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Grass–endophyte symbiosis: A neglected aboveground interaction with multiple belowground consequences

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A B S T R A C T

Most terrestrial plants form symbiosis with a large variety of microorganisms that can be major determinants of morphological and physiological host traits that, ultimately, affect the structure and function of communities. Epichloae fungal endophytes (Clavicipitaceae) are widespread symbionts of grasses that asymptomatically inhabit aerial host tissues, and confer defense to herbivores and tolerance to multiple stresses. Research on symbionts of plants has focused on soil organisms such as nitrogen-fixing bacteria and mycorrhizal fungi, although recent studies suggest that endophytes of great agronomic interest can also generate major shifts in the belowground subsystem. We conducted a meta-analysis to examine the available knowledge on the effects of endophytes on the soil. Our objective was to provide a synthesis of existing data that, comprehensively and quantitatively, evaluates the different pathways by which the grass–endophyte symbiosis may affect belowground components and processes, and highlights gaps in knowledge relevant to agroecosystem management. The presence of endophyte was found to reduce root biomass and mycorrhizal colonization of plants. In turn, a significant stimulation of root exudates by endophytes was detected. Effects of endophytes on soil respiration and litter decomposition were not significant. No pattern was found in the response of the soil fauna to endophyte presence. Our review revealed that we still need to improve our understanding about the impact of endophytes on other mutualistic symbionts. The grass–endophyte symbiosis is a promising interaction that, like those involving nitrogen-fixing bacteria and mycorrhizal fungi, may become an important tool for sustainable agriculture.

1. Introduction

Virtually all terrestrial plants interact with a large variety of microorganisms, forming symbioses that may range from mutualism to parasitism (Brundrett, 2009; Douglas, 2010; Hartley and Gange, 2009; Rodriguez et al., 2009). These symbionts can be major determinants of host eco-physiology, ultimately affecting the structure and function of communities (Clay and Holah, 1999; Clay and Schardl, 2002; Douglas, 2010; Malinowski and Belesky, 2006; Omacini et al., 2001). Symbionts involving soil microorganisms are the most studied, probably because they are critical to many plants in low-nutrient environments (Douglas, 2010; Van der Heijden and Sanders, 2002). In contrast, other plant microsymbionts, such as systemic leaf fungal endophytes (Clavicipitaceae, Balansieae), have been scarcely considered. About 96% of the published articles between 1961 and 2010 provided studies on the associations of plants with nitrogen-fixing bacteria of the genus *Rhizobium* or phosphorus-providing mycorrhizal fungi of the genus *Glomus*, whereas less than 4% focused on the symbiosis between grasses and endophytes (Fig. 1). These fungi (hereafter endophytes) are widespread symbionts of cool-season grasses (Clay and Schardl, 2002; Iannone et al., 2011). Moreover, recent studies suggest their asymptomatic presence may generate shifts in multiple above- and belowground ecosystem components over both short and long time scales (Omacini et al., 2005; Rudgers and Clay, 2007; Saikkonen et al., 2006).

Many grass species form symbioses with systemic fungal endophytes of the genus *Neotyphodium* (formerly *Acremonium* sect. Albolanosa Morgan-Jones, Glenn et al., 1996). According to phylogenetic studies, the different *Neotyphodium* species have arisen independently from hybridization of sexual ancestors of genus *Epichloë* (Clay and Schardl, 2002; Iannone et al., 2009; Moon et al., 2000). Hyphae of *Neotyphodium* grow systemically among cells in sheathes of leaves, stems and reproductive host tissues without causing any symptoms of disease (Rodriguez et al., 2009). Unlike their pathogenic ancestors, *Neotyphodium* endophyte fungi are not

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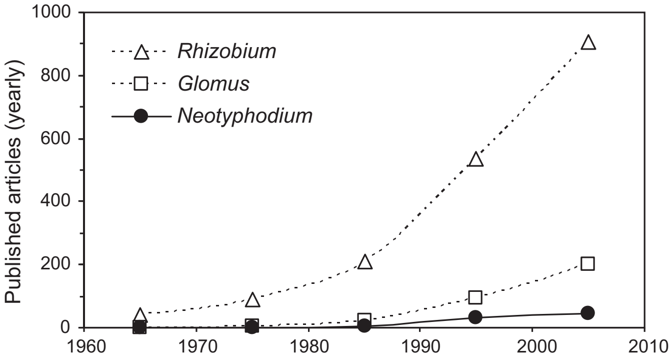

Fig. 1. Mean annual articles published between 1960 and 2011, compiled by Scopus database. Selected articles included at least one of the terms *Rhizobium* or *Bradyrhizobium*, *Glomus*, and *Neotyphodium*, which represent the most important genera of nitrogen fixing bacteria, mycorrhizae, and fungal endophytes respectively. The search on *Neotyphodium* also included Epichloe and the former nomenclatures of *Neotyphodium coenophialum* or *N. lolii* (*Acremonium coenophialum* or *A. lolii*).

capable of sexual reproduction and horizontal transmission, which means that they reproduce asexually and vertically through host grass seeds (Type III, or class I endophytes according to White, 1987, and Rodriguez et al., 2009, respectively). Thus, the grass–endophyte symbiosis appears to be essential for the fungus, while facultative for host plants. Evolutionary theory predicts that this interaction should result in a mutualism. While the host plant provides nutrition and means of propagation and dispersion to the endophytic fungus, the symbiont confers the plant with resistance to herbivory (mediated by alkaloids) and tolerance to stress (Clay and Schardl, 2002; Malinowski and Belesky, 2000, 2006; Rodriguez et al., 2009; White and Torres, 2009). Those features have triggered great agronomic interest since this endophyte is often essential for pasture persistence and productivity but is less desirable for animal production.

The first publication that revealed endophytes in grasses dates from 1904 (Freeman, 1904). However, the great agronomic interest for the grass–endophyte symbioses started around the 1970s when the toxicosis in cattle that grazed on tall fescue (*Lolium arundinaceum*, formerly named *Festuca arundinacea*) and perennial ryegrass (*Lolium perenne*) was associated with the presence of asexual endophytes (Bacon and Hill, 1997). Then, after finding that fungal alkaloids were responsible for host grass resistance to herbivores, an increasing interest has arisen in the ecological role of this defensive mutualism (Clay, 1988). Today, it is well known that endophyte presence in many weeds, invaders and forage grasses sometimes changes host biochemistry, physiology and morphology, which modify the symbiosis relationship with the biotic and abiotic environments (Clay and Schardl, 2002; Malinowski and Belesky, 2000, 2006; Rasmussen et al., 2008). Although the relationship between grass–endophyte symbiosis and aboveground components has been studied, a limited understanding of its relationship with the belowground subsystem still persists.

Understanding the impact of the interaction between grasses and endophytic fungi on fundamental ecosystem processes has become an important goal of ecological and agricultural research. The aim of this article was to critically analyze the potential pathways by which these endophytic fungi modify the relationships between host plants and soil biota. Based on a review and meta-analyses including all published studies on the interaction between cool season grasses and endophytes, this work proposes a general ecological framework to improve our knowledge on relevant soil processes mediated by endophytes. Gaps in knowledge about the implications of this symbiosis for sustainable agroecosystem management are also identified. Research on this constitutive and promising symbiosis should be encouraged. The reason for this is that the use of this strategic symbiosis may help to design environmentally friendly systems, for instance, by reducing the use of agrochemical inputs (Malinowski and Belesky, 2006; Tikhonovich and Provorov, 2007).

2. Materials and methods

To analyze how the grass–endophyte symbiosis can affect the structure and activity of belowground components of the ecosystems, the Scopus database was used to find articles and reviews that include the terms “soil”, “*Epichloë*” and “*Neotyphodium*” (or its former nomenclature *Acremonium lolii* and *A. coenophialum*). From the articles obtained, we considered those which satisfied two criteria: (1) studies in which endophyte infection was experimentally manipulated to achieve low and high infected populations or infected and non-infected individuals from an originally single population, and (2) studies in which statistical information (either in graphical or numerical form) was provided. Thus, not included in the meta-analysis were those articles that did not provide measures of error, number of replicates, or control treatments for the comparisons. Data available in graphical form were digitalized using GetData Graph Digitizer 2.24. Standard error values were converted to standard deviation from the product of SE and the square root of the number of replicates. We then, compiled a database with a total of 133 measurements (data pairs) that were grouped in the following categories: root biomass, arbuscular mycorrhizal fungi, soil fauna, root exudates, microbial respiration and litter decomposition (Table 1). It was not possible to analyze the response of *Rhizobium* sp.

A meta-analysis was carried out using Metawin 2.0 (Rosenberg et al., 2000). Effect sizes were measured using Hedges’d instead of response ratio because it is not biased by small sample sizes (Rosenberg et al., 2000). Hedges’d was calculated as,

$$d = \frac{\bar{X}^{C} - \bar{X}^{\in}}{S} \times J$$

where $\bar{X}^{C}$ and $\bar{X}^{\in}$ are control and experimental means, S is the pooled standard deviation and J is a correction factor accounting for small sample size (Rosenberg et al., 2000). Cumulative effect size, confidence intervals, and total heterogeneity (Q_t) (Hedges and Olkin, 1985) were calculated for each group of studies. Cumulative size effect was considered significant if the 95% confidence intervals did not overlap zero (Rosenberg et al., 2000). If the effect size was found to be significantly different from zero, the fail-save number (n_{fs}) was calculated as described in Rosenberg (2005). This number provides an estimation of publication bias because it estimates the number of null effect studies needed to turn results not significant. An n_{fs} greater than $5n+10$, where “n” represents the number of studies included in the analysis, is a strong indicator of the absence of a bias effect. For each category, we analyzed its total heterogeneity (Q_t). If a given categorical group displayed a significant Q_t, we performed a further subdivision of the data, whenever possible. Those categories including results obtained from a single study were not considered in the meta-analysis.

3. Results

From a total of 246 articles obtained, only 27 articles satisfied the criteria for meta-analysis. All the papers have been published during the last 20 years (Table 1). From a total of 133 data pairs obtained, 90 corresponded to *L. arundinaceum* and 16 to *L. perenne* grass species, and their endophyte-specific symbionts (Table 1). A statistically significant influence of the grass–endophyte symbiosis was found on three of the six analyzed variables (Fig. 2, Table 2).

Table 1

Published articles on the grass–endophyte symbiosis that evaluate any aspect of the belowground ecosystem. Data column indicates the number of data pairs (endophyte infected plants and endophyte-free plants) used from each study for the meta-analysis; the cross indicates that the data set is discussed but not included in the meta-analysis. Host grasses: Ah: *Agrostis hyemalis*, Bs: *Bromus setifolius*, La*: *Lolium arizonica*, La: *L. arundinaceum*, Fp: *Lolium pratensis*, Lm: *Lolium multiflorum*, Lp: *L. perenne*, Lsp: *Lolium sp*, Pa: *Poa alsodes*, Pb: *P. bonariensis*. Endophyte: Ea: *Epichloë amarillans*, Nc: *Neotyphodium coenophialum*, Nl: *Neotyphodium lolii*, No: *Neotyphodium occultans*, Nsp: *Neotyphodium* sp., *Neotyphodium uncinatum*.

Belowground component/process	Reference	Data	Grass/endophyte species
Root biomass	van Hecke et al. (2005)	1	La/Nc
	Casas et al. (2010)	1	Lm/No
	De Battista et al. (1990)	X	La/Nc
	Mack and Rudgers (2008)	1	La/Nc
	Novas et al. (2005)	X	Bs/Nsp
	Omacini et al. (2006)	2	Lm/No
	Kannadan and Rudgers (2008)	2	Pa/nsp
	Lehtonen et al. (2005)	2	Lp/Nsp
	Ren et al. (2009)	2	Lp/Nl
	Eerens et al. (1998a)	3	Lp/Nl
	Faeth et al. (2004)	1	La*/Nsp
	Matthews and Clay (2001)	X	La/Nc
	Davitt et al. (2011)	X	Ah/Ea
	Rudgers and Orr (2009)	X	La/Nc
Micorrhiza	Guo et al. (1992)	2	La/Nc
	Chu-Chou et al. (1992)	23	La/Nc
	Mack and Rudgers (2008)	1	La/Nc
	Müller (2003)	6	Lp/Nl
	Novas et al. (2005)	X	Bs/Nsp
	Novas et al. (2009)	X	Pb/Nsp
	Omacini et al. (2006)	1	Lm/No
	Popay and Jensen (2005)	2	La/Nsp
	Antunes et al. (2008)	1	La/Nc
Rhizobium	Eerens et al. (1998b)	X	Lp/Nl
Soil Fauna	Lemons et al. (2005)	5	La/Nc
	Popay and Jensen (2005)	4	La/Nsp
	Pennell et al. (2005)	1	Lsp/Nl
	Popay and Gerard (2007)	5	La/Nsp
	Chu-Chou et al. (1992)	5	La/Nc
	Eerens et al. (1998a)	3	Lp/Nl
	Humphries et al. (2001)	2	La/Nc
	Kimmons et al. (1990)	X	La/Nc
	West et al. (1988)	X	La/Nc
	Cook et al. (1991)	X	Lp/Nl
	Elmi et al. (2000)	X	La/Nc
Exudates	Malinowski et al. (1998)	4	La/Nc
	Malinowski et al. (2004)	4	La/Nc
	van Hecke et al. (2005)	2	La/Nc
	Vila Aiub et al. (2003)	6	Lm/No
	Franzluebbers and Stuedemann (2005)	X	La/Nc
	Belesky and Fedders (1995)	X	La/Nc
	Malinowski et al. (1998)	X	La/Nc
	Franzluebbers et al. (1999)	X	La/Nc
	Franzluebbers and Hill (2005)	X	La/Nc
	Franzluebbers (2006)	X	La/Nc
Decomposition	Lemons et al. (2005)	5	La/Nc
	Omacini et al. (2004)	2	Lm/No
	Siegrist et al. (2010)	2	La/Nc
Respiration	van Hecke et al. (2005)	1	La/Nc
	Casas et al. (2010)	2	Lm/No
	Franzluebbers et al. (1999)	X	La/Nc
	Franzluebbers and Stuedemann (2005)	X	La/Nc

Root biomass and mycorrhizal colonization of endophyte infected plants were 9% and 31% lower than their non-infected counterparts (Fig. 2). Non publication bias was detected for these latter variables (Fig. 2, Table 2). In both cases, variation among effect sizes of studies (Q_t) was greater than expected by sampling error, suggesting that the data set may be structured and explanatory variables should be investigated (Table 2). We searched for further subdivision of data, including host species as category, but the number of cases was too low.

No significant response of soil fauna to the endophyte presence was found. This variable encompassed a wide variety of organisms from different taxonomic groups, trophic levels and body sizes, and exhibited a significant heterogeneity (Table 2). Further subdivision in microfauna (organisms with less than 200 μm such as nematodes) and mesofauna (organisms with 200–10,000 μm such as collembola and oribatid mites) + macrofauna (the larger animals such as earthworms, myriapods) (Coleman et al., 2004) reduced the group heterogeneity of microfauna, although it did not reveal endophyte effects either.

Results showed a slight but significant stimulating effect of the symbiosis on root exudates (+9%) and a positive, although not statistically significant, effect on soil microbial respiration (+36%) (Fig. 2, Table 2). Finally, a non significant reduction (−7%) in aerial litter decomposition produced by endophyte-infected plants was found with respect to non-infected plants (Fig. 2, Table 2).

4. Discussion

This study reveals that over the past 20 years there has been significant progress in the understanding of the impact of the

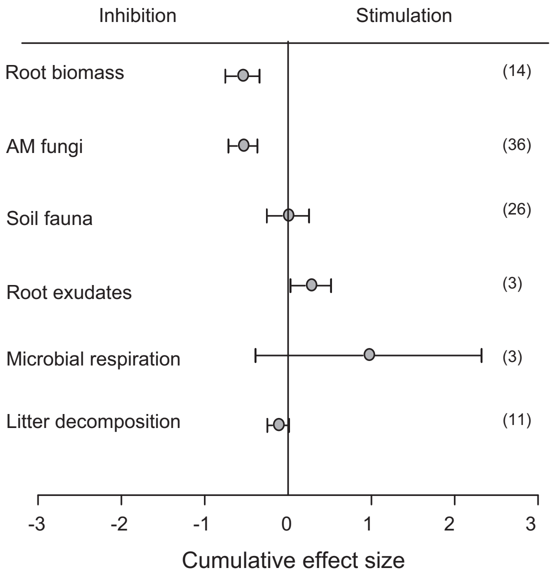

Fig. 2. Cumulative effect size of grass–endophyte symbiosis on six components/processes of the belowground ecosystem. Circles indicate mean values and bars denote the 95% confidence interval. Numbers in parentheses indicate the number of replicates for each variable.

grass–endophyte symbiosis on the components and processes of the belowground part of the ecosystem. Over the last 10 years, the volume of information generated matches that published in the 1960s for *Rhizobium*. Even though the progress achieved has allowed us to describe some patterns and identify controversial issues that still need research (Fig. 3). Our review indicates that there are significant changes in root biomass, root exudates, and mycorrhizal colonization of infected plants compared to the non-infected counterparts, suggesting that these aerial endophytes can alter the host plant rhizosphere by multiple pathways. We did not find a pattern in the response of the soil fauna to endophyte presence and neither did we detect significant changes in the rate of relevant processes such as soil microbial respiration and litter decomposition. Moreover, we were not able to evaluate other soil processes and components (e.g. nutrient mineralization, nitrogen fixation, contribution of bacteria and fungi to soil activity) due to the virtual absence of empirical data. Many questions remain unanswered largely because soil is often considered a black box in ecological experimentation and several linkages between above- and belowground subsystems are still omitted.

Table 2
Summary of the analyses performed on the effects of the grass–endophyte symbiosis on different belowground components or processes (see Fig. 2). Q_t indicates the between group heterogeneity and n_{sf} is the fail-save number (see methods).

Categorical variable	N	Q_t	n_{sf}
Root biomass	14	202.6^{*}	162
AM fungi	36	117.6^{*}	572
Soil fauna	26	43.6	
Macro/meso	15	29.9^{*}	
Micro	11	13.7	
Root exudates	3	50.8^{*}	6.1
Microbial respiration	3	2.3	
Litter decomposition	11	8.9	

* $p < 0.05$.

The lower root biomass of endophyte infected plants with respect to their non-infected counterparts was clearly an unexpected result, that seems to contradict the greater drought tolerance of infected grasses (Cheplick and Faeth, 2009; Malinowski and Belesky, 2000, 2006). We believe that longer hairs and fine roots of endophyte infected grasses might counteract the lower biomass, thus the tolerance could be explained by a different root architecture rather than biomass (Cheplick and Faeth, 2009; Malinowski and Belesky, 2000, 2006). In addition, there are other traits associated with endophyte infection (i.e., decreased stomatal conductance and osmotic adjustment) that may contribute to counteract and even overcome the lower root biomass (Malinowski and Belesky, 2000). Far from being clarified, this issue needs more research. The significant change in the compounds secreted into

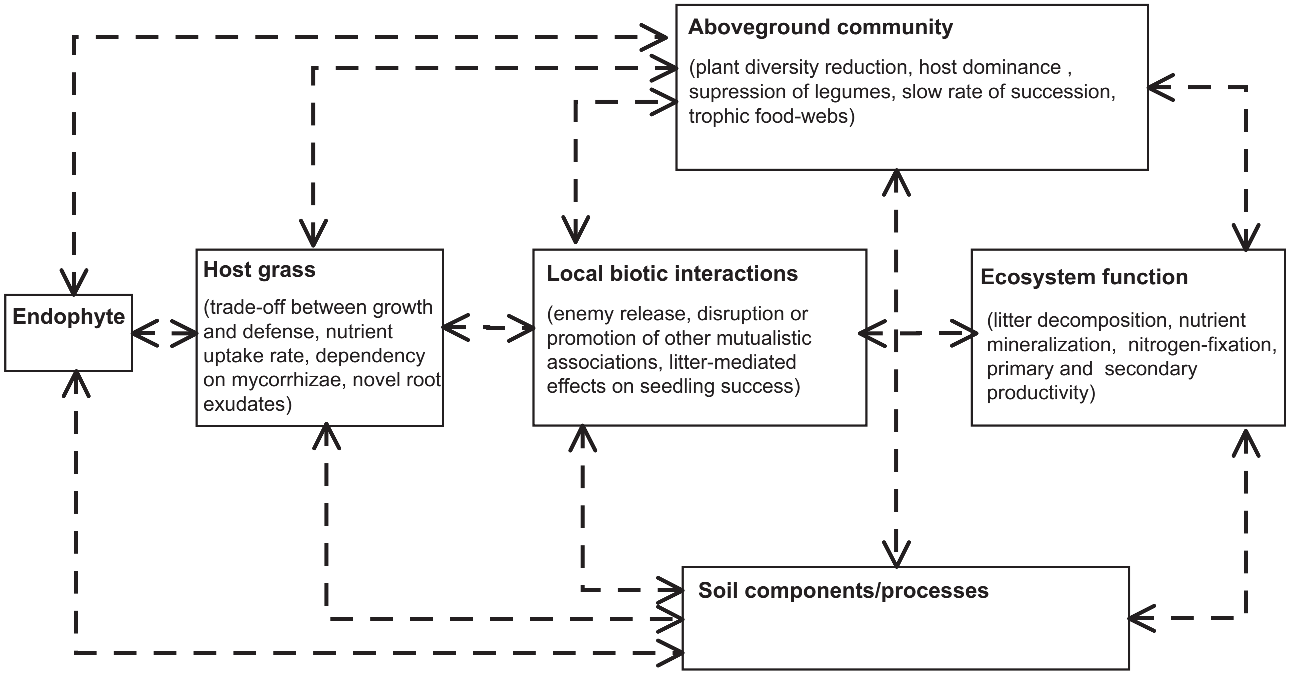

Fig. 3. Synthesis of the potential impacts of fungal endophytes on ecosystem function through the soil. Although endophytes do not cause visible symptoms, studies have recently shown that their presence may generate shifts in multiple above-and belowground ecosystem components over short and long time scales. Arrows in both directions indicate that the aboveground and belowground consequences involve feedbacks that may alter plant fitness and endophyte transmission efficiency within the population.

the soil by host plant, broadly referred to as root exudates, can be another advantage of this symbiosis (Fig. 3). For instance, it was observed that infected plants of *L. arundinaceum* show an increased ability to reduce Fe^{3+} and to bind copper in the rhizosphere (Malinowski et al., 1998, 2004), and that root exudates of infected plants stimulate soil microbial respiration (van Hecke et al., 2005). Recently, Casas et al. (2010) found that the presence of *N. occultans* in the annual grass *L. multiflorum* affected the catabolic diversity of the soil microbial community (i.e., increased the soil metabolic capacity with almost all the compounds added as substrates). Additionally, an increment in the relative contribution of fungi to soil activity was detected, which in turn can influence nutrient cycling and ecosystem processes (Casas et al., 2010).

The negative effect of the grass–endophyte symbiosis on AM fungi colonization is clearly observed in our analysis, supporting the view that the fungi would interact antagonistically (Guo et al., 1992; Omacini et al., 2006; Mack and Rudgers, 2008; Müller, 2003). The mechanisms underlying the endophyte effect on the mycorrhizal colonization are still unknown. Nonetheless there is evidence that secondary compounds from root tissues or exudates (e.g. flavonoids) act as chemical attractants or repellants in the rhizosphere (Malinowski et al., 2004; Ponce et al., 2009; Steinkellner et al., 2007). In a field survey conducted with the Argentine native grasses *Poa bonariensis*, a positive correlation between mycorrhizal colonization and endophyte infection levels was observed (Novas et al., 2009). This apparently opposite result deserves further research considering that the controls of multiple symbiotic interactions may be much more complex under natural settings and that the surveyed populations may diverge in features other than the endophyte infection level. A variety of plant responses to simultaneous infection by asexual endophytes and AM fungi has been detected (Larimer et al., 2010). Moreover, there is evidence that both fungi mediate complex interactions with cascades of effects on other trophic levels. For example, the endophyte can reduce the growth of lepidopteran larvae in the absence of AM fungi, but not when both symbionts are co-infecting the same host (Vicari et al., 2002). Furthermore, results from manipulative experiments demonstrated that the grass–endophyte symbiosis can increase the mycorrhizal colonization of co-occurring endophyte-free plants (Fig. 3, Omacini et al., 2006). Therefore, ignoring the endophyte as a component of the ecosystem may lead to misinterpretations, since it may modify the functioning of AM fungi and thus their consequences in the structure and functioning of communities.

Studies that evaluate different aspects of the interaction between soil fauna and grasses with contrasting levels of endophyte infection have demonstrated that there is a great variability in the responses. Hence, future analyses should take into account the traits or the processes of the studied organisms to find a pattern. The same conclusion has been drawn from previous authors reviewing the relationship between *Neotyphodium* endophytes and nematodes mainly considered pests (Cook and Lewis, 2001; Popay and Bonos, 2005). In spite of the existence of several studies showing negative endophyte effects on parasitic or phytophagous nematodes (e.g. Chu-Chou et al., 1992; Pedersen et al., 1988), the number of published studies to analyze those groups separately is not enough for a meta-analysis. The published literature did not allow us to establish a clear picture in the response to plant cultivar, endophyte strains, and invertebrate feeding habits. Our analysis of endophyte-mediated effects on microfauna considered the responses of nematodes from different trophic levels and life history traits. As far as could be observed, endophyte effects on plant tolerance to diseases caused by fungi, bacteria and nematodes have not been assessed as thoroughly as the same effects on insects. Analyses of endophyte impact on invertebrates from the meso and macrofauna show that their responses also vary widely and suggest that there are also relatively few studies to assess the response of different functional groups such as herbivores, carnivores or detritivores. Finally, it is commonly suggested that endophytes contribute to the invasion success of exotic grasses through enemy release and that the chemical compounds produced by endophytes are responsible for these effects (Fig. 3, Cheplick and Faeth, 2009; Rudgers and Clay, 2007). Future experimental research will illuminate the nature of these speculative mechanisms and effects attributed to the endophyte.

Our analysis revealed that the grass–endophyte symbiosis had a slight inhibiting effect on litter decomposition. Endophyte infected plants unpalatable to many herbivores can be a substrate which decomposes more slowly than the litter of non-infected plants (Lemons et al., 2005; Omacini et al., 2004; Siegrist et al., 2010). Moreover, it has been already demonstrated that endophyte presence in the previous generation of plants can alter the soil microenvironmental conditions and, consequently, the decomposition of other substrates by changing the quantity and quality of the litter on the soil surface or belowground (Omacini et al., 2004). The use of systematic review and meta-analytic methods to summarize research is limited by the scarcity of data. However, the observed effects of endophyte on the host litter quality for detritivores by different authors cannot be ignored. The same applies to the impacts of endophyte on litter persistence which can alter the availability of nutrients for the co-occurring species (Franzluebbers et al., 1999) together with biotic and abiotic conditions for germination and establishment of the subsequent plant cohort (Antunes et al., 2008; Omacini et al., 2009).

5. Synthesis and applications

Our analyses support the growing appreciation that plants mediate the interactions between above- and belowground communities, and that their symbionts can be driving forces for the diversity and function of the whole ecosystem (Bever, 1994; van der Putten et al., 2009; Wardle, 2002; Wardle et al., 2004). We have identified multiple endophyte impacts on the soil, although we still need to understand the interaction with other common plant symbionts (e.g. mycorrhizal fungi, nitrogen-fixing bacteria), pathogens and herbivores. It is remarkable that endophytes have so far been neglected in the research, particularly in pastoral systems where dependence on soil symbionts grasses and legumes is promoted. The ecological linkages between microbial symbionts of neighboring plants promise a myriad of responses that will enhance our understanding of the regulation and functional significance of biodiversity and of the impacts of human-induced global change phenomena.

The review showed that the literature is strongly taxonomically biased toward certain host species (i.e., tall fescue and perennial ryegrass) and that many others, especially native and annual species are little studied. Other authors have already discussed the implications of this historical development of grass–endophyte symbiosis research (Rudgers and Clay, 2007; Saikkonen et al., 2006). Increased experimentation with other species under different ecological contexts will enhance understanding of the mechanisms involved in endophyte effects on above- and belowground processes and on other microsymbionts in the community. The grass–endophyte symbiosis still lacks a clear and strong identity among ecologists working on symbiosis or mutualism in general (e.g. Douglas, 2010; Kiers et al., 2010), or on plant-mediated interactions in particular (e.g. Heil, 2011; Johnson, 2011).

The increased use of agrochemicals in agronomical practices and the environmental impact of production on human health have encouraged scientists to find more sustainable alternatives in the management of agroecosystems (Malinowski and Belesky, 2006; Tikhonovich and Provorov, 2007). Thus, for example, knowledge has been generated that allowed the development of tools for biological pest control, symbiotic fixation of nutrients, microbial growth promoters and others. In this sense, being able to increase the natural benefits of *Neotyphodium* endophytes' presence appears to be a challenging alternative. Great efforts are underway to find genotypes, or modify the existing ones, in order to optimize endophyte use and plant productivity (Easton, 2007). Similarly, the existence of natural variability in the profile of alkaloids produced by endophyte according to the strain, the host grass genotype or the environment, has led to the inclusion of this symbiont in plants to improve turf and ornamental grasses (Easton, 2007) or for phytoremediation of metal-polluted soils (Ren et al., 2011). It is necessary to expand our knowledge about the factors that control desirable and undesirable effects of endophyte on the ecosystem as well as symbiosis persistence.

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