of companion plants on *Myzus persicae* populations and yellows symptoms in sugar beet crops. The efficiency of the lever will be assessed alone and in combination with aphicide protection to determine the value of companion plants compared to existing protection strategies used by farmers. The second objective is to identify a crop management that limits yield losses due to competition. Three companion plant species are being worked on: spring oat, spring barley and spring faba bean.

To assess the benefits of companion plants, trials have been set up on a network of farmers' plots located in northern France, over two years of experimentation (2021 and 2022). The experimental design involves alternating strips of sugar beet with and without companion plants. Companion plants populations are described from emergence to destruction. Aphid populations are monitored during the sugar beet's sensitive period, and yellows symptoms are assessed at the end of the crop cycle.

Methods

A network of farmers' plots

The trials were set up in 2021 and 2022, on farmers' plots in the Fermes Pilotes d'Expérimentation (FPE) network created in 2021 as part of the PNRI. This network is coordinated and monitored by the ITB and the Agronomic Services of Sugar Companies (Tereos, Cristal Union, Saint Louis Sucre). The plots are



spread across the main sugar beet regions in France to evaluate the efficiency of the levers tested in diverse situations. 86% of the plots are managed under conventional farming, and 14% under organic farming.

Among the trials, some were excluded from the analysis due to low companion plant populations (seeds batch with poor germination quality for oat, impact of herbicides or frost for faba bean). The absence of aphids also led to the exclusion of some trials. Thus, the results presented in this article include 54 trials. They were set up in 47 plots within the FPE network, 3 plots in agricultural schools involved in the PNRI, and 4 plots outside the network (Figure 1).

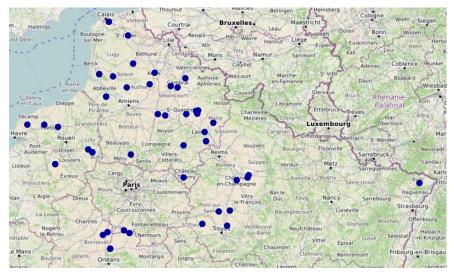


Figure 1 Location of the 54 plots used to analyse the effect of companion plants on sugar beet yellows in 2021 and 2022. A blue dot corresponds to a municipality close to one or more plots in the network - Geobatch_v1.5

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Sowing and destruction of companion plants

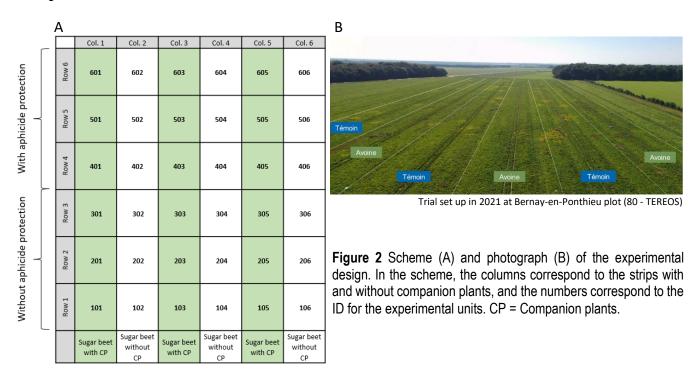
The companion plants were sown at the same time as sugar beet sowing, or 2 or 3 weeks before to promote their development when the aphids arrive. The target seeding density for grasses was 100 grains/m² in 2021 and 75 grains/m² in 2022. It was reduced in 2022 because a loss of sugar beet yield due to competition from grasses was observed in 2021. Faba bean were sown at a density of 20 grains/m² in 2021 and 2022. Grasses destruction was carried out from the 6-leaf stage of sugar beets, while faba bean was done at the 8- to 10-leaf stage.

Experimental design

The experimental design to assess the effect of companion plants on aphids and yellow disease is a stripplot design, with alternating strips of sugar beets with and without companion plants. In the scheme in Figure 2A, the strips correspond to the columns. This non-randomised design was chosen to work on large areas and to simplify the sowing of companion plants by the farmer. This limitation is compensated by the large number of trials conducted. Each strip is at least 12 m wide and 30 m long. In most of the trials, the length of the strips is usually the entire length of the plot, as shown in the photo in Figure 2B. A companion plant species may be tested on one or more strips in a given trial. Moreover, the three species are not systematically tested in a trial. The choice of species tested depended on the context of the plot and the farmer's interest in one or more of the proposed species.



In some plots, the experimental design was duplicated to assess the effect of companion plants in combination with aphicide protection, evaluating the value of the lever in a protection strategy commonly used by farmers (Figure 2A). The maximum aphicide protection involved one treatment of Teppeki® (flonicamid at 0.14 kg/ha), followed by two treatments of Movento® (spirotetramat at 0.45 L/ha) with an estimated residual effect of 10 to 14 days. However, the program was adapted to the aphid populations in each trial and could be reduced in the case of low populations, where a single aphicide was sufficient. The first aphicide treatment was made when the infestation level reached 10% of sugar beets being colonised by wingless aphids. Subsequent treatments were made when the threshold was exceeded again.



Monitoring

The monitoring period extends from sugar beet emergence to harvest.

Sugar beet and companion plant populations

At the 2 to 4-leaf stage, the number of sugar beets is counted on 4 rows x 10 m to describe the population and note any heterogeneity in the emergence across the trial. The number of companion plants per square metre was also measured using quadrats.

At each observation date, the stage of the sugar beets and companion plants is noted for each object (1 value per object). An object corresponds to a cross between two modalities (e.g. With companion plant X Without aphicide protection, Without companion plant X With aphicide protection, etc.).

Aphid populations

In each experimental unit, green aphids (*Myzus persicae*), black aphids (*Aphis fabae*), wingless and winged aphids were counted on 2 rows x 5 beets (i.e. 30 beets per object). The counts start when the first green aphid is observed in the field, then every 15 days, until 15 days after the destruction of companion plants (8 to 12-leaf stage of the sugar beets, depending on the trial). Observations were made in the center of each experimental unit (Figure 2), at least 20 m away from the edges of the field.



Yellows symptoms

Before harvest, yellows symptoms are measured using a visual severity rating scale, supplemented by drone photography: 0 - 0% of the area affected; 1 - 1 to 10% of the area affected; 2 - around 20% of the area affected; ...; 10 - 100% of the area affected. At each observation, a severity score for yellows symptoms is recorded for each object (1 value per object). In some cases, serological tests (ELISA) are carried out to confirm the diagnosis.

Sugar beet yields

To assess the impact of companion plants on sugar beet yield, sugar beets without visible yellows symptoms were sampled from areas with and without companion plants, for each companion plant species tested in each trial. A sample corresponds to 4 rows of sugar beets over 4 m, and 3 to 4 samples (replicates) spaced 20 m apart were taken in the two targeted areas. Productivity (t/ha) and sugar content (%) were then measured on a dedicated platform.

Data analysis

Efficiency on wingless green aphids

To accurately estimate the efficiency of companion plants, combined with or without aphicide protection, a hierarchical Bayesian statistical model was fitted to the data, taking into account the variability of treatment effects between site-years and temporal evolution of the number of green aphids. The model is based on the one used by Laurent *et al* (2023) to evaluate the efficiency of different aphicide treatments against aphids. It was adapted to assess the effect of companion plants, and the results of 36 factorial trials evaluating the efficacy of Teppeki® (method and results presented in Laurent *et al.*, 2023) were added to complete the database. The statistical model estimates the efficiency of each treatment (companion plant and/or aphicide treatment), with statistical replications corresponding to the experimental units, and the trial was considered as a random effect.

Efficiency on yellows symptoms

The efficiency of companion plants on yellows symptoms was calculated for each trial, and a boxplot was made to summarise all the trials. Efficiency is a percentage and corresponds to the relative difference in yellows symptoms intensity observed on sugar beets with companion plants (score_yellow_CP) compared to beets without companion plants (score_yellow_control): Efficiency (%) = (1 - (score_yellow_CP / score_yellow_control)) x 100. The efficiency of companion plants on yellows symptoms was first calculated without aphicide protection, then with aphicide protection.

Impact of competition on sugar beet yields

The yield loss due to companion plants was calculated for each trial, and a boxplot was made to summarise all the trials. Yield loss is a percentage and corresponds to the relative difference in yield between sugar beets with companion plants (yield_PC) compared with sugar beets without companion plants (yield_control): Yield loss (%) = $(1 - (yield_PC/ yield_control)) \times 100$. This yield loss was calculated for different companion plant destruction periods to determine which one allows the least impact on yield.



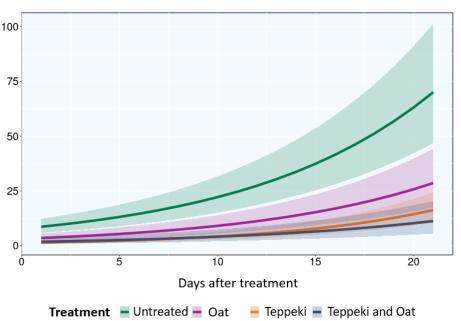
Results

Companion plant populations

The growth of oat, barley and faba bean has been affected by several factors over the two years of trials: frost, poor climate conditions (low temperatures, especially in 2021), and some herbicides used in sugar beet management. Grasses were not much affected by herbicides, although barley was more sensitive than oat, and was more puny. Faba bean were mainly affected by herbicides, especially by the triflusulfuron-methyl molecule, but were rarely totally destroyed. In 2022, some of the oat sown had poor germination quality (seed batches with weevil damage), leading to lower populations than expected in half of the trials for that year.

Efficiency on wingless green aphids

Statistical analysis was carried out on 52 trials where monitoring of aphids was carried out (out of 54 trials). Below, we present the results for the aphicide treatment (Teppeki[®]), for oat as a companion plant, and for the combination of both. Companion plants and aphicide treatment have different action on aphids, therefore the efficiency was compared 14 days after the aphicide treatment to assess a situation where the additional value of companion plants could be observed due to the limited persistence of the aphicide treatment. If no aphicide was applied on the trial, the fictitious treatment date t0 is defined as the average of the treatment dates in all trials including an aphicide treatment.



Prediction of the number of aphids (for 10 sugar beets)

Figure 3 Dynamics of wingless green aphids for several treatments: oat as a companion plant (n = 30), aphicide treatment (Teppeki®, n = 53) and the combination of both (n = 11), compared with the untreated control (n = 74). Aphid dynamics are assessed in terms of the number of days after the aphicide treatment in order to compare the efficiency of the treatments.

At 14 days after application, the aphicide treatment alone allows an aphid reduction of 80.5% [95% confidence interval = (74.5; 85.4)]. For oat as a companion plant (without aphicide treatment), the results show an aphid reduction of 45.5% [95% confidence interval = (22.6; 63.4)] (**Erreur ! Source du renvoi introuvable.**). Faba bean and barley were slightly less effective companion plants than oat, with 43.2 and 39.4% of aphid reduction respectively (with a higher level of uncertainty due to the smaller number of



trials). A slightly higher level of control was obtained by combining the aphicide treatment (Teppeki®) with oat as a companion plant. The combination show an aphid reduction of 82.5% at 14 days [95% confidence interval = (68.5; 91.5)]. However, this result remains similar to that obtained with the Teppeki® treatment alone (**Erreur ! Source du renvoi introuvable.**). Finally, the number of aphids observed over the monitoring period is acceptable for Teppeki® alone (and the combination with oat), but too high for oat alone at 14 days and beyond.

Efficiency on yellows symptoms

The efficiency of companion plants on yellows symptoms was assessed on the 34 trials where yellows symptoms were observed in untreated control. Visually, differences in yellows symptoms can be observed in several trials (an example in Figure 4A). Grasses allow an average reduction of yellows symptoms of 34% for oat and 31% for barley. Faba bean allows an average reduction of 19% (Figure 4B). Companion plants alone are not sufficient to reduce yellows symptoms, as symptoms are still present, but they sometimes show an important reduction.

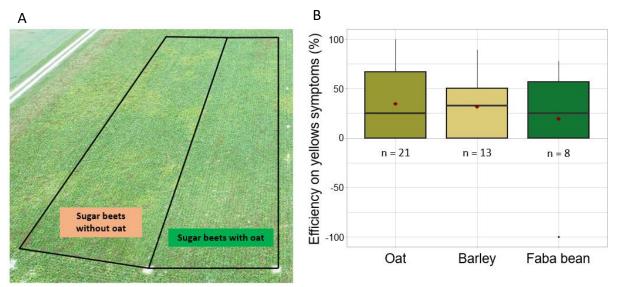


Figure 4: Efficiency of companion plants on yellows symptoms without aphicide treatment. Drone photograph of the ITB trial at Somme-Vesle (51) in 2022, visually showing the difference in yellows symptoms between sugar beets with and without oat as a companion plant (A). On the boxplot (B), red dots show the mean value, and n values correspond to the number of trials per companion plant species.

In the area with one or more aphicide treatments, the three species of companion plant allow an additional reduction in yellows symptoms in some trials (6 trials out of 16). In the other ones, aphicide protection was sufficient to control the disease.

Impact of competition on sugar beet yields

The dry year of 2022 is particularly interesting for analysing the competitive effect of companion plants under limited water resources. The climatic conditions were optimal in 2021. Sugar beets without visible yellows symptoms were sampled from 20 trials.

Trials show a loss of yield whatever the companion plant species tested (excepted two trials), which could be more or less important: 2 (minimum) to 39% (maximum) of yield loss compared to sugar beets without companion plant.

In 18 of the 20 trials, the leaf stage of the sugar beets was noted when the companion plants were destroyed (**Erreur ! Source du renvoi introuvable.**). Oat and barley were grouped together as "grasses"



because the results were similar for both species. Highest yield losses observed with grasses, averaging 19%, are explained by a later destruction of grasses, at 8-leaf stage of sugar beets. Lower yield losses, averaging 3%, are observed when companion plants were destroyed at the 6-leaf stage of sugar beets. For faba bean, destruction was carried out at the 12-leaf stage of sugar beets and an average yield loss

of 15% is observed. No effect of companion Grasses Faba bean plants richness on sugar n = 8 was 40 observed. n = 5 30 Yield loss (%) 20 n = 6 10 n = 3 0 6-leaf 8-leaf > 8-leaf > 8-leaf

Figure 5 Yield loss according to the leaf stage of sugar beets at the destruction of companion plant species (18 trials). Red dots show the mean value, and n values correspond to the number of trials per companion plant species.

Discussion

Efficiency on wingless green aphids

The value of companion plants was assessed for wingless green aphids (mainly represented by *Myzus persicae*), which are the main vector of yellow disease of sugar beet (Audran, 2020).

Without aphicide treatment

Without aphicide protection, results show a reduction of wingless green aphids populations on sugar beets in the presence of all three companion plant species. Oat provides the highest reduction, but a larger number of trials were carried out with this specie. High heterogeneity in the results were observed, partly explained by heterogeneous populations densities (number of plants/m²) due to oat sown with poor germination quality (seed batches with weevil damage) in 2022. Faba bean are mainly affected by the herbicide program commonly used in sugar beet management.

The experimental design was not intended to address the question of the mechanism explaining the effect of companion plants on aphid behaviour (Döring, 2014). Our hypothesis to explain the mechanism is that the presence of companion plants may alter the recognition of sugar beet by *M. persicae* through a visual and/or olfactory mechanisms. Therefore, the strip-plot design chosen in this study could accentuate the contrast between areas with and without companion plants, overestimating their efficiency on aphids and



yellows symptoms. To minimise this potential effect, future trials will be carried out on minimum 2-hectare areas per modalities tested. Continuing on the topic of mechanisms, it is not excluded that the presence of grasses could reduce aphid growth rate as demonstrated by Davis and Radcliffe (2008). Lastly, companion plants may promote certain beneficial organisms, enhancing their impact on aphids.

With aphicide treatment

Aphicide treatment has shown an efficiency 14 days after treatment. As expected, the efficiency on aphids' populations is higher than companion plants. When aphicide treatment is combined with oat as companion plant, the additive effect of oat is marginal compared with the aphicide treatment. This result supports that in situations where aphicide treatment are sufficient to manage aphids' populations, there is no value to sow companion plants. Although these situations are the majority of those encountered during the last two years of experimentation, the year 2020 showed that this was not always the case, as aphicide treatments were not sufficient to control aphid populations. Furthermore, companion plants could potentially reduce the number of aphicide treatments, which will be explored in future trials using experimental design that will cross companion plants with an increasing number of aphicide treatments (1, 2 and 3 treatments).

Efficiency on yellows symptoms

The value of companion plants was evaluated on yellows symptoms to confirm results observed on aphid populations.

Without aphicide treatment

Without aphicide protection, initial analyses carried out show a reduction in yellows symptoms on sugar beets in the presence of the three companion plant species. Oat and barley show an equivalent reduction in symptoms, while faba bean showed a lower reduction. Variability was observed between trials, but as with aphids, this can be partly explained by heterogeneous companion plant populations and levels of development. Moreover, the link between aphid populations in the field and the intensity of yellows symptoms is generally not linear, depending on the virulence of the aphids (Werf *et al.*, 1992).

With aphicide treatment

Few situations show an additional value of companion plants in addition to aphicide treatment, which is sufficient in the majority of trials carried out. This result for yellows is consistent with what was observed previously for aphids.

Impact of competition on sugar beet yields

Competition can occur between companion plants and sugar beets. The impact on sugar beet yield must therefore be assessed to evaluate the overall value of companion plants and to adapt their management.

Yield losses are observed in most trials for the three companion plant species, and they are higher if companion plants were destroyed late. An earlier destruction can limit the impact on sugar beet yields (**Erreur ! Source du renvoi introuvable.**), therefore a destruction at the 4 to 6-leaf stage of sugar beets will be considered in future trials. Microplot trials comparing different companion plant management (species, seeding densities, destruction stages) will be carry out to optimise the technical management more precisely. In addition, trials involving mechanical destruction of companion plants on organic farms will be carried out in 2023 to assess feasibility.



Conclusion

Companion plant lever must reconcile a reduction in the number of wingless green aphids, a reduction in yellows symptoms and an absence of competition with sugar beets. These two years of trials have shown that companion plants (without aphicide treatment) can reduce *Myzus persicae* populations by an average of 45.5% for oat, 43.2% for barley and 39.4% for faba bean, with heterogeneous results especially for the latter two species. Yellows symptoms were also reduced by an average of 34% for oat, 31% for barley and 19% for faba bean. However, the value of companion plants in an overall protection strategy including aphicide treatment has not yet been demonstrated, as aphicide treatment alone has been sufficient in the majority of trials carried out where moderate aphid populations (and yellows symptoms) were observed over the two years. Lastly, yield loss due to competition between companion plants and sugar beets can be high, averaging 19% for grasses when destruction is late. Less risky practices are possible to limit competition, and they should be assessed on yield, aphids and yellow disease. In the PNRI's work, companion plants are combined with other levers such as biocontrol products (paraffin oil, entomopathogenic fungi), flower strips or releases of beneficial insects (lacewings) to assess the value of management solutions against yellow disease that could save aphicide treatments.

Ethics

The authors declare that the experiments were carried out in compliance with the applicable national regulations.

Declaration on the availability of data and models

The data supporting the results presented in this article are available on request from the author of the article.

Declaration on Generative Artificial Intelligence and Artificial Intelligence Assisted Technologies in the Drafting Process.

The authors have used artificial intelligence-assisted technologies to translate from French to English.

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