

Research review

Nematodes as suppressors and facilitators of plant performance

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Summary

Plant–nematode interactions are mainly considered from the negative aspect with a focus on plant-parasitic nematodes (PPNs), which is justified considering the agronomic losses caused by PPNS. Despite the fact that PPNS are outnumbered by nonparasitic free-living nematodes (FLNs), the functional importance of FLNs, especially with regard to plant performance, remains largely unknown. Here, we provide a comprehensive overview and most recent insights into soil nematodes by showing direct and indirect links of both PPNS and FLNs with plant performance. We especially emphasize the knowledge gaps and potential of FLNs as important indirect players in driving plant performance such as stimulating the resistance to pests via improving the disease suppressive activity of the rhizobiome. Together, we present a holistic view of soil nematodes as positive and negative contributors to plant performance, accentuating the positive but underexplored role of FLNs.

Introduction: importance of soil nematodes for plant performance

Plants are the basis of almost all life on Earth, especially that of animals including humans. We have modified most land on Earth for our need to grow human-essential plants in agriculture that led to fundamental increases in food production. However, constant food production is not guaranteed, partly because plant performance can be reduced by pests, here referring to all organisms, including pathogens that impair plant nutrition, growth, and defense (Kranz, 2005; Döring *et al.*, 2011). These pests lead to *c.* 40% global crop yield losses annually (IPPC Secretariat, 2021). The pests are often controlled by pesticides, but at a cost: negative side-effects, such as losses of nontarget biodiversity and detrimental impacts on the environment might in the long-run outweigh the short-term benefits of plant stimulation (Oka, 2020). To promote a more sustainable plant protection, the European Commission aims to halve the current use of pesticides by 2030 (European Commission, 2022), putting yield at risk.

Among these pesticides are nematicides that act against plant-parasitic nematodes (PPNs). Plant-parasitic nematodes are notorious plant pests that cause annual yield losses of over 80 billion US\$ worldwide (Abd-Elgawad & Askary, 2015). There are over 4100 currently described PPN species that parasitize virtually all plant species, including all crops (Decraemer &

Hunt, 2006). Therefore, nematodes are mostly viewed as negative contributors to plant performance. However, nematodes can also positively contribute to plant performance. For example, non-parasitic free-living nematodes (FLNs) (here defined as non-parasitic soil nematodes excluding entomopathogenic nematodes as obligate insect parasites) are the most abundant soil animals that outnumber PPNS in terms of abundance and diversity (van den Hoogen *et al.*, 2019). Free-living nematodes display different functions in the soil food web and can act, for example, as consumers of bacteria, fungi, protists, and other nematodes. Their rapid response to environmental disturbances makes them good bioindicators of soil health (Ferris *et al.*, 2001). The abundance and trophic nature of FLNs are often used for measuring different nematode-based indicators that can reflect the ecosystem status of soil (Du Preez *et al.*, 2022). Furthermore, FLNs can more or less selectively feed on members of the soil microbiome (Rønn *et al.*, 2012; Liu *et al.*, 2017), acquire microorganisms on their surface (Dirksen *et al.*, 2016), transfer microbiomes to remote habitats (Finkel *et al.*, 2017), and serve as a food to higher trophic groups (Stirling, 2014). These interactions may shape the rhizobiome, here defined as the biodiversity attached to and influenced by roots, and ensure nutrient transfer between microbiomes and plants to eventually shape plant performance (Trap *et al.*, 2016). However, the functional links between FLNs and plant performance remain surprisingly little studied.

In this review, we collected reports and presented a multitude of pathways in which PPNs and FLNs inhibit and most importantly promote plant performance. More specifically, we show that direct links that are governed by nematode feeding on roots are prevalent between PPNs and plants and, thereby, negatively affect plant performance. On the contrary, indirect links, often being plant-beneficial, are more pronounced between FLNs and plants via interlinkages of FLNs with the rhizobiome through predation and dispersal. We especially highlight the knowledge gaps in understanding the role of FLNs to assist plant protection against pests and explore how their multitrophic position in the food web can contribute to this aspect.

Negative impact of nematodes on plant performance

Direct negative impact

Deeply studied, well-known and widely recognized and feared among farmers and gardeners are PPNs that directly damage plants via feeding mainly on roots. Among these PPNs, endoparasitic PPNs (especially migratory and sedentary groups that we feature below) cause the most damage as they are in the most intimate contact with the plant (Jones *et al.*, 2013). The damage threshold for notorious root-knot nematodes was estimated to be only between 1 and 8 eggs per cm³ of root material (Greco & Di Vito, 2009). In addition to forming progenies that can amount to

several hundreds of eggs per only one female, some PPNs have the highest spreading rate of all plant pests due to their wide host range of many thousands of plant species (Bebber *et al.*, 2014). Endoparasitic PPNs use short stylets to penetrate the root and feed directly from the parenchymal cells in the vascular cylinder (Jones *et al.*, 2013) (Fig. 1). Sedentary endoparasitic PPNs are represented by two subgroups: root-knot nematodes and cyst nematodes. Root-knot nematodes infect various plant species, including vegetables, grasses, shrubs, and trees. Some root-knot nematode species have a wider host range than others; for instance, *Meloidogyne incognita* is ubiquitous and infects > 3000 plant species including major crops, while *Meloidogyne mali* parasitizes mostly woody perennial plants. Cyst nematodes generally have a narrower host range than root-knot nematodes, but can be devastating to their host. *Heterodera schachtii* infections alone can cause 95% of the annual yield losses in sugar beet (Müller, 1999).

Sedentary endoparasitic PPNs have different life stages. Only the second-stage juveniles (J2s) infect plant roots. Once inside the root, the J2s take symplastic (cyst nematodes) or apoplastic (root-knot nematodes) routes to reach the permanent sites for feeding. For example, root-knot nematodes, being unable to break through the root endodermis, enter the root at the elongation zone of the root tips and use meristem tissue to pass to the vascular cylinder (Grundler *et al.*, 1992), while cyst nematodes directly pass through the endodermis barrier (Holbein *et al.*, 2016). At their destination, PPNs manipulate defense mechanisms in the host plant, including

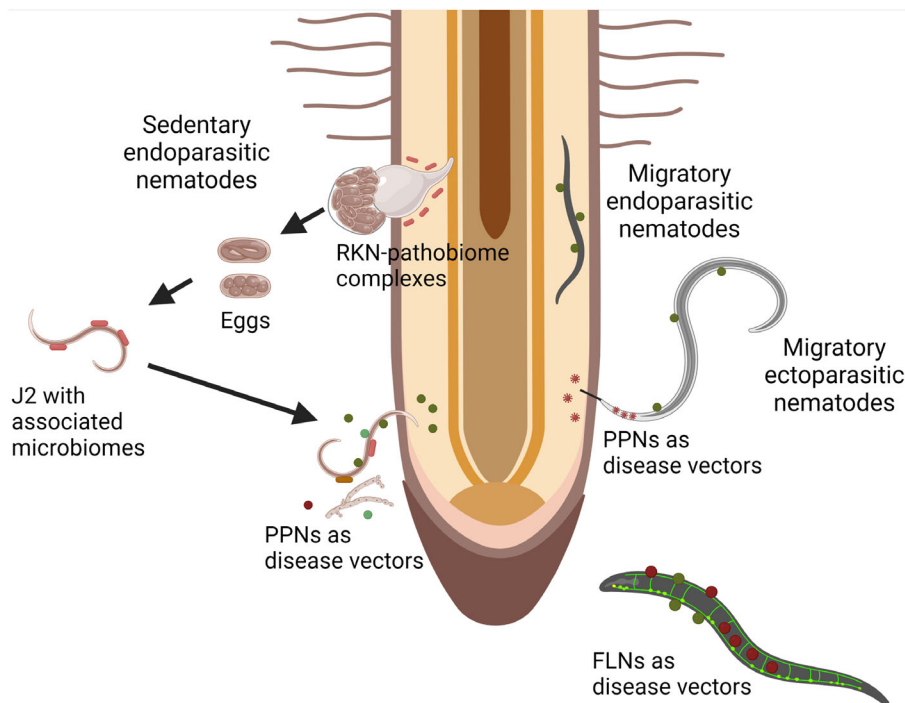


Fig. 1 Negative impact of nematodes on plant performance. Direct negative impact (parasitism): Nematode groups that feed on plants are sedentary endoparasitic nematodes, migratory endoparasitic nematodes, and ectoparasitic nematodes. Root-knot nematodes (RKN) form galls on the roots with hundreds of eggs in egg masses. Migratory endoparasitic nematodes reside outside and inside the roots, where they make necrotic lesions during moving and feeding. Ectoparasitic nematodes reside outside the roots and only insert the stylet within the root, while some of them are vectors of plant viruses. Indirect negative impact: All plant-parasitic nematodes (PPNs) create wounds for the entrance of secondary pathogens. PPNs and free-living nematodes (FLNs) can acquire plant-deleterious microorganisms endogenously or on their surface, transport them to the roots, and stimulate the formation of a pathobiome.

the repression of cell wall barrier defense and changes in diverse hormonal pathways (Holbein *et al.*, 2016). Nematode feeding directly damages plant tissue, leading to necrosis and anatomical alterations of the roots, such as formation of root galls by root-knot nematodes. Deformations and impaired root growth can result in reduced water uptake, photosynthesis and plant growth.

Migratory endoparasitic nematodes, including root lesion nematodes (*Pratylenchus* spp.) and burrowing nematodes (*Radopholus* spp.), are also among devastating PPNs (Jones *et al.*, 2013). All stages of migratory endoparasitic nematodes are mobile and vermiform. A yellow disease of black pepper caused by *Radopholus similis* in 1950s was responsible for the loss of 20 million black pepper trees in Indonesia (Hubert, 1957). The root lesion nematode *Pratylenchus thornei* was reported to cause between 25% and 28% of grain yield losses in wheat and barley when present at densities between 150 and 250 nematodes g⁻¹ soil (Fanning *et al.*, 2020).

In contrast to endoparasitic PPNs, ectoparasitic PPNs use their long stylets to feed on different root parts without invading the roots. Economically, they are less important than the endoparasitic nematodes, especially if they primarily feed on root hairs. However, they can also cause injuries to the outer layers of cortical tissue and progressive deterioration of the root system as in case of *Helicotylenchus* spp. (Riascos-Ortiz *et al.*, 2020).

Indirect negative impact

Indirectly, PPNs can negatively affect plants via multifaceted interactions with microorganisms (Fig. 1). Plant-parasitic nematodes can form disease complexes with some plant-pathogenic members of the rhizobiome, including bacteria, fungi, and viruses (Back *et al.*, 2002; Lamelas *et al.*, 2020). Notable examples are diseases caused by synergistic interactions between *Meloidogyne* spp. and *Fusarium* wilt pathogens, or by *Pratylenchus*- and *Globodera-Verticillium dahliae* complexes (Back *et al.*, 2002). Besides using the wounds created by PPNs for the entrance into the roots, some secondary pathogens can also use PPNs for phoresy (being transported on the nematode body without harming the nematode). For instance, some ectoparasitic PPNs, such as taxa in the families Trichodoridae and Longidoridae, transmit plant-pathogenic viruses (nepoviruses and tobnaviruses) via their stylet when feeding on roots (Decraemer & Robbins, 2007). Arguably, the most notorious pest on vineyards is the dagger nematode *Xiphinema index* that transmits Grapevine fanleaf virus (Jones *et al.*, 2013). In addition, some plant-pathogenic rhizobiomes seem to hitchhike on nematodes (Karimi *et al.*, 2000; Adam *et al.*, 2014). It was also shown that root-lesion nematodes can impair the symbiosis between the soybean roots and nitrogen fixing bacteria by impairing the formation of bacterial nodules (Elhady *et al.*, 2020).

Recent insights into plant–nematode interactions reveal that root infections by endoparasitic PPNs shift microbial functions such as enhancing degradation of root compounds that increase PPN performance and plant damage (Tian *et al.*, 2015; Yergaliyev *et al.*, 2020). Collectively, PPNs and microorganisms that support PPN development can be referred to as a pathobiome (Bass *et al.*, 2019). The precise interactions within this potential

pathobiome and the mechanisms of how nematodes may facilitate the spread of other pathogens remains unknown. It has been suggested that the specific rhizobiomes that are transmitted via the cuticle of endoparasitic PPNs into the roots can protect PPNs against the PPN-antagonistic root endophytes (Brown, 2018; Topalović & Vestergård, 2021). Better understanding the microbial role in the life cycle of PPNs on plants would be pivotal for disrupting the optimal host conditions for PPN development.

In contrast to PPNs, there is little information on how FLNs may negatively impact plant performance. Several studies suggested that FLNs can also serve as vectors of plant deleterious microorganisms. *Caenorhabditis elegans* and *Pristionchus* sp. were shown to transmit soft-rot bacteria endogenously and on their surface to the plant roots (Nykyri *et al.*, 2013). *Diplogaster* sp. was reported as a vector of several plant pathogens including *Agrobacterium tumefaciens*, *Erwinia amylovora*, *Erwinia carotovora*, *Pseudomonas phaseolicola*, and a phage of *A. tumefaciens* (Chantanao, 1969). It was shown that nematodes feed on these bacteria, which then survive the passage through the nematode digestive tract and get excreted outside. More recently, Kanfra *et al.* (2018) reported that an increased abundance of some FLNs, such as *Acrobeles* and *Acrobeloides*, and their associated microbes in apple replant diseased field soils, was associated with more serious disease symptoms (Kanfra *et al.*, 2018). However, the exact mechanisms behind this interaction remain to be determined.

Positive impact of nematodes on plant performance

Direct positive impact

Unlike arbuscular mycorrhizal fungi (Bennett & Groten, 2022) and some nitrogen (N) fixing bacteria (Schroeder *et al.*, 2020) that directly enhance nutrient levels in plants and increase stress tolerance, such direct positive relationship between plants and nematodes likely does not exist.

Indirect positive impact

Indirectly, nematodes can stimulate plant performance by promoting the activity of plant growth-promoting rhizobiome (PGPR) including biocontrol agents via root puncturing (PPNs), microbe-induced carbon sequestration (FLNs), and regulation of pests and pathogens (FLNs) (Fig. 2). In some cases, the roots punctured by sedentary endoparasitic nematodes are less attractive to other herbivores due to nematode-induced systemic defense responses in plants (Wondafraash *et al.*, 2013; Arce *et al.*, 2017). However, this happens only in primed plants or plants resistant to sedentary endoparasitic nematodes and cannot be regarded as a positive contribution of PPNs to plant defense. There is evidence that microorganisms that attach to the surface of endoparasitic PPNs can induce an array of early plant defense responses against the invading nematodes, including expressions of jasmonic acid/ethylene- and salicylic acid-signaling genes and an oxidative burst (Topalović *et al.*, 2020). On the contrary, it was shown that when the population of a clover cyst nematode *Heterodera trifolii* is below the damage threshold of the clover, it may stimulate the release of

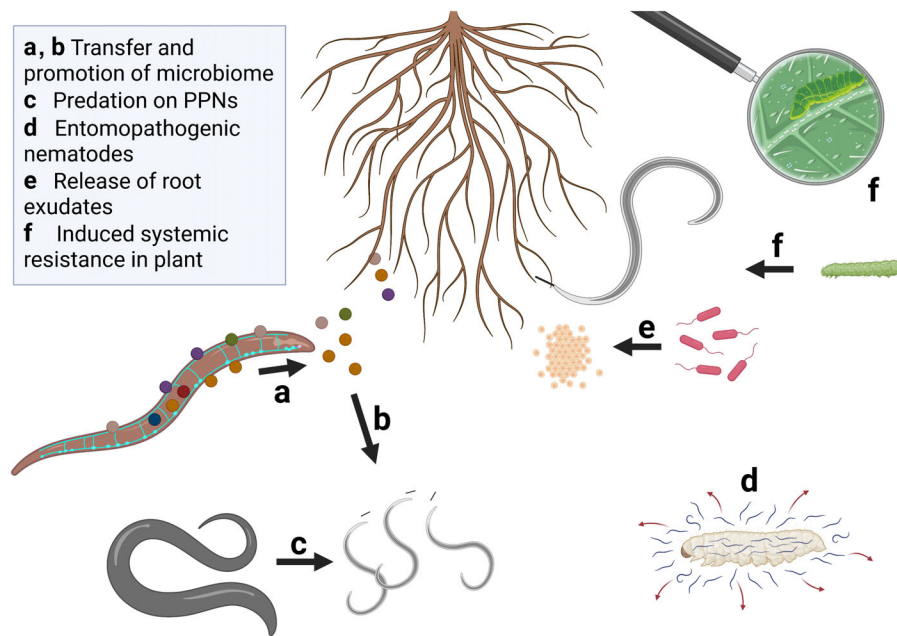


Fig. 2 Positive impact of nematodes on plant performance. Major positive contributions to plant performance are attributed to free-living nematodes (FLNs) and their multitrophic position in the soil food web. The transfer and activation of plant growth promoting microorganisms (a, b): FLNs predate on soil bacteria and fungi and release nutrients for plant uptake. Predation on plant-parasitic nematodes (PPNs) (c): some FLNs predate on PPNs and reduce their proliferation. Entomopathogenic nematodes (d): the infective stages of entomopathogenic nematodes receive signals from the plant when endangered by larvae of insect pests and together with their endosymbiotic bacteria invade the larvae and intoxicate them. Release of root exudates for microbial growth (e): below damage-threshold populations of some PPNs (e.g. *Heterodera trifolii*) can promote PGPR by puncturing the roots with their stylets and releasing root exudates. (f) Induced systemic resistance in plants against secondary pests: in plants resistant to PPNs, PPN attack can trigger plant defense against aboveground and belowground herbivores.

root exudates and promote microbial growth in the rhizosphere. This can increase the biomass of the host and neighboring plants (Denton *et al.*, 1998; Bardgett *et al.*, 1999; Yeates, 1999). We lack insights into whether this positive PPN-plant link in natural systems might be applicable in agricultural settings.

Arguably, the largest proportion of positive contributions of nematodes to the plant come from FLNs. These contributions are driven by plant inputs in soil (bottom-up regulation) and multitrophic functions of FLNs, especially as predators of bacteria and fungi (Thakur & Geisen, 2019). Some predatory FLNs directly feed on PPNs, but as predatory FLNs hardly grow in high numbers and feed on other FLNs that may be plant beneficial, they have never reached a wider application (Khan & Kim, 2007). Microbivorous FLNs and protists feed on the microbiome through selective and nonselective consumption (Liu *et al.*, 2017). Similar to protists, FLNs have a C : N ratio similar to that of their microbial prey that will be lowered in the predator through respiration of carbon (Ferris *et al.*, 1997). The extra N and N rich compounds such as $\text{NH}_4\text{-N}$ are released and available for plant uptake (Ingham *et al.*, 1985). Zheng *et al.* (2022) also revealed that nematode predation induces competitive interactions and a higher diversity of alkaline phosphomonoesterase (ALP)-producing bacteria and increased ALP activity in the rhizosphere of rapeseed plants that enhanced P availability. Another study has shown that selective predation of fungivorous FLNs can increase the plant colonization by arbuscular mycorrhizal fungi leading to enhances in plant P and productivity (Jiang *et al.*, 2020).

Besides predation, bacterial cells and fungal spores in soil can attach to the surface of FLNs and be transported from nutrient-poor niches (away from rhizosphere) to nutrient-rich (rhizosphere) niches through phoresy, where some microorganisms can be beneficial to plant performance (Horiuchi *et al.*, 2005; García-Sánchez *et al.*, 2021). However, it is difficult to discern the ingested and surface-attached FLN-associated microbiomes in their natural environment (Dirksen *et al.*, 2016), and most studies on microbial attachment to the nematode surface are performed with PPNs that do not ingest microorganisms (Elhady *et al.*, 2017; Topalović *et al.*, 2019; Mohan *et al.*, 2020). Phoresy itself may not involve high specificity as compared to the attachment of parasites (Elhady *et al.*, 2017) or nematode-protective microbiomes (Topalović & Vestergård, 2021), especially if microorganisms quickly detach from the nematode. Nonetheless, phoresy could play important roles in transport and activation of plant growth- and plant defense-promoting microorganisms. There is evidence, which shows that the plant itself mediates the phoretic events between FLNs and plant-beneficial bacteria. Horiuchi *et al.* (2005) have shown that the legume *Medicago truncatula* releases specific volatiles, such as dimethyl sulfide, which attract bacterivorous *Caenorhabditis elegans* to roots. Thereby, *C. elegans* bring nitrogen-fixing *Sinorhizobium meliloti* endogenously and on their cuticle closer to the roots. Another study reported that root colonization by the plant-growth-promoting rhizobacterium *Pseudomonas fluorescense* increased in the presence of soil nematodes (Knox *et al.*, 2004). More targeted approaches such as labeling and microscopically

tracing microbial hitchhikers both in the rhizosphere and *in planta* would provide a more precise mechanistic understanding on the role FLNs play in an active microbial transport and its role in plant stimulation.

Although in our review, we exclude entomopathogenic nematodes from the definition of FLNs, it is important to mention their indirect positive contribution to plant health. The entomopathogenic rhabditid nematodes, *Steinernema* spp. and *Heterorhabditis* spp. form symbiotic complexes with bacterial species within the genus *Xenorhabdus* and *Photorhabdus* that jointly kill the insect host after entering its gut (Brivio & Mastore, 2018). The entomopathogenic nematode-microbial symbiotic complexes are used commercially against various insect pests (Koppenhöfer *et al.*, 2020).

How can FLNs assist plant resistance to pests?

Although FLNs are among the most abundant soil organisms spanning across all trophic levels, their potential to suppress plant pests is underexplored beyond the well-known commercial use of entomopathogenic nematodes. For improved plant performance, the FLNs in the rhizosphere should promote activities of plant-beneficial microorganisms and reduce activities of plant-deleterious microorganisms ('disease-suppressive FLNs'). One way to establish 'disease suppressive FLNs' in soil is by employing agricultural practices that facilitate their establishment. There are several empirical examples that display the correlation between the FLN community response to certain sustainable agricultural practices and plant disease suppression. For instance, Masson *et al.* (2020) have reported that the presence of *Meloidogyne graminicola* and some other PPNs in rice roots decreased 7 yr after converting conventionally managed rice field to conservation agriculture consisting of no-tillage,

application of mulch and inclusion of cover crops. The reduction of PPNs in rice roots under conservation agriculture was correlated with increased microbial richness and diversity and increased relative abundance of different groups of nematodes, especially omnivores. In addition, Schmidt *et al.* (2020) have shown that a reduced incidence of pea root rot disease under long-term organic minimum tillage with cover cropping and mulch application was positively correlated with different nematode-based indicators including increased nematode abundances, higher metabolic enrichment, and higher bacterivore carbon footprints. However, although there are positive links between different nematode-based indicators and FLN groups with the suppression of plant pests in previous studies, we need more studies that combine laboratory assays with field experiments to unravel whether this relationship is causative and how it can be utilized for improved plant performance (see the Section [Outstanding questions and solutions](#)).

Another promising mechanism that can be exploited to make FLNs applicable is their potential to disperse immobile biocontrol agents (King & Bell, 2022). For that, FLNs could be added together with PGPR and thereby help their establishment and efficiency. Having this in mind, we propose how FLNs may assist direct and indirect PPN suppression via interacting with different microorganisms (Fig. 3). For microorganisms that require attachment to the PPN surface for PPN suppression, FLNs may transport these microorganisms closer to PPNs and increase the probability of microbial attachment to the PPN surface and PPN suppression (Fig. 3a,c). In addition, selective predation of FLNs on certain rhizobiome taxa can increase the abundance and activity of PPN antagonists by inhibiting their competitors (Fig. 3b,c). Finally, FLNs can transport some plant-beneficial rhizobiome taxa with plant-defense promoting functions closer to the roots and prevent root invasion by PPNs (Fig. 3d). Thus, through feeding and

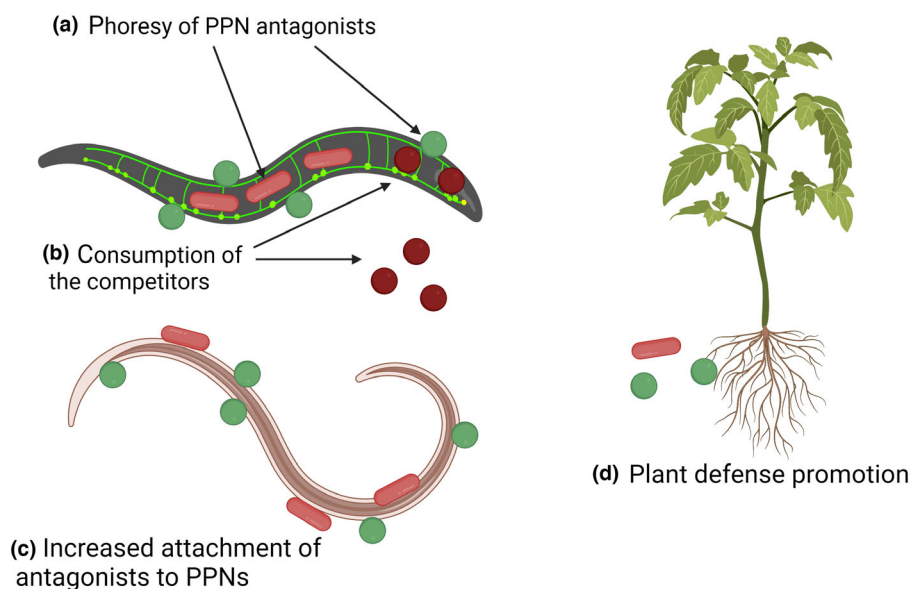


Fig. 3 Indirect contribution of free-living nematodes (FLNs) to plant defense against plant-parasitic nematodes (PPNs). FLNs transfer antagonists endogenically and via their cuticle close to PPNs (a). FLNs consume the competitors of PPN antagonists (b). FLNs increase the attachment of antagonists to the surface of PPNs (c). FLNs mediate the plant response to PPNs in the rhizosphere via modulating the rhizobiome (d).

Box 1 Molecular aspects of plant-FLN links

The links between PPN recognition and establishment in plants have been extensively studied and reviewed due to their well explored direct negative impact on plant performance (Holbein *et al.*, 2016; Gheysen & Mitchum, 2019; Siddique *et al.*, 2022). On the other hand, there are fewer studies on the precise molecular mechanisms of FLN-plant interactions. For example, ascarosides are signaling molecules widely conserved among nematodes, but their role in nematode recognition by the plant has only been studied for PPNs (Choe *et al.*, 2012). Plants can read and edit the chemical composition of a specific ascaroside (asc#18), sometimes leading to a repulsive response against PPN infection (Manoslava *et al.*, 2015; Manohar *et al.*, 2020). For FLNs, the role of ascarosides was determined mostly in relation to behavioral cues, such as mating and dispersal (Kaplan *et al.*, 2020) or in relation to other nematode groups, but not in their relation to the plant. For example, some species of root-knot nematodes are repelled by ascarosides from *C. elegans* under laboratory conditions (Kaplan *et al.*, 2012), and it would be important to study whether this repulsion is partly plant-mediated. Yet, plants might 'cry for help' for entomopathogenic FLNs when endangered by insect larvae by producing sesquiterpene (E)- β -caryophyllene (Ebc) that attract nematodes close to the roots and larvae (Rasmann *et al.*, 2005). Conversely, some compounds, such as specific metabolites of benzoxanoids synthesized by maize plants, act as general repellents against some FLN taxa (Sikder *et al.*, 2022). Future studies should be aimed at understanding how plants respond to the assembly of FLN communities and how it affects plant performance (Section [Outstanding questions and solutions](#)).

phoresy, FLNs may assist PPN suppression by increasing the abundance and activity of nematode-suppressive rhizobiome taxa.

However, the complexity of soil food webs may still impose challenges in applying FLNs *in situ* (Berlinches de Gea *et al.*, 2022). For instance, FLNs are always endangered by their own pathogenic microorganisms in soil (Katzen *et al.*, 2021). In addition, as mentioned above, plant pathogens can be transferred or consumed by FLNs (Chantanao, 1969; Nykyri *et al.*, 2013; Kanfra *et al.*, 2018). To make it more complex, many microorganisms protect themselves against predation. For instance, when endangered by nematodes, some bacteria release urea that alters the feeding lifestyle of the nematode-trapping fungus *Arthrobotrys oligospora* from saprophyagy to predation, such as on nematodes (Wang *et al.*, 2014). Other bacteria produce biofilms that reduce consumption by FLNs (Darby *et al.*, 2002).

More fundamental and applied research should be performed to understand the multitude of interactions between FLNs, plants (Box 1), disease-suppressive microbial communities in soil and applied biocontrol agents (see the Section [Outstanding questions and solutions](#)). This could result in mass-production of specific FLN taxa that promote disease suppression by interacting with other soil biodiversity and in their application as 'bio-additives' either alone or in a combination with biocontrol agents.

Conclusions

Soil nematodes may positively and negatively affect plant performance. Plant-parasitic nematodes mostly negatively affect

plant performance by direct feeding as endoparasites and ectoparasites and as facilitators of secondary pathogens in disease complexes and disease vectors. In turn, FLNs commonly indirectly stimulate plant performance via interactions with other soil biodiversity, especially bacteria and fungi. Via microbial feeding and phoresy, FLNs enhance microbial activity that might lead to increased carbon sequestration, nutrient cycling and plant growth. Furthermore, entomopathogenic FLNs are commercially available antagonists of insect pests.

However, the role of FLNs as plant beneficial organisms such as their role in suppressing pests remains poorly known despite evident links reviewed here, and therefore not exploited at all in applied products. This calls for an urgent need to expand ecological work on soil nematodes, particularly on their potential as plant promoting agents. A visionary way forward in using FLNs in enhancing plant performance in agricultural systems is to promote FLNs through adapted agricultural practices that are plant beneficial, either by catalyzing soil nutrient cycling or by manipulating the soil microbiome in favor of plant beneficial rather than pathogenic microorganisms. Another way can be in the application of FLNs as biological agents that shape the microbiome in a plant-enhancing manner. In any case, we are convinced that FLNs are key elements driving plant performance that should be more closely considered in both fundamental and applied research.

Outstanding questions and solutions

(1) How can we implement a more positive view on nematodes as plant promoting agents that extends the dominating negative view driven by PPNs?

Designing and conducting functional studies that focus on FLNs beyond simple compositional analyses.

(2) Which levels of PPNs can be beneficial for host and non-host plant species and under which conditions?

PPN-centered studies should involve diverse host and non-host plant species and varying levels of PPNs to test plant performance.

(3) How do we bridge scientific insights into the positive role of nematodes for plant performance to application?

Replicated field experiments to study nematodes in response to other management practices and identifying key nematode taxa linked to plant performance.

(4) What are the links between FLNs and plant diseases?

Screening the effects of diverse FLN species with disease-causing microorganisms on different plant species in the greenhouse experiments.

(5) Which methods are best to examine the mechanisms underlying plant performance increases by FLNs?

Integrative methods including laboratory, greenhouse and field experiments followed by microscopy, transcriptomics, metagenomics and metabolomics to study the specificity and mechanisms of plant-FLN-microbe associations.

(6) How to find the most effective nematode-microbial species combinations that enhance plant performance?

Targeted cultivation and screening of the potential plant species- and genotype-specific FLN-microbial combinations that promote plant performance.

(7) How to bring FLNs as biological agents to the market? Standardization of protocols for a large-scale multiplication and application of FLN products in the field as exemplified by entomopathogenic nematodes.

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Competing interests

None declared.

Author contributions

OT and SG contributed with the ideas, concepts, and writing. SG supervised and revised the manuscript.

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References

- Abd-Elgawad MMM, Askary TH. 2015. Impact of nematodes on agriculture economy. In: Askary TH, Martinelli PRP, eds. *Biocontrol agents of phytonematodes*. Wallingford, UK: CAB International, 3–49.
- Adam M, Westphal A, Hallmann J, Heuer H. 2014. Specific microbial attachment to root knot nematodes in suppressive soil. *Applied and Environmental Microbiology* 80: 2679–2686.
- Arce CCM, Machado RAR, Ribas NS, Cristaldo PF, Ataíde LMS, Pallini Â, Carmo FM, Freitas LG, Lima E. 2017. Nematode root herbivory in tomato increases leaf defenses and reduces leaf miner oviposition and performance. *Journal of Chemical Ecology* 43: 120–128.
- Back MA, Haydock PPJ, Jenkinson P. 2002. Disease complexes involving plant parasitic nematodes and soilborne pathogens. *Plant Pathology* 51: 683–697.
- Bardgett RD, Denton CS, Cook R. 1999. Below-ground herbivory promotes soil nutrient transfer and root growth in grassland. *Ecology Letters* 2: 357–360.
- Bass D, Stentiford GD, Wang H-C, Koskella B, Tyler CR. 2019. The pathobiome in animal and plant diseases. *Trends in Ecology & Evolution* 34: 996–1008.
- Bebber DP, Holmes T, Gurr SJ. 2014. The global spread of crop pests and pathogens. *Global Ecology and Biogeography* 23: 1398–1407.
- Bennett AE, Groten K. 2022. The costs and benefits of plant-arbuscular mycorrhizal fungal interactions. *Annual Review of Plant Biology* 73: 649–672.
- Berlinches de Gea A, Hautier Y, Geisen S. 2022. Interactive effects of global change drivers as determinants of the link between soil biodiversity and ecosystem functioning. *Global Change Biology* 29: 296–307.
- Brivio MF, Mastore M. 2018. Nematobacterial complexes and insect hosts: different weapons for the same war. *Insects* 9: 117.
- Brown AMV. 2018. Endosymbionts of plant-parasitic nematodes. *Annual Review of Phytopathology* 56: 225–242.
- Chantanao A. 1969. Transmission of plant pathogenic bacteria and a bacteriophage of *Agrobacterium tumefaciens* (Smith and Townsend) Conn. by a saprozoic nematode, *Diplogaster lheritieri* Mupas, 1919. *Dissertation Abstract* 29: 3571.
- Choe A, von Reuss SH, Kogan D, Gasser RB, Platzer EG, Schroeder FC, Sternberg PW. 2012. Ascaroside signaling is widely conserved among nematodes. *Current Biology* 22: 772–780.
- Darby C, Hsu JW, Ghori N, Falkow S. 2002. *Caenorhabditis elegans*: plague bacteria biofilm blocks food intake. *Nature* 417: 243–244.
- Decraemer W, Hunt DJ. 2006. Structure and classification. In: Perry RN, Moens M, eds. *Plant Nematology: advances in impact assessment*. Wallingford, UK: CAB International, 3–32.
- Decraemer W, Robbins RT. 2007. The who, what and where of Longidoridae and Trichodoridae. *Journal of Nematology* 39: 295–297.
- Denton CS, Bardgett RD, Cook R, Hobbs PJ. 1998. Low amounts of root herbivory positively influence the rhizosphere microbial community in a temperate grassland soil. *Soil Biology and Biochemistry* 31: 155–165.
- Dirksen P, Marsh SA, Braker I, Heidland N, Wagner S, Nakad R, Mader S, Petersen C, Kowallik V, Rosenstiel P *et al.* 2016. The native microbiome of the nematode *Caenorhabditis elegans*: gateway to a new host-microbiome model. *BMC Biology* 14: 38.
- Döring TF, Pautasso M, Frinckh MR, Wolfe MS. 2011. Concepts of plant health – reviewing and challenging the foundations of plant protection. *Plant Pathology* 61: 1–15.
- Du Preez G, Daneel M, De Goede R, Du Toit MJ, Ferris H, Fourie H, Gesien S, Kakouli-Duarte T, Korthals G, Sánchez-Moreno S *et al.* 2022. Nematode-based indices in soil ecology: application, utility, and future directions. *Soil Biology and Biochemistry* 169: 108640.
- Elhady A, Giné A, Topalovic O, Jacquiod S, Sørensen SJ, Sorribas FJ, Heuer H. 2017. Microbiomes associated with infective stages of root-knot and lesion nematodes in soil. *PLoS ONE* 12: e0177145.
- Elhady A, Hallmann J, Heuer H. 2020. Symbiosis of soybean with nitrogen fixing bacteria affected by root lesion nematodes in a density dependent manner. *Scientific Reports* 10: 1619.
- European Commission. 2022. *Proposal for a regulation of the European parliament and of the council on the sustainable use of plant protection products and amending regulation (EU) 2021/2115*.
- Fanning JP, Reeves KL, Forknall CR, McKay AC, Hollaway GJ. 2020. *Pratylenchus thornei*: the relationship between presowing nematode density and and yield loss in wheat and barley. *Phytopathology* 110: 674–683.
- Ferris H, Bongers T, De Goede RGM. 2001. A framework for soil food web diagnostics: extension of the nematode faunal analysis concept. *Applied Soil Ecology* 18: 13–29.
- Ferris H, Venette RC, Lau SS. 1997. Population energetics of bacterial-feeding nematodes: carbon and nitrogen budgets. *Soil Biology and Biochemistry* 29: 1183–1194.
- Finkel OM, Castrillo G, Herrera Paredes S, Salas González I, Dangl JL. 2017. Understanding and exploiting plant beneficial microbes. *Current Opinion in Plant Biology* 38: 155–163.
- García-Sánchez M, Souche M, Trives-Segura C, Plassard C. 2021. The grazing activity of sp. drives phytate mineralisation within its trophic relationship with bacteria. *Journal of Nematology* 53: 1–13.
- Gheysen L, Mitchum M. 2019. Phytoparasitic nematode control of plant hormone pathways. *Plant Physiology* 179: 1212–1226.
- Greco N, Di Vito M. 2009. Population dynamics and damage levels. In: Perry RN, Moens M, Starr JL, eds. *SRoot-knot nematodes*. Wallingford, UK: CAB International, 246–274.
- Grundler F, Munch A, Wyss U. 1992. The parasitic behaviour of second-stage juveniles of *Meloidogyne incognita* in roots of *Arabidopsis thaliana*. *Nematology* 38: 98–111.
- Holbein J, Grundler FMW, Siddique S. 2016. Plant basal resistance to nematodes: an update. *Journal of Experimental Botany* 67: 2049–2061.
- van den Hoogen J, Geisen S, Routh D, Ferris H, Traunspurger W, Wardle DA, De GRGM, Adams BJ, Ahmad W, Andriuzzi WS *et al.* 2019. Soil nematode abundance and functional group composition at a global scale. *Nature* 572: 194–198.
- Horiuchi J, Prithiviraj B, Bais HP, Kimball BA, Vivanco JM. 2005. Soil nematodes mediate positive interactions between legume plants and rhizobium bacteria. *Planta* 222: 848–857.
- Hubert FP. 1957. Diseases of some export crops in Indonesia. *Plant Disease Reporter* 41: 55–64.
- Ingham RE, Trofymow JA, Ingham ER, Coleman DC. 1985. Interactions of bacteria, fungi, and their nematode grazers: effects on nutrient cycling and plant growth. *Ecological Monographs* 55: 119–140.

- IPPC Secretariat. 2021. *Scientific review of the impact of climate change on plant pests – a global challenge to prevent and mitigate plant pest risks in agriculture, forestry and ecosystems*. Rome, Italy: FAO on behalf of the IPPC Secretariat. doi:10.4060/cb4769en.
- Jiang Y, Luan L, Hu K, Liu M, Chen Z, Geisen S, Chen X, Xu Q, Bonkowski M, Sun B. 2020. Trophic interactions as determinants of the arbuscular mycorrhizal fungal community with cascading plant-promoting consequences. *Microbiome* 8: 142.
- Jones JT, Haegeman A, Danchin EGJ, Gaur HS, Helder J, Jones MGK, Kikuchi T, Manzanilla-López R, Palomares-Rius JE, Wesemael WML *et al.* 2013. Top 10 plant-parasitic nematodes in molecular plant pathology. *Molecular Plant Pathology* 14: 946–961.
- Kanfra X, Liu B, Beerhues L, Sørensen SJ, Heuer H. 2018. Free-living nematodes together with associated microbes play an essential role in apple replant disease. *Frontiers in Plant Science* 9: 2679.
- Kaplan F, Alborn HT, von Reuss SH, Ajredini R, Ali JG, Akyazi F, Stelinski LL, Edison AS, Schroeder FC, Teal PE. 2012. Interspecific nematode signals regulate dispersal behavior. *PLoS ONE* 7: e38735.
- Kaplan F, Perret-Gentil A, Guirintano J, Stevens G, Erdogan H, Schiller KC, Mirti A, Sampson E, Torres C, Sun J. 2020. Conspecific and heterospecific pheromones stimulate dispersal of entomopathogenic nematodes during quiescence. *Scientific Reports* 31: 5738.
- Karimi M, van Montagu M, Gheysen G. 2000. Nematodes as vectors to introduce *Agrobacterium* into plant roots. *Molecular Plant Pathology* 1: 383–387.
- Katzen A, Chung H-K, Harbaugh WT, Della Iacono C, Jackson N, Yu SK, Flavell SW, Glimcher PW, Lockery SR. 2021. The nematode worm *C. elegans* chooses between bacterial foods exactly as if maximizing economic utility. *BioRxiv*. doi:10.1101/2021.04.25.441352.
- Khan Z, Kim YH. 2007. A review on the role of predatory soil nematodes in the biological control of plant parasitic nematodes. *Applied Soil Ecology* 35: 370–379.
- King WL, Bell TH. 2022. Can dispersal be leveraged to improve microbial inoculant success? *Trends in Biotechnology* 40: 12–21.
- Knox OGG, Killham K, Artz RRE, Mullins C, Wilson M. 2004. Effect of nematodes on rhizosphere colonization by seed-applied bacteria. *Applied and Environmental Microbiology* 70: 4666–4671.
- Koppenhöfer AM, Shapiro-Ilan DI, Hiltbold I. 2020. Entomopathogenic nematodes in sustainable food production. *Frontiers in Sustainable Food Systems* 4: 125.
- Kranz J. 2005. Interactions in pest complexes and their effects on yield. *Journal of Plant Diseases and Protection* 112: 366–385.
- Lamelas A, Desgarenes D, López-Lima D, Villain L, Alonso-Sánchez A, Artacho A, Latorre A, Moya A, Carrión G. 2020. The bacterial microbiome of *Meloidogyne*-based disease complex in coffee and tomato. *Frontiers in Plant Science* 11: 365.
- Liu T, Yu L, Xu J, Yan X, Li H, Whalen JK, Hu F. 2017. Bacterial traits and quality contribute to the diet choice and survival of bacterial-feeding nematodes. *Soil Biology and Biochemistry* 115: 467–474.
- Manohar M, Tenjo-Castano F, Chen S, Zhang YK, Kumari A, Williamson VM, Wang X, Klessir DF, Schroeder FC. 2020. Plant metabolism of nematode pheromones mediates plant-nematode interactions. *Nature Communications* 10: 208.
- Manoslava P, Manohar M, von Reuss SH, Chen S, Koch A, Kaplan F, Choe A, Micikas RJ, Wang X, Kogel K-H *et al.* 2015. Conserved nematode signaling molecules elicit plant defenses and pathogen resistance. *Nature Communications* 6: 7795.
- Masson A-S, Ho Bich H, Simonin M, Nguyen Thi H, Czernic P, Moulin L, Bellafiore S. 2020. Deep modifications of the microbiome of rice roots infected by the parasitic nematode *Meloidogyne graminicola* in highly infested fields in Vietnam. *FEMS Microbiology Ecology* 96: fiae099.
- Mohan S, Kumar KK, Sutar V, Saha S, Rowe J, Davies KG. 2020. Plant root-exudates recruit hyperparasitic bacteria of phytonematodes by altered cuticle aging: implications for biological control strategies. *Frontiers in Plant Science* 11: 763.
- Müller J. 1999. The economic importance of *Heterodera schachtii* in Europe. *Helminthologia* 36: 205–213.
- Nykyri J, Fang X, Dorati F, Bakr R, Pasanen M, Niemi O, Palva ET, Jackson RW, Pirhonen M. 2013. Evidence that nematodes may vector the soft rot-causing enterobacterial phytopathogens. *Plant Pathology* 63: 747–757.
- Oka Y. 2020. From old-generation to next-generation nematocides. *Agronomy* 10: 1387.
- Rasmann S, Köllner TG, Degenhardt J, Hiltbold I, Töpfer S, Kuhlmann U, Gershenson J, Turlings TCJ. 2005. Recruitment of entomopathogenic nematodes by insect-damaged maize roots. *Nature* 434: 732–737.
- Riascos-Ortiz D, Mosquera-Espinosa AT, De AFV, De OCMG, Muñoz-Florez JE. 2020. An integrative approach to the study of *Helicotylenchus* (Nematoda: Hoplolaimidae) Colombian and Brazilian populations associated with Musa crops. *Journal of Nematology* 52: 1–19.
- Rønn R, Vestergård M, Ekelund F. 2012. Interactions between bacteria, protozoa and nematodes in soil. *Acta Protozoologica* 51: 223–235.
- Schmidt JH, Theissen LV, Sisis A, Finckh MR. 2020. Increased resilience of peas toward root rot pathogens can be predicted by the nematode metabolic footprint. *Frontiers in Sustainable Food Systems* 6: 881520.
- Schroeder JW, Dobson A, Mangan SA, Petticord DF, Herre EA. 2020. Mutualist and pathogen traits interact to affect plant community structure in a spatially explicit model. *Nature Communications* 11: 2204.
- Siddique S, Coomer A, Baum T, Williamson VM. 2022. Recognition and response in plant–nematode interactions. *Annual Review of Phytopathology* 60: 143–162.
- Sikder MM, Vestergård M, Kyndt T, Topalovic O, Kudjordjie EN, Nicolaisen M. 2022. Genetic disruption of *Arabidopsis* secondary metabolite synthesis leads to microbiome-mediated modulation of nematode invasion. *The ISME Journal* 16: 2230–2241.
- Stirling GR. 2014. Nematodes, mites and Collembola as predators of nematodes, and the role of generalist predators. In: Stirling GR, ed. *Biological control of plant-parasitic nematodes: soil ecosystem management in sustainable agriculture, 2nd edn*. Willingford, UK: CAB International, 157–192.
- Thakur MP, Geisen S. 2019. Trophic regulations of the soil microbiome. *Trends in Microbiology* 27: 771–780.
- Tian B-Y, Cao Y, Zhang K-Q. 2015. Metagenomic insights into communities, functions of endophytes, and their associates with infection by root-knot nematode, *Meloidogyne incognita*, in tomato roots. *Scientific Reports* 5: 17087.
- Topalovic O, Bredenbruch S, Schleker ASS, Heuer H. 2020. Microbes attaching to endoparasitic phytonematodes in soil trigger plant defense upon root penetration by the nematode. *Frontiers in Plant Science* 11: 138.
- Topalovic O, Elhady A, Hallmann J, Richert-Pöggeler K, Heuer H. 2019. Bacteria isolated from the cuticle of plant-parasitic nematodes attached to and antagonized the root-knot nematode *Meloidogyne hapla*. *Scientific Reports* 9: 11477.
- Topalovic O, Vestergård M. 2021. Can microorganisms assist the survival and parasitism of plant-parasitic nematodes? *Trends in Parasitology* 37: 947–958.
- Trap J, Bonkowski M, Plassard C, Villenave C, Blanchart E. 2016. Ecological importance of soil bacterivores for ecosystem functions. *Plant and Soil* 398: 1–24.
- Wang X, Li G-H, Zou C-G, Ji X-L, Liu T, Zhao P-J, Liang L-M, Xu J-P, An Z-Q, Zheng X *et al.* 2014. Bacteria can mobilize nematode-trapping fungi to kill nematodes. *Nature Communications* 5: 5776.
- Wondafra M, van Dam NM, Tytgat TOG. 2013. Plant systemic induced responses mediate interactions between root parasitic nematodes and aboveground herbivorous insects. *Frontiers in Plant Science* 4: 87.
- Yeates GW. 1999. Effects of plants on nematode community structure. *Annual Review of Phytopathology* 37: 127–149.
- Yergaliyev TM, Alexander-Shani R, Dimerets H, Pivonia S, Bird DM, Rachmilevitch S, Szitenberg A. 2020. Bacterial community structure dynamics in *Meloidogyne incognita*-infected roots and its role in worm-microbiome interactions. *mSphere* 5: e00306-20.
- Zheng J, Dini-Andreote F, Luan L, Geisen S, Xue J, Li H, Sun B, Jiang Y. 2022. Nematode predation and competitive interactions affect microbe-mediated phosphorus dynamics. *MBio* 13: e0329321.