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Can above-ground ecosystem services compensate for reduced fertilizer input and soil organic matter in annual crops?

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Running title:

Above-ground and below-ground effects on yield

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Summary

1. Above-ground and below-ground environmental conditions influence crop yield by pollination, pest pressure, and resource supply. However, little is known about how interactions between these factors contribute to yield. Here, we used oilseed rape *Brassica napus* to test their effects on crop yield.

2. We exposed potted plants to all combinations of high and low levels of soil organic matter (SOM) and fertilizer supply, and placed all treatments at a variety of field sites representing a gradient in pollinator visitation rate and pest exposure. We determined the relative contribution of pollinators and pests, SOM and fertilizer supply to yield. We also tested whether SOM can moderate effects of fertilizer on yield and whether soil conditions influence the relationship between above-ground conditions and yield.

3. Increases in pollinator visitation rate and decreases in pest pressure enhanced yield more than increase of fertilizer supply. Although higher SOM content resulted in plants with more biomass and flowers, under our experimental conditions SOM neither enhanced yield, nor influenced effects of fertilizer, pollinators or pests on yield.

4. The relationships between yield, pollinator visitation rate and pest pressure did not depend on the level of fertilization suggesting that the effects of fertilizer application and above-ground (dis)services on yield were additive. In contrast, pollinator visitation rate was more strongly related to yield at low pest pressure than at high pest pressure indicating trade-offs between above-ground services and disservices.

5. *Synthesis and applications.* Our results show that it is possible to increase oilseed rape yield by enhancing pollination, irrespective of supplying mineral fertilizer. Moreover, the fact that below-ground conditions did not alter the effect of above-ground conditions, suggests that farmers may obtain even higher yields by maximizing both above-ground ecosystem services and external inputs. Further studies are needed to understand at which point the positive relationships between pollinator visitation and yield, as well as between fertilizer and yield will level off. Considering above-ground and below-ground services and inputs in agro-ecosystems in conjunction is crucial in order to optimize external inputs for crop yield from an economic and ecological perspective.

Key-words: agro-ecosystem, *Brassica napus*, crop yield, *Evergestis extimalis*, fertilizer, insect pests, oilseed rape, pollination, rapeseed, soil organic matter

Introduction

Crop plants are exposed to multiple above-ground and below-ground environmental factors that all together determine biomass production and crop yield. These factors operate at a variety of spatial and temporal scales and are under the influence of multiple management strategies. Nutrient status, water availability, and soil organic matter (SOM) are managed mainly by farmers through fertilizer supply, returning organic matter to the soil, and soil tillage. These factors influence each other as well – increased percentage of SOM can reduce dependence on mineral nitrogen fertilizer (Brady *et al.* 2015). Exposure to pests, biocontrol of these pests and pollination are influenced by farmers through applying biocides and putting out bees, but these (dis)services are also provided by the surrounding landscape where pests and their natural enemies (Thies & Tscharntke 1999), as well as pollinating insects (Steffan-Dewenter & Tscharntke 1999) survive periods without crops. Growing crops and management to optimize yield therefore require explicit consideration of both below-ground and above-ground ecosystem properties and services. Relatively little is known about potential trade-offs and synergies between management practices focusing on either above-ground or below-ground subsystems (Tscharntke *et al.* 2012; Bommarco, Kleijn & Potts 2013; Setälä *et al.* 2014). Here, we study possible trade-offs and synergies among SOM content, fertilizer supply, and above-ground influences of pollinators, pests, and pest control organisms as provided by the surrounding landscape.

Above-ground, field conditions may differ in the influx of pests, predators, and pollinators, because the supply of these organisms from surroundings can vary. For example, the quantity of semi-natural habitats near a field is generally positively related to arthropod diversity and subsequently to arthropod-delivered ecosystem services such as pollination and pest control (Steffan-Dewenter *et al.* 2002; Thies, Steffan-Dewenter & Tscharntke 2003; Kennedy *et al.* 2013). In addition, the quality (e.g. flower abundance) of habitats near a field influence (dis)service-providing organisms (Albrecht *et al.* 2007), for example by providing overwintering refuges and alternative food sources for pollinators (Scheper *et al.* 2013), herbivores (Landis, Wratten & Gurr 2000; Veres *et al.* 2013), and their natural enemies (Rusch *et al.* 2013; Sarthou *et al.* 2014). Knowledge of the influences of off-field management on delivery of ecosystem services and disservices is rapidly increasing, but information about how comprehensive management strategies may enhance net effects of all local ecosystem services together is still mostly lacking (Schellhorn *et al.* 2008).

Below-ground, management interventions that influence soil structure and nutrient availability can have contrasting consequences. For example, ploughing and supplying mineral fertilizers may lead to a short-term improvement of soil conditions that enhance yield, but negatively affect soil biota such as earthworms and arbuscular mycorrhizal fungi that generally enhance crop growth by improving soil structure and nutrient availability in the long term (Helgason *et al.* 1998; Crittenden *et al.* 2014). Moreover, long term high input–output agriculture based on intensive soil tillage and nutrients predominantly supplied as mineral fertilizers usually lead to a decrease in SOM (Reeves 1997), which is an important ecosystem property that influences nutrient availability, prevents nutrient leaching and increases soil structure and water-holding capacity (Hendrix, Coleman & Crossley 1992; Six, Elliott & Paustian 2000; Lal 2006). Increasing SOM content is expected to enhance plant growth and reduce dependence on mineral fertilizer (Brady *et al.* 2015).

Nutrient availability in soil may also influence performance of above-ground antagonistic and mutualistic insects (Poveda *et al.* 2003; Otieno *et al.* 2011). However, fertilizer supply may also negatively affect herbivore performance (Mattson 1980). Effects of fertilization on insect performance are indirect: mineral nitrogen (N) fertilization leads to more available N and a lower carbon (C):N ratio (Zhang *et al.* 2012), which alters plant chemistry and growth (Bender & van der Heijden 2015). Indirectly, fertilizer supply may change the nectar content of flowers (Viik *et al.* 2012), the number of flowers (Lau & Stephenson 1993), and plant nutritional quality (Nowak & Komor 2010). Subsequently, all these plant properties are known to attract pollinators (Ohashi & Yahara 1998; Viik *et al.* 2012; Klatt *et al.* 2013) and herbivores (Nowak & Komor 2010), but little is known about the overall effects on yield.

Here, we examined how fertilizer supply interacts with SOM content in affecting the effect of a realistic exposure to pollinators and arthropods (both crop pests and their antagonists) on crop yield. We grew oilseed rape *Brassica napus* plants in pots under two levels of fertilizer supply and SOM. At the onset of flowering, potted plants were placed in fields that differed in the complexity of the surrounding semi-natural habitat, which was used as a proxy for pollinator abundance. After being pollinated, all plants were placed under the same growth conditions in order to examine the net effects of pollinator and pest exposure, fertilizer supply and SOM on above-ground biomass, number of flowers and seed yield. We addressed the following questions: (i) What is the relative importance of SOM, mineral fertilizer, pollinator visitation rate and pest pressure to yield? (ii) Can

SOM, at least partly, replace the effects of mineral fertilizer on yield? (iii) How are the relationships between yield on the one hand and pollination and pest pressure on the other influenced by fertilizer application or SOM content?

Materials and methods

Experimental design

We used oilseed rape *Brassica napus* (variety *Petranova*; a spring-sown variety) as our model crop. Maximum yield of *B. napus* requires insect pollination (Klein *et al.* 2007) and the crop is susceptible to damage from various insect pest species (Valantin-Morison, Meynard & Dore 2007). *Brassica napus* is an important (energy) crop worldwide; in Europe it is even the most abundant oilseed species (Verhoog 2002) and it has expanded in the last decade because of the renewable fuel directive of 2003 (Breeze *et al.* 2014). We grew *B. napus* plants under factorial combinations of two levels of SOM content and two levels of fertilizer addition, and placed four replicates of all four soil treatments in 15 different field sites, resulting in 2 × 2 × 4 × 15 = 240 experimental units. We used landscape complexity and quality as a proxy for pollinator abundance (Albrecht *et al.* 2007; Kennedy *et al.* 2013) and selected the 15 sites so that the experimental plants were positioned in a gradient of pollinator visitation rates (see Appendix S1 in Supporting Information). At four locations, we placed four extra plants (one for each treatment) covered by a gauze net (mesh size: 0.25–0.75 mm) to exclude all insects (pollinators and herbivores), bringing the total to 240 + 16 = 256 experimental units. We used the caged plants to check the level of seed set without insect pollination. Under the cages wind pollination was possible to some extent as the size of *B. napus* pollen ranges from 32–35 μm (Huesken & Dietz-Pfeilstetter 2007).

Plant growth conditions

In the summer of 2013 we germinated *B. napus* seeds on glass beads, planted the seedlings in potting soil ('Lentse potgrond' no.4; Lent, the Netherlands), and placed them in a greenhouse at 20 + 1 °C. After three weeks, seedlings were transplanted to 10-litre pots and placed in an open greenhouse with less control over the temperature. Seedlings were randomly assigned to one of the four soil treatments (2 SOM levels × 2 fertilizer levels). High and low SOM treatments contained 3.0 and 1.3% SOM, fitting within the range between approximately <1 and 3% reported by (Brady *et al.* 2015). The SOM treatments were created using loamy fine sand from a long-term experiment (Korthals *et al.* 2014) at Vredepeel, south-eastern Netherlands (51°32'26.0"N 5°51'13.0"E). The top soil (upper 25 cm) contained 3.3% SOM, while the subsoil from 100 cm below the soil surface was of the same origin but contained hardly any SOM at all. We used this difference to create our experimental soils with 3.0 and 1.3 % SOM contents by thoroughly homogenizing a 9:1 mixture (high SOM) and a 2:3 mixture (low SOM) of top soil and sub soils (dry w/w). Mineral fertilizer was supplied as 0.4 g nitrogen (N) per plant, which corresponds with the average nitrogen supply per plant under regular farming practices, equalling an amount of 200 kg N ha^{-1}. Nitrogen was supplied in a dissolved form of NO_3^-, (mainly $Ca(NO_3)_2$), enriched with 0.5 L ½ Hoagland solution (High), or as 0.5 L of tap water with 10% of this solution (Low) (see Hewitt 1966 for composition of the solution). Fertilizer was supplied when plants were transplanted to the 10 L pots. After another three weeks, all plants received 100 ml ½ Hoagland solution without additional N. When the first plants started to flower all plants were placed at their randomly assigned field sites.

We standardized conditions among sites by placing the experimental plants in an open field boundary at the corner of an arable field. Herbivory from e.g. roe deer and cattle was prevented; plants were placed outside managed grazing areas and fenced with a small wire fence. To ensure that plants received ample water, each pot was placed in a construction of two stacked crates. The top crate contained the plants, while the bottom crate contained water. Water absorbing coir rope was extended through holes in the bottom of the top crate into the bottom crate, keeping the soils of the potted plants moist for a longer period of time. In addition, plants were watered every three days. After five weeks in the field when most plants had finished flowering, all plants were returned to the greenhouse to let the seeds mature under standard conditions. Here, plants were not watered anymore to stimulate senescence.

Observations

In each site, eight observations were made about pollinators on the experimental plants. During each site visit, pollinators were observed for 15 minutes in order to record the number of unique pollinators that visited each plant (see Appendix S1 for more details). Such methodology is widely used in similar studies (e.g. Steffan-Dewenter & Tscharntke 1999; Poveda *et al.* 2003).

We counted flower visits eight times and characterized pollinator visitation rates of every field site by averaging the number of bees and hoverflies at one site across all eight counting events. Prior to each pollinator count we counted the number of flowers per plant. For analyses we used the average number of counted flowers per plant per counting event.

Pods were harvested just before ripening in order to avoid loss of seeds and were stored at 4ºC. After all pods had been harvested we determined the total number of pods per plant and the weight of the seeds (g plant^{-1}). Prior to opening pods, we also inspected them for signs of damage by herbivores. As pods were frequently damaged by larvae of the moth *Evergestis extimalis*, proportion of infected pods was determined. Remaining above-ground plant biomass was harvested, dried at 70ºC for at least 48 hours, weighed and expressed as g plant^{-1}.

Analyses

To examine which of our investigated variables significantly affected *B. napus* yield and whether the effects of one variable was influenced by the level of another, we related yield (g seeds plant^{-1}) to the factors of SOM and fertilizer and the covariates pollinator visitation rate at the site level and proportion of infected pods at the plant level, using linear mixed models with the random intercept and field site as the random factor. Collinearity of explanatory variables can affect model outcome; however, in our case pollinator visitation rate and proportion of infected pods were not correlated (see Appendix S1). To avoid having too few replications per variable we only included all main effects and two-way interactions. We log-transformed seed weight in order to obtain normally distributed residuals according to the Shapiro-Wilk test.

To understand the mechanisms underlying the observed yield patterns, we also ran models with average number of flowers per plant across the visits and total plant biomass at harvest. As we did not expect to find a relationship between the number of flowers or plant biomass on the one hand and pollinator visitation rate or proportion of infected pods on the other, these models initially only included the factors of SOM and fertilizer as explanatory variables. However, exploratory analyses showed highly significant relationships with pollinator visitation rate and the proportion of infected pods, which were therefore included in these models making the analyses essentially the same as for *B. napus* yield. In all analyses, we only included plants for which we had data for all response and explanatory variables; plants that died during the study were excluded, bringing the total number of experimental plants to 213. We established that the number of excluded plants did not differ among treatments, or among the different field sites. Afterwards, we ran the models again with only significant explaining variables to estimate the relative contribution of these variables to yield.

A separate analysis was done for the four sites where additional plants had been grown under insect cages. Caged plants were compared with open pollinated plants, resulting in 79 plants on the four field sites. We examined effects of excluding all pollinators on plant biomass, flower number, seed weight and additionally the proportion of infected pods. There was not enough statistical power to test these sites for all treatments simultaneously. Therefore, as the treatments using SOM and fertilizer did not interact in affecting yield, flower number, plant biomass, and the proportion of infected pods we only tested effects of insect exclusion on these variables. Since normality could not be met for all variables of interest we used a Kruskal-Wallis test for these analyses.

All analyses were performed with R 3.1.1 (R Core Team), using the *nlme package* for linear mixed models (Pinheiro *et al.* 2014).

Results

Yield (total seed weight)

Yield was significantly affected by fertilizer, but not by soil organic matter (SOM) and there was no significant interaction between fertilizer and SOM (Table 1). Model-predicted yields of plants from the high fertilizer treatment were 80% higher than of plants from the low fertilizer treatment (Fig. 1a). There was a significantly positive relationship between pollinator visitation rate and yield (Fig 2a, Table 1). Hoverfly visitation rate was significantly related to yield, whereas bee visitation rate and pollinator diversity were not (see Appendix S2). At field sites with the highest number of pollinators, model-predicted yields were 165% higher than at field sites with the lowest number of pollinators. There was no significant interaction effect on yield between pollinator visitation rate and SOM content, or fertilization, suggesting that the effects of pollinators and fertilizers are additive and do not strengthen or weaken one another. Yield was negatively related to the proportion of pods infected by *Evergestis extimalis* larvae (Fig. 2d). This negative relationship was more pronounced with increasing pollinator visitation rate (Fig. 3a, Table 1). At field sites with the highest proportion of *E. extimalis* infected pods, model-predicted yields were 153% lower than at sites with the lowest infestation rates. The relationship between yield and the proportion of infected pods was not influenced by SOM or fertilizer (Table 1).

Number of flowers

The number of flowers was highest in the high SOM and fertilizer treatments, but there was no significant interaction between SOM and fertilizer (Fig 1b, Table 1). The number of *B. napus* flowers showed an overall positive relationship with pollinator visitation rate (Fig. 2b, Table 1). However, this relationship was more pronounced for plants with high than with low fertilizer supply, as indicated by the significant interaction between fertilizer and pollinator visitation rate on the number of flowers (Table 1). The number of flowers was significantly negatively related to the proportion of infected pods (Fig. 2e) and this relationship was stronger at field sites with higher pollinator visitation rates (Fig 3b, Table 1).

Plant biomass

Above-ground biomass, excluding pod mass, was significantly enhanced by high fertilizer supply, as well as by SOM content (Fig 1c, Table 1).There was a negative relationship between above-ground biomass and pollinator visitation rate (Fig. 2c, Table 1). However, there was a positive relationship between above-ground biomass and the proportion of infected pods (Fig. 2f). The relationship between plant biomass and pollinator visitation rate was not affected by SOM or fertilizer, as there was no significant interaction between these two soil factors (Table 1).

Pollinator exclusion

Caged plants produced less yield (average 0.11 g vs. 1.33 g; Kruskal test, X^2= 13.0623, *P*= 0.0003, d.f.=1) and fewer flowers (average 2.01 vs. 3.88; Kruskal test, X^2= 4.2119, *P*= 0.0401) than plants that were exposed to all insects. Biomass production was not significantly affected by pollinator exclusion (average 25.10 g vs. 29.52 g; Kruskal test, X^2= 1.7227, *P*= 0.1894). Cages also reduced pod infection by *E. extimalis* moths, resulting in a significantly lower proportion of infected pods (average 9% vs. 37%; Kruskal test, X^2= 5.5490, *P*=0.0185).

Discussion

We studied how soil organic matter (SOM), mineral fertilizer supply, and exposure to variations in pollinators and crop pests influence above-ground biomass, numbers of flowers, seed yield, and pod infestation of a partially insect-pollinated crop. We analysed how interactions between these below-ground and above-ground factors contribute to crop yield using oilseed rape *Brassica napus* as a model crop. Our study showed that the relationships between yield, pollinator visitation rate and pest pressure did not depend on the level of below-ground services (nutrient supply) implying that effects of above-ground and below-ground (dis)services on yield were additive. In contrast, pollinator visitation rate was more strongly related to yield at low pest pressure than at high pest pressure. Under the present experimental conditions, differences in real-world pollinator visitation rate and pest pressure influenced yield of *B. napus* more strongly than fertilizer application.

A more pronounced relationship of yield with pollination than with fertilizer supply is to be expected in crops that largely depend on insect pollination for fruit set, such as cucumber (*Cucumis sativus*; Motzke *et al.* 2015). In the case of a partially insect-pollinated crop, such as *B. napus*, the contribution of insect pollination to maximum yield is generally considered to be relatively small (Klein *et al.* 2007). An explanation for the strong yield response to variability in pollinator visitation rate in our study may be that pollination did not only affect seed set directly (Steffan-Dewenter & Tscharntke 1999; Garibaldi *et al.* 2013), but that exposure to pollinators also affected growth allocation patterns in *B. napus*. At sites with the highest pollinator visitation rates, experimental plants produced more flowers but less above-ground non-flower biomass. This could point at a trade-off between producing above-ground biomass and flowers; well-pollinated plants produced large numbers of flowers at the expense of other plant organs. Indeed, above-ground biomass and the number of flowers correlate negatively if the effects of fertilizer supply are accounted for (see Appendix S3).

Poorly pollinated plants may initially invest more in biomass possibly resulting in a more prolonged period of flower production, which enhances the likelihood of pollination later in the season. This is in line with findings by Brann and Lehtila (2007) that hand-pollination significantly enhances flower number in wild radish *Raphanus raphanistrum*, a close relative of *B. napus*. Under field conditions pollinator visitation may even relate stronger to yield than after artificial pollination, as an increase in the number of flowers could subsequently also attract more pollinators to the plant (Ohashi & Yahara 1998). Further testing is needed to unravel the relative contribution of this energy-reallocation effect (more energy allocated to flowers after pollination) and attraction effect (more pollinators due to a higher number of flowers), as well as their reinforcing interactions between energy-reallocation and attraction.

Infestation of pods by *Evergestis extimalis* larvae resulted in substantial yield losses. Herbivory-related yield losses were greater than yield reductions associated with the cessation of fertilizer applications. Under high pollinator visitation rates the negative relationship of *E. extimalis* with yield became stronger, virtually neutralizing all benefits from pollination. These results are in line with Lundin *et al.* (2013) who found a synergistic effect between pest control and pollination. In addition, we found that the proportion of pods infested by insect larvae was negatively related to flower number and positively related to leaf and stem biomass. As the moth *E. extimalis* places eggs on the leaves and larvae stay on the leaves until seed set when they migrate to the pods (Muus 2014 - Microlepidoptera.nl), it is unlikely that the moth had a direct effect on flower number. However, it is unclear which mechanism may explain the relationship between the proportion of infected pods and the number of flowers, or above-ground plant biomass.

Whether *E. extimalis* has similar effects on oilseed rape planted at the field scale will depend on its population size, which may fluctuate strongly from year to year. This species is predominantly found in field edges, but rarely occurs in the centre of the field (Jeffrey A. Harvey, *personal communication*). This might explains why *E. extimalis* is not considered an important pest of *B. napus* in the Netherlands. Simultaneously, key pest species that normally occur on *B. napus* were not found in our study, possibly because the relatively small experimental patches were not attractive for pest species such as the pollen beetle *Meligethes spec.* that usually operates at a larger spatial scale (Zaller *et al.* 2008). This reduces the potential for direct generalizations from our study for day to day farming, but does not affect our main conclusion that effects of pollinators and pests are not influenced by soil conditions.

Fertilizer supply increased seed yield by 80%, whereas doubling the amount of SOM (increase from 1.3% to 3.0%) did not have a significant effect. These results do not support earlier studies showing that SOM significantly enhances crop yield (van Eekeren *et al.* 2010) and modelling studies predicting that higher SOM content can allow for lower fertilizer application rate to obtain maximum yield (Brady *et al.* 2015). Soil organic matter enhanced the number of flowers and above-ground biomass produced by the oilseed rape plants, suggesting that providing nutrients through SOM or mineral fertilizer results in different temporal resource allocation patterns. The duration of plant exposure in the field in our experiment might have prevented finding a positive SOM effect, as seed set was not completed in the field, but under controlled experimental conditions. Considering that nutrients become available faster from mineral fertilizer than from SOM, as organic matter first needs to be decomposed by bacteria and fungi (Hendrix, Coleman & Crossley 1992), plants with high fertilizer may have developed more rapidly, within the time frame of the experiment, than plants with high SOM content. Moreover, we have controlled moisture availability of all soil treatments. SOM usually has a positive effect on plant production by promoting soil water-holding capacity (Lal 2006).

We had expected that higher nutrient availability (high SOM plus high fertilizer) would lead to more attractive plants, resulting in more pollinator visits and, therefore, a more rapid saturation in pollination service demand. In line with earlier work (Otieno *et al.* 2011) fertilizer supply and SOM indeed led to more flowers, implying greater resource availability for pollinators. However, the relationship between pollination and yield was not more pronounced on soils with high fertilizer or high SOM levels than in soils with low fertilizer or low SOM. A number of recent studies addressing interacting effects of nutrient provision and pollination result in contrasting results. In line with the present study, Bartomeus, Gagic and Bommarco (2015) found no interaction between soil properties and pollinator visitation rate on *B. napus* yield, whereas Marini *et al.* (2015), found the greatest effects of pollination on oilseed rape yield at low fertilizer application rates. Finally, Tamburini *et al.* (2015) observed less pronounced effects of pollination on sunflower *Helianthus annuus* yield on low- than on high-nutrient soils. Therefore, whether soil nutrient status enhances or reduces the effects of pollination on yield it is most likely context dependent and will differ among crop species and varieties, level and type of nutrient supply, and density and species composition of pollinator communities (Hudewenz *et al.* 2014; Marini *et al.* 2015).

Synthesis and management implications

Management focussing on combining both below-ground and above-ground ecosystem services requires integration of activities that operate locally, such as supplying fertilizer or organic matter, with landscape-scale activities focussing on promoting pollinator and pest control services (Tscharntke *et al.* 2012). However, soil organic matter (SOM) management and nutrient supply are usually considered independent from managing pollinators and pest control organisms. Our results show that real-world variation in pollinator visitation rate and pest pressure have yield effects that may be even larger than those of factors that have traditionally been the focus of farming practices, such as fertilizer application and management of SOM. Effects of pests and pollinators add up to those from fertilizer supply and, although less prominent in our study, SOM management. Moreover, as pollination is known to also enhance oil content of oilseed rape seeds (Bommarco, Marini & Vaissiere 2012), the importance of considering pollination in combination with SOM and nutrient management might even be higher than appearing from our results. Therefore, we suggest that farmers can improve yield by optimizing both the nutrient provisioning of their crops and the services that are supplied largely by the landscape. Because the production value of insect-pollinated field crops is generally low (FAO 2014), enhancing crop pollination in such systems might best be achieved by managing existing field margins and semi-natural habitats in a more pollinator-friendly way rather than creating costly new pollinator habitat such as wildflower fields (Schellhorn, Gagic & Bommarco 2015). Further field studies are required to determine when the positive relationship between pollinator visitation and yield, and between fertilizer and yield, will level off. This information is crucial in order to optimize ecosystem services and external inputs from an economic and ecological perspective.

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Data accessibility

Data available from Dryad Digital Repository: doi:10.5061/dryad.s08f7 (van Gils, van der Putten & Kleijn 2016).

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Appendix S1. Field selection

Appendix S2. Community effects of pollinators on yield

Appendix S3. Correlation between biomass and the number of flowers

Table 1. Below-ground (soil organic matter [SOM] and mineral fertilizer) and above-ground (pollinator visitation rate, proportion of infected pods) factors and covariates explaining yield and performance indicators of *Brassica napus*. Bold values represent a significant relationship (*P*<0.05). All analyses were performed using a linear mixed model with random intercept. Cases were included only when all data were available, resulting in 213 plants, across 15 landscape sites

Response	**Total seed weight (yield;ln+1)**			**Average number of flowers**			**Above-ground biomass plant**		
	Value	F	*P*	Value	F	*P*	Value	F	*P*
Intercept	**0.129**	**156.77**	**<.0001**	**1.014**	**305.30**	**<.0001**	**20.2125**	**1655.01**	**<.0001**
Below-ground									
SOM	0.043	1.06	0.3045	**0.608**	**6.07**	**0.0147**	**7.9667**	**81.65**	**<.0001**
Fertilizer	**0.335**	**26.06**	**<.0001**	**1.886**	**59.59**	**<.0001**	**15.4403**	**401.50**	**<.0001**
Above-ground									
Pollinator visitation rate	**0.099**	**8.89**	**0.0106**	**0.556**	**18.83**	**0.0008**	**-0.6004**	**8.14**	**0.0136**
Proportion of infected pods	**0.252**	**46.87**	**<.0001**	**1.114**	**18.59**	**<.0001**	**0.3081**	**5.71**	**0.0178**
Interactions									
SOM:Fertilizer	0.017	0.15	0.6944	-0.557	0.40	0.5295	-1.7127	1.88	0.1725
SOM:Pollinator visitation rate	0.019	0.21	0.6512	0.117	0.24	0.6278	-0.2023	0.61	0.4359
Fertilizer:Pollinator visitation rate	-0.002	0.57	0.4527	**0.217**	**4.22**	**0.0414**	-0.1605	0.69	0.4084
SOM:Proportion of infected pods	-0.216	2.56	0.1111	0.290	0.09	0.7697	1.1801	0.42	0.5172
Fertilizer:Proportion of infected pods	-0.232	1.12	0.2911	-0.722	0.32	0.5712	0.9299	0.17	0.6786
Pollinator vis. rate:Prop. inf. pods	**-0.134**	**8.27**	**0.0045**	**-0.636**	**5.23**	**0.0233**	0.2437	0.27	0.6011

Figure 1. Effects of soil organic matter (SOM) and mineral fertilizer on performance of *Brassica napus*: a) total seed weight, b) average number of flowers, c) above-ground biomass (excluding pods and seeds). Error bars represent standard error. Letters above the bar graph (a, b, c) indicate a significant (P<0.05) difference between specific treatment combinations after a Least Squares Difference (LSD) contrast test.

Figure 2. Factors (soil organic matter, mineral fertilizer) and covariates (pollinator visitation rate, proportion of pods infected by *Evergestis extimalis*) affecting yield (total seed weight; a&d), average number of flowers (b&e) and above-ground biomass (c&f) of *Brassica napus*. Graphs are based on linear mixed models with field site (being a proxy for insect abundance) as the random factor. S0: low soil organic matter; S1: high soil organic matter; F0: low fertilizer; F1: high fertilizer. Only significant factors and covariates are used in the models. If significant covariates are not displayed, we used the average value in the model. Thin lines represent low fertilizer (F0 in legend) and thick lines represent high fertilizer (F1). If soil organic matter was significant we separated the treatment effect. Solid lines represent low (S0), and dashed high (S1) soil organic matter.

Figure 3. Relationship between pollinator visitation rate and yield (a) and average number of flowers (b) of *Brassica napus* under different proportions of *Evergestis extimalis* infected pods. Graphs are based on linear mixed models with field site (being a proxy for insect abundance) as the random factor. Line thickness from thick to thin represents the proportion of infected pods by *Evergestis extimalis* (0% infected, 25% infected, 50% infected, 75% infected). Treatments were visualized with the same symbols as explained in the legend of Fig. 2.

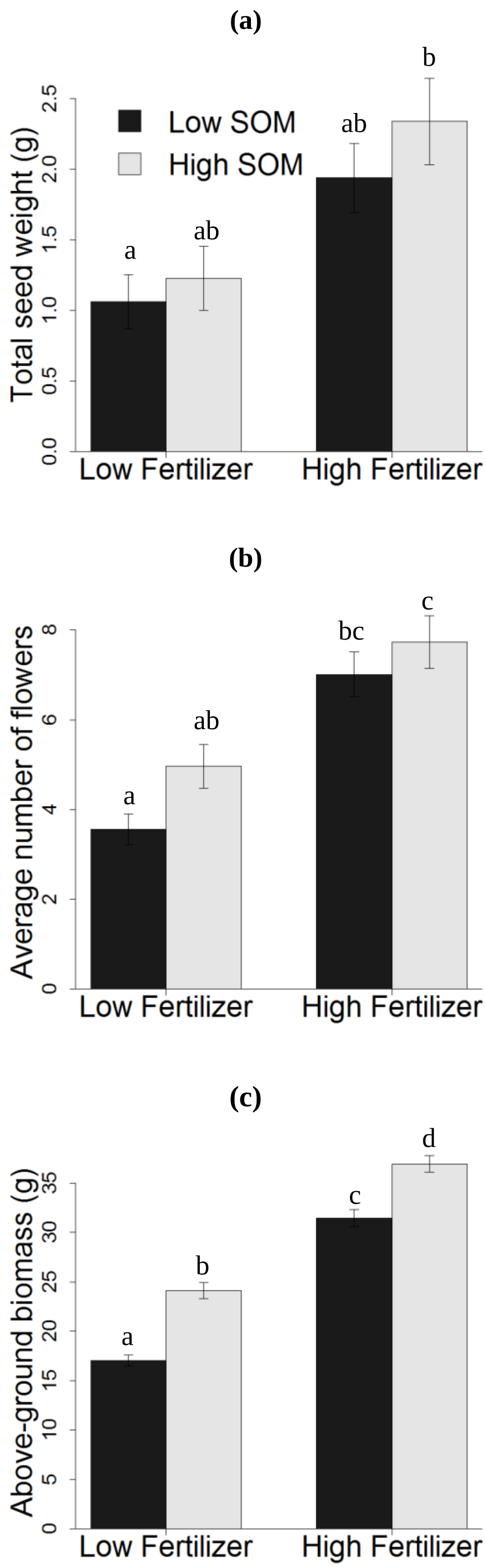
(a)
Total seed weight (g)
0.0
0.5
1.0
1.5
2.0
2.5
Low SOM
High SOM
a
ab
ab
b
Low Fertilizer
High Fertilizer
(b)
Average number of flowers
0
2
4
6
8
a
ab
bc
c
Low Fertilizer
High Fertilizer
(c)
Above-ground biomass (g)
0
5
10
15
20
25
30
35
a
b
c
d
Low Fertilizer
High Fertilizer

Figure 1.

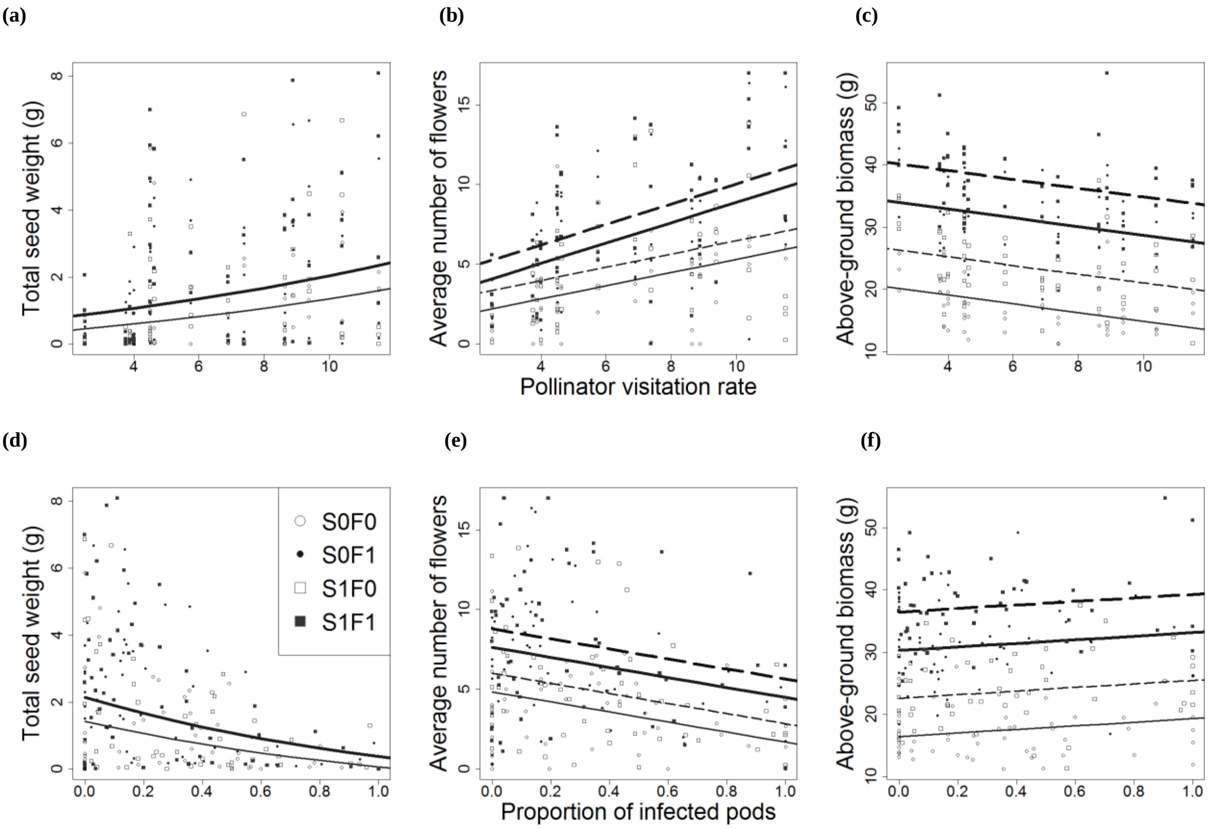
(a)
(b)
(c)
Total seed weight (g)
Average number of flowers
Above-ground biomass (g)
Pollinator visitation rate
(d)
(e)
(f)
S0F0
S0F1
S1F0
S1F1
Proportion of infected pods

Figure 2.

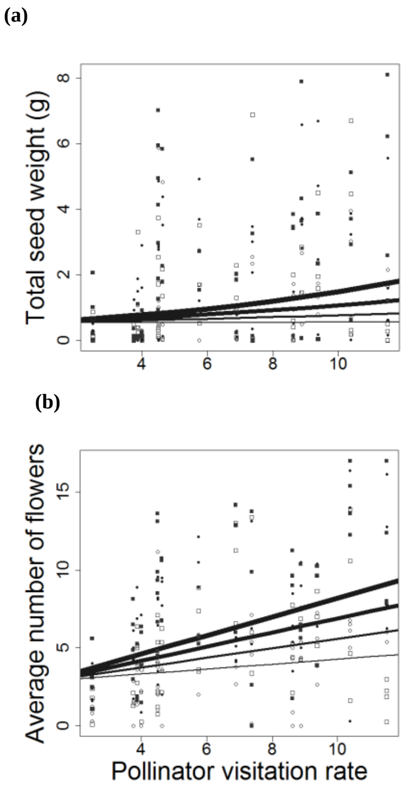
(a)
Total seed weight (g)
0
2
4
6
8
4
6
8
10
(b)
Average number of flowers
0
5
10
15
4
6
8
10
Pollinator visitation rate

Figure 3.