bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

1 **Title**: Ectomycorrhizas accelerate decomposition to a greater extent than arbuscular mycorrhizas
2 in a northern deciduous forest
3
4 **Authors**: Alexis Carteron*, Fabien Cichonski & Etienne Laliberté
5
6 Institut de recherche en biologie végétale, Département de sciences biologiques, Université de
7 Montréal, Montréal, Canada
8
9 *Corresponding author: E-mail: alexis.carteron@umontreal.ca

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

Abstract

It has been proposed that ectomycorrhizal (EcM) fungi slow down decomposition by competing with free-living saprotrophs for organic nutrients and other soil resources (known as the "Gadgil effect"), thereby increasing soil carbon sequestration. As such, this Gadgil effect should depend on soil organic matter age and quality, but this remains unstudied. In addition, the Gadgil effect is not expected to occur in arbuscular mycorrhizal (AM) forests since AM fungi cannot access directly nutrients from soil organic matter, yet few direct comparisons between EcM and AM forests have been made. We performed a two-year reciprocal decomposition experiment of soil organic horizons (litter - L, fragmented - F, humic - H) in adjacent temperate deciduous forests dominated by EcM or AM trees. Litterbags were made of different mesh sizes allowing or excluding ingrowth of external fungal hyphae, which are primarily mycorrhizal in these forests other than for the most-recent superficial litter horizon. As expected, organic matter originating from deeper horizons and from EcM forests was of lower quality (e.g. higher lignin to nitrogen ratios) and decomposed more slowly. However, contrary to the Gadgil effect, organic matter exposed to external fungal hyphae (i.e. primarily mycorrhizal) actually decomposed faster in both forest types, and this effect was strongest in EcM forests, particularly in the F horizon. Unexpectedly, organic matter decomposition was faster in EcM than in AM forests, regardless of organic matter origin. Overall, our study reinforces the view that temperate EcM forests store greater amounts of soil organic carbon than AM forests, but suggests that this is due to factors other than the Gadgil effect.

Keywords

Organic matter decomposition; carbon cycle, nitrogen cycle, temperate forest, vertical segregation; Gadgil effect; mycorrhizal fungi; *Acer saccharum*; *Fagus grandifolia*

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

Introduction

Forests cover much of the land surface, and represent the largest terrestrial carbon (C) pool globally (Dixon and others 1994; Baldrian 2017). A majority of that C is stored in forest soils, especially in northern forests (Lal 2005; Crowther and others 2019). Soil C storage is controlled by many abiotic and biotic factors such as climate, vegetation, topography and nutrient availability that interact together (Averill and others 2014; Carvalhais and others 2014; Wiesmeier and others 2019). However, belowground biotic factors, such as microorganisms, also play an important role, directly influencing soil C inputs (i.e. litter quantity and quality) and outputs (i.e. decomposition) (Schimel and Schaeffer 2012). For example, soil microorganisms such as fungi can produce recalcitrant organic matter that decomposes slowly or they can produce extracellular enzymes that break down organic matter (Frey 2019). As a result, soil fungi play a major role in forest C cycling (Kubartová and others 2008; Bardgett and Wardle 2010; Orwin and others 2011).

A long-standing hypothesis about the effects of fungi on the soil C cycle is the “Gagdil effect” (Gadgil and Gadgil 1971; Fernandez and Kennedy 2016). This hypothesis suggests ectomycorrhizal (EcM) fungi slow down litter decomposition, potentially due to competition between EcM fungi and free-living saprotrophs for organic nutrients. Because EcM fungi acquire their C in highly labile form via plant hosts (Smith and Read 2008) in exchange for nutrients such as nitrogen and phosphorus, they would leave behind C-rich but nutrient-poor organic matter, potentially favoring soil C accumulation (Read and others 2004; Averill and others 2014). On the other hand, some EcM fungi have the capacity to oxidize organic matter, directly influencing decomposition and indirectly influencing saprotrophic organisms (Lindahl and Tunlid 2015;

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

Verbruggen and others 2017). Saprotrophic fungi could also be impacted by EcM fungi through mycoparatism, antibiosis and alteration of abiotic conditions (Fernandez and Kennedy 2016; Zak and others 2019). The Gagdil effect has only been supported by a few studies but seems to be largely context dependent, for example to litter quality (Smith and Wan 2019) and moisture level (Koide and Wu 2003). Compared to EcM fungi, it is considered that arbuscular mycorrhizal (AM) fungi lack the capacity to produce enzymes that break down organic matter (Tisserant and others 2013; Tedersoo and Bahram 2019). AM fungi would not compete directly with saprotrophic fungi, therefore it is expected that decomposition would be quicker in AM forests compared to EcM forests, but this still remains an open question (Fernandez and Kennedy 2016; Frey 2019). In fact, AM fungi may even enhance directly organic matter decomposition in some cases via a "priming effect", promoting the activity of free-living saprotrophs (Hodge 2017; Frey 2019). A better understanding of the roles that different mycorrhizal types play in organic matter decomposition is thus needed.

Because fungal types and taxa differ strongly in their vertical distribution, especially in well-stratified soil such as podzols (Dickie and others 2002; Rosling and others 2003; Bahram and others 2015), the strength and direction of the Gadgil effect could vary across soil organic horizons, yet most previous studies have only considered the uppermost litter layer. Strong vertical segregation of fungal guilds occurs across podzol profile: saprotrophic fungi dominate the litter horizon, and can still be abundant in upper organic horizons where mycorrhizal fungi increasingly dominate (Lindahl and others 2007; Clemmensen and others 2015; Carteron and others 2020). It is recognized that overlapping niches between different groups of fungi can generate competition for soil resources (Bödeker and others 2016; Mujic and others 2016). Therefore, the greatest potential for mycorrhizal fungi to inhibit saprotrophs, and thus slow down

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

82 organic matter decomposition, should lie just below the layer of fresh litter. It has been suggested
83 that these interactions might help to explain differences in the amount and vertical distributions
84 of soil C in EcM systems (Clemmensen and others 2013; Kyaschenko and others 2017) and
85 between EcM- and AM-dominated forests at different depth or horizons (Phillips and others
86 2013; Soudzilovskaia and others 2015; Craig and others 2018). By competing with saprotrophs
87 for organic nutrients, EcM fungi may promote C accumulation more than AM fungi that cannot
88 directly access these resources. These vertically segregated interactions among fungal guilds need
89 to be better understood because they play an important role in regulating organic matter
90 accumulation (Frey 2019).
91
92 Local adaptation to microbial guilds based on soil properties could also be an important factor
93 influencing decomposition via what has been termed the “home-field advantage” (HFA; van der
94 Wal and others 2013) hypothesis. This HFA predicts that litter decomposition is faster in “home
95 soils” due to adaptation of the decomposer community to the chemical composition of the “home
96 litter” (Gholz and others 2000; Austin and others 2014). Using published data on mass loss from
97 125 reciprocal litter transplants, Veen and others (2015) have shown that this HFA increases
98 decomposition rates by 7.5% on average. However, the strength of the HFA might depend on the
99 context such as plant identity, litter quality and moisture level (Veen and others 2015; Wang and
100 others 2020). Some studies suggest that AM litter shows higher HFA than EcM litter (Midgley
101 and others 2015; Jacobs and others 2018). On the other hand, because EcM litter tends to be more
102 recalcitrant (Keller and Phillips 2019), it could be expected that EcM litter decays faster in EcM
103 forest with saprotrophs better adapted to decompose recalcitrant organic matter. In any case,
104 further investigation is needed to better understand the effect of microbial decomposers driving

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

105 the HFA depending on litter type and the stage of litter decomposition (Li and others 2020; Lin
106 and others 2020).
107
108 The main objective of our study was to assess the impact of EcM and AM strategies on the
109 decomposition of soil organic matter in organic horizons in northern forests. First, we determined
110 stocks of C and nutrients in the upper 20 cm of soil in adjacent forest plots dominated by AM or
111 EcM trees. Then, we performed a litterbag experiment using a reciprocal transplant of organic
112 matter from AM and EcM forest enabling us to isolate site *vs.* organic matter quality effects on
113 decomposition. Litterbags were composed of different mesh size that allowed (44 µm) or
114 excluded (1 µm) ingrowth of fungal hyphae. It is well established that fungal hyphae can move
115 freely across large pore-size (30-50 µm) mesh but prevents in-growth of roots, while small pore-
116 size (0.5-1 µm) mesh further blocks fungal hyphae (Johnson and others 2001; He and others
117 2004; Teste and others 2009). Since mycorrhizal fungi development requires association with
118 living plant roots, only saprotrophs can develop inside the small-mesh litterbags and access
119 organic matter. Mycorrhizal fungi can colonize organic-rich soil (Lindahl and others 2007; Bunn
120 and others 2019) and, can even be abundant in organic horizons (this system; see Carteron and
121 others 2020). Therefore, large-mesh litterbags allow to follow the decomposition of organic
122 matter in the presence of external fungal hyphae, which are primarily mycorrhizal in these forests
123 other than for the most-recent superficial litter horizon where saprotrophs dominate (Carteron and
124 others 2020). Decomposition of the three upper organic horizons (litter - L, fragmented - F,
125 humic - H) was followed by measuring changes in soil mass, and changes in C and nitrogen (N)
126 over two years. In addition, the fate of C fractions was followed in decomposing L samples and
127 potential access of N by mycorrhiza in the F samples. We hypothesized that the impact of
128 mycorrhizas on organic matter decomposition would differ between AM and EcM forests. More

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

129 specifically, we expected based on the Gadgil effect hypothesis that EcM forests would store a
130 higher amount of C in the topsoil and show slower organic matter decomposition due to the
131 inhibition of saprotrophs by EcM fungi and lower litter quality, whereas these effects would be
132 smaller in AM forests. In addition, we hypothesized that the slowing down of C cycle by EcM
133 fungi would be strongest in the fragmented (F) horizon where litter-derived organic materials,
134 free-living saprotrophs, mycorrhizal fungi and roots coincide (Clemmensen and others 2013;
135 Cotrufo and others 2015; Carteron and others 2020). Due to microbial adaptations of the
136 decomposer community, we also hypothesized that litter would decompose fastest in their
137 "home" forests relative to "away" forests (Veen and others 2015). Specifically, mass loss of
138 organic matter from AM soil would be highest when incubated in AM forest and mass loss of
139 EcM organic matter highest in EcM forest.
140
141 **Material and methods**
142 *Study area and site selection*
143 Our study was conducted in a northern temperate forest at the Université de Montréal's field
144 station (Station de biologie des Laurentides, Saint-Hippolyte, Québec, Canada). The mean annual
145 temperature is 4.3 °C and total annual precipitation is 1195 mm, with ~25% falling as snow
146 (based on 1981–2010 data, meteorological station #7037310, Saint-Hippolyte). Soils consist of
147 podzols with moder humus formed from Precambrian anorthosite (Bélanger and others 2004;
148 Courchesne and others 2005). We selected ten 20 m × 20 m plots from Carteron and others
149 (2020), either dominated by EcM or AM trees (Table S1), and grouped into five clusters or
150 "blocks" (n = 5 blocks, each containing one plot of each of the two mycorrhizal types, EcM and
151 AM). These pairs of EcM-AM sites were clustered together to minimize variation in
152 environmental conditions (e.g. slope, aspect, elevation) within each block. Previous root

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

colonization and molecular analyses on the same sites showed that forests dominated by EcM trees had the highest EcM fungal abundances while forests dominated by AM trees had the highest AM fungal abundances. Carteron and others (2020) also found strong shifts from saprotrophic to mycorrhizal fungal dominance with increasing soil depth in both forest types, especially across surface organic horizons.

Soil carbon and nutrient stocks

Carbon and nutrient stocks were quantified by measuring C, N, phosphorus (P) concentrations and thickness for all horizons in the upper 20 cm of soil, as reported in Carteron and others (2020). Soil bulk density was measured simultaneously for the five horizons in three randomly-positioned locations replicates per plot using an auger, and values from these locations were averaged across sites. The horizons considered were litter (L), fragmented (F), humic (H), and mineral horizons Ae and B.

Organic matter collection

In each plot, organic matter samples were collected separately from the three organic horizons, namely: L, F and young H (i.e. most recent layer) from two pits. Samples were homogenized by horizon within each plot. Samples were collected in July 2016. A subsample from each horizon by plot was preserved at 4 °C as inoculum (see below). Another subsample was oven-dried at 60 °C for 72 h and ground for chemical analyses. The rest of the organic matter was air-dried before being used to fill the bags.

Litterbag design

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

Litterbags were 15 cm × 15 cm in size and designed to have three compartments (L, F, H; in the same order in which they occur through the soil profile) separated by 44 µm-pore polyethylene mesh (PETEX® 07-40/12; Sefar Inc., Buffalo, NY, USA). Our use of 44 µm-pore mesh ensured that hyphae could grow across compartments within each bag, an important process for decomposition (i.e. to allow for translocation of nutrients and C across horizons), while still keeping L, F, and H horizons separate for later retrieval. The outer mesh of the litterbags was made with either the same 44 µm-pore polyethylene mesh described above or 1 µm-pore mesh from the same material (PETEX® 07-1/2; Sefar Inc., Buffalo, NY, USA). Large pore size (30-50 µm) mesh has been widely used to assess the effect of mycorrhizal hyphal colonization since more than a decade ago (Johnson and others 2001), by excluding fine roots but not fungal hyphae (He and others 2004; Teste 2008). Thus, our litterbags made with 44-µm pore size mesh allow to study decomposition in the presence of mycorrhizal hyphae (and other saprotrophic fungi located outside of the bag). By contrast, the small 1-µm pore size mesh prevents most external fungal hyphae to grow through the litterbag (Teste and others 2006). Because most mycorrhizal hyphae cannot grow within the bag (as mycorrhizal fungi are obligate biotrophs), this bag design allows us to study organic matter decomposition in the absence of mycorrhizal fungi. Litterbags of 50 µm-pore size mesh have been found to allow ingrowth of mycorrhizal fungi (i.e. Teste and others 2006; Sterkenburg and others 2018), which are abundant in our F and H horizons of our plots (Carteron and others 2020). By contrast, free-living saprotrophic fungi should be present in all bags (of 1-µm and 44-µm pore sizes) since all bags were inoculated with horizon-specific organic matter from the same plot prior to being installed in the field. For this reason, while we recognize that the 1-µm mesh bags exclude all external hyphal ingrowth (mycorrhizal and free-living), for simplicity we refer to this treatment as “mycorrhizal exclusion” hereafter since 1-µm mesh bags

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

exclude this particular fungal guild. Mycorrhizal fungal hyphae should be present in the 44-µm mesh bags, being a very important component of the fungal community in the soil other than for the L horizon (Carteron and others 2020). Polyethylene mesh was selected over nylon mesh (e.g. NITEX®, Sefar Inc. Buffalo, NY, USA) because it is much more resistant to degradation when buried in soil (Colin and others 1981). Microscope observations showed no evidence that the bags were breached after two years. Previous studies using 0.5 µm-pore size mesh litter bags found that environmental conditions, particularly moisture levels, were similar inside and outside the bags (Allison and others 2013) and, that soil water moved freely across the mesh within minutes (Teste and others 2009). Our own observations confirmed that water moved freely across membranes of both mesh size via capillary action as long as there was contact between the litter inside the bag and the membrane itself; such conditions were maintained throughout the field experiment since the litterbags were buried under the litter layer and secured firmly on the ground. The 1 and 44 µm-pore size mesh have air permeability values of > 95 ±15 $l.(m^2.s)^{-1}$ at 200 Pa (provided by Sefar inc.). In total, 160 litterbags were used (Fig. S1), from which 90 prevented most hyphal ingrowth of all external fungi. Each bag was stored within a 1-mm mesh nylon bag to provide additional physical protection for the less robust 44- or 1-µm PETEX® mesh.

Litterbag preparation and collection

Weighed air-dry organic matter was transferred to litterbags (2.85 g for L and 4.75 g for F and H horizons). Horizon specific fresh inoculum (~5 % of dry-weight equivalent) was added to each horizon from the receiving plot to ensure that plot-specific microbial biota, including free-living saprotrophic fungi, could colonize each litterbag. Water content was determined from oven-dried

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

sub-samples at 60 °C for dry-mass inoculum conversion. Filled litterbags were put back *in situ* October 2016, directly on top of the H horizon (with L horizon facing up) and covered by a thin layer of fresh litter. Litterbags were secured on the ground with small stakes and tied together with nylon fishing line to a central stake to facilitate retrieval of bags. Two spatial replicates within each plot were installed. A total of 160 bags were collected after one and two years of residence (i.e. field incubation) for 480 samples analyzed (Fig. S1).

Soil analysis

Initial subsamples of ground horizons L, F and H were weighed (5.0, 6.0 and 7.0 mg ± 0.2 respectively) and analyzed to estimate C and N contents by dry combustion in a CN analyzer (Vario Micro Cube; Elementar, New-Jersey, United States, dx.doi.org/10.17504/protocols.io.udces2w). The concentrations of soluble cell contents (e.g. non-structural carbohydrates), hemicellulose, cellulose and lignin (% dry weight) were also determined on these initial samples by sequential digestion (Fiber Analyzer 200; ANKOM technology, dx.doi.org/10.17504/protocols.io.yinfude). After one and two years, organic matter samples were retrieved from litterbags, oven-dried at 60 °C for at least 72 h and then weighed to estimate mass loss percentage. These samples were then ground with a cyclone mill (Cyclone Sample Mills, UDY Corporation, Colorado, United States), using a 2-mm screen. Concentrations of C and N were also determined using the method described above. Thirty subsamples of the initial horizons, and all the F horizons after two years of residence were analyzed for $\delta^{15}N$ with a Micromass model Isoprime 100 isotope ratio mass spectrometer coupled to an Elementar Vario MicroCube elemental analyser in continuous flow mode.

Statistical analyses

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

Differences in organic matter stocks among forest types were evaluated using a linear mixed-effects model with forest type (AM or EcM as soil provenance) as a fixed factor and block as a random factor. Horizon was added as fixed factor for the modeling of initial soil chemistry. To predict the changes in mass (within the litterbags), linear mixed-effects models were also used by adding as fixed factors outside fungal hyphae (i.e. size of mesh pore) excluded (1 µm) or not (44 µm). Finally, forest of residence (AM or EcM forest) and time (one or two years) were added as fixed factors to compare decomposition in the two forest types including relevant interactions among fixed factors (see Table S2 for more details). Models were compared using the Akaike information criterion corrected for small sample size (AIC_c). Validation of the models was done by visual inspection of the residuals. Spatial replicates within one plot were averaged prior to analyses. Eleven bags with damaged mesh were removed from the analysis. Statistical analyses were performed using the R software (R Core Team 2018) and the following packages *dplyr* (Wickham and others 2017), *emmeans* (Lenth 2019), *ggplot2* (Wickham 2016), *ggpubr* (Kassambara 2018), *nlme* (Pinheiro and others 2012). Data and R scripts can be found at https://github.com/alexiscarter/decompo_myco.

Results

Organic matter stocks

Stocks of C were higher in EcM forest stands compared to AM stand within the upper 20 cm of soil (one-way analysis of variance, $P < 0.001$; Fig. S2) as observed in the organic horizons (Table 1). Stands dominated by EcM trees stored 14% more C than AM stands in surface soils. The soil C:N ratio also differed among forest types, with higher values in EcM stands ($P = 0.024$; Fig. S3). By contrast, there were no differences in soil C:P ratio among forest types (Fig. S4).

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

Initial soil chemistry

Soil C, cellulose and hemicellulose concentrations decreased from L to H horizons in both forest types, while lignin was highest in the F horizon and in EcM stands overall (23% in AM forest and 26 % in EcM forest; Table S3). By contrast, soil total [N] increased slightly with soil depth in both forest types (Table S3). As a result, soil C:N and lignin:N ratios were higher in EcM forest for the three organic horizons compared to AM forest (Table S3). $\delta^{15}N$ values showed similar increases from L to H horizons in both forest types but the F horizon in EcM forest was slightly enriched (but not significantly, $P = 0.224$; Table 1). Horizons tended to be thicker in EcM forest (Table 1).

Effect of residence on decomposition: AM vs. EcM forests

In AM and EcM stands, older (i.e. deeper) horizons decomposed more slowly than younger ones (Fig. 1). Organic matter loss was slower in the litterbags of 1 µm-pore size mesh in all horizons of both types of forests. However, the slowing down of decomposition due to mycorrhizal fungal exclusion was only statistically significant in the F horizons in stands dominated by AM (-3.7 %, $P = 0.02$; Fig. 1a) and EcM (-4.4 %, $P = 0.019$; Fig. 1b). Differences in the effects of the mycorrhizal exclusion treatments among forest types increased between one and two years of incubation (Fig. 2). Overall, decomposition was slower in AM compared to EcM stands, ranging from -0.8% ($P > 0.05$) of mass loss after one year to -3% ($P < 0.001$) after two years of incubation. After two years, decomposition of organic matter originating from EcM and AM soils was higher in EcM stands (Fig. 2).

Effect of provenance: AM vs. EcM forests

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

293 Litter originating from AM stands decomposed more quickly than EcM litter after one year but
294 this was no longer the case after two years ($P > 0.05$; Fig. S5). While changes in the litter C:N
295 ratio remained similarly low for both soil origins (~1, Fig. S6), the EcM litter showed a clear
296 increase (~1.2) suggesting a lower loss in N compared to C. Similarly, changes in lignin:N ratio
297 were also stronger for EcM litter (Fig. S7).
298
299 *Effect of provenance and residence on C fractions and N*
300 Mycorrhizal exclusion did not affect concentrations of soluble contents nor hemicellulose in litter
301 incubated in AM and EcM stands. Compared to EcM stands, decomposition was slower in AM
302 stands for litter cellulose (-4.12 %, $P = 0.001$; Fig. S8) and lignin (-6.56 but $P > 0.05$; Fig. S9).
303 Mycorrhizal exclusion slowed down decomposition of cellulose (-3.79 %, $P = 0.003$) and lignin
304 (-5.13% but $P > 0.05$). Overall, N loss was reduced by mycorrhizal exclusion (-2.83 %, $P <$
305 0.001) and reduced in AM stands (+2.7 %, $P < 0.001$). After two years, ^{15}N enrichment was
306 higher in EcM stands (F horizons only, $P = 0.046$) but effect of mycorrhizal exclusion was rather
307 low (Fig. S10).
308
309 **Discussion**
310 No evidence of a Gadgil effect in either forest mycorrhizal type was observed. In fact, the
311 opposite effect was observed, in that decomposition was faster in the presence of EcM or AM
312 fungi than in their absence. Contrary to our hypothesis, decomposition was faster in EcM- than in
313 AM-dominated forests. However, as predicted, decomposition was higher in upper horizons (i.e.
314 “younger” soil), and the net effect of the external fungal network on decomposition was
315 significant in the fragmented (F) horizons. The F horizon is located just below the litter (L),
316 where most decomposition studies tend to focus. Our results suggest that the Gadgil effect is not

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

317 a universal pattern in EcM forests, and that mycorrhizal fungi may actually accelerate rather than
318 slow down decomposition (Frey 2019). In agreement with these results, we found that
319 decomposition was faster in EcM forests regardless of organic matter origin, suggesting an HFA
320 in EcM but not AM forests.
321
322 Several abiotic and biotic factors can impact litter decomposition, such as climate and soil fauna
323 (Hättenschwiler and others 2005; Steidinger and others 2019). However, given the importance of
324 fungi in soil decomposition processes, there has been much interest in exploring the potential
325 effects of interguild fungal interactions over C and nutrient dynamics (Dighton and others 1987;
326 Verbruggen and others 2017). Mycorrhizal fungi can inhibit saprotrophs by competing for
327 nutrients, resulting in slower organic matter decomposition and promotion of C accumulation
328 (Frey 2019). We took advantage of a natural experiment of co-occurring patches of AM and EcM
329 trees under similar environmental conditions but distinct fungal communities and soil chemistry
330 (Carteron and others 2020) to test if contrasting mycorrhizal strategies exerted different control
331 on organic matter decomposition (Phillips and others 2013; Dickie and others 2014). However,
332 contrary to the Gadgil effect hypothesis, our results showed that both EcM and AM fungi
333 accelerate organic matter decomposition in this northern deciduous forest. This might occur if the
334 overall positive effect of mycorrhizal hyphae and other external fungi on decomposition was
335 greater than any potential negative impacts of competition with saprotrophs. In addition,
336 mycorrhizal fungi combined with their local microbial community in EcM forests tended to
337 degrade cellulose and lignin more quickly compared to AM forests. By isolating the effect of
338 mycorrhizas, microbial communities and local environmental conditions, our study shows that
339 decomposition tends to be higher in EcM than AM forests regardless of soil origin and incubation
340 time. Our results challenge the view that EcM fungi slow down litter and soil decomposition

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

341 compared to AM fungi (Tedersoo and Bahram 2019 and references therein). They also suggest
342 that more attention should be paid to priming *vs.* inhibitory effects of different mycorrhizal types
343 on the decomposition of organic matter (Kuzyakov 2010).
344
345 Ectomycorrhizal fungi have traditionally been suggested to slow down litter decomposition via
346 their negative competitive effects on free-living saprotrophs (Gadgil and Gadgil 1971; Fernandez
347 and Kennedy 2016). In our field experiment, we have found that EcM fungi in fact accelerated
348 the decomposition across the three upper organic horizons over two years, particularly in the
349 fragmented (F) horizon. Fernandez & Kennedy (2016) suggested a number of important
350 environmental factors that could modulate the inhibiting impact of EcM fungi on free-living
351 saprotrophs, which might help to explain our results. First, organic matter recalcitrance was
352 relatively low in this broadleaf forest, with lignin:N ratios below 20. Similarly, the C:N ratio was
353 below 30, making N less limiting for saprotrophs compared to other studies where a Gadgil effect
354 was observed (Smith and Wan 2019). Secondly, the studied podzols were well-stratified and
355 exhibited a strong vertical segregation with distinct fungal communities with strong shifts from
356 saprotrophic to mycorrhizal fungal dominance with increasing soil depth (Carteron and others
357 2020), thereby reducing opportunities for interguild competition. Finally, decreased soil moisture
358 due to EcM fungi can impede decomposition processes (Koide and Wu 2003), but our system is
359 located in a northern temperate forest characterized by a humid continental climate with
360 precipitations throughout the year, where water is not thought to be limiting. At least three other
361 experimental studies have found a positive combined effect of roots and EcM fungi on
362 decomposition (Zhu and Ehrenfeld 1996; Subke and others 2011; Malik 2019). In our case, it is
363 worth noting that the strongest positive net effect was observed in the fragmented horizon where
364 there are: (i) High root colonization by EcM and AM fungi, (ii) high abundance of saprotrophic

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

365 and mycorrhizal fungi and (iii) high fine root density (Carteron and others 2020). Most studies
366 that have studied the impact of mycorrhizas on decomposition have focus on the most recent litter
367 (L) layer, whereas here we show that important processes occur in deeper (organic) horizons. Our
368 results suggest that vertical stratification should be taken into account to better understand the
369 effect of mycorrhizas on the decomposition process.
370
371 Arbuscular mycorrhizal fungi are known to produce compounds that can, for example, alter
372 microbial community or promote soil aggregation thus modulating decomposition rate (e.g.
373 Hodge and others 2001; Gui and others 2017; Xu and others 2018). Decomposition can even be
374 reduced by AM fungi, potentially through antagonistic interactions with free-living saprotrophs
375 (Leifheit and others 2015; Carrillo and others 2016). In our field experiment, we found no
376 evidence of a Gadgil effect exerted by AM fungi that would counterbalance their positive impacts
377 on decomposition. As expected, decomposition in the upper three organic horizons in AM forest
378 was not reduced with the 1-µm mesh bags (in which mycorrhizal hyphae were excluded), but in
379 fact tended to increase. Given that AM fungi lack a strong degradation machinery (Tedersoo and
380 Bahram 2019), our results support the view that priming of organic matter decomposition might
381 be an important nutrient acquisition strategy for them (Wurzburger and Brookshire 2017).
382 Greater priming in AM systems may result from AM fungal necromass and the lack of genetic
383 capacity from AM fungi to directly access organic nutrients (Frey 2019). It is worth noting that
384 with the 1-µm mesh bags, decomposition in AM forests tended to be slower than in EcM forests,
385 suggesting that their free-living saprotrophic communities have different capacities to degrade
386 organic matter (see results after two years in Fig. 2). Microbial communities in AM forest might
387 be less efficient at degrading organic matter due to their more easily-decomposed litter contrary
388 to what has been observed for other AM systems in microcosm experiments (Taylor and others

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

2016). Evaluating the response of the saprotrophic community using molecular tools over long-term experiment would be an interesting way to better understand decomposition processes *in situ*, in order to complement studies that focus on laboratory manipulations of mycorrhizal abundance (Verbruggen and others 2017). It would also allow us to experimentally assess if the abundance of saprotrophs shifts in deeper horizons when AM and EcM fungi are excluded (e.g. Lindahl and others 2010; Sietiö and others 2019).

Leaf litter decomposition rates are known to be positively linked with initial N concentration and inversely with lignin (Prescott 2005; Berg and McClaugherty 2014). Overall net effect of mycorrhizas over decomposition is known to be controlled by substrate quality and local microbial community composition (Fernandez and others 2019; Smith and Wan 2019). As expected, we found a strong effect of soil depth with deeper (i.e. “older”) horizons decomposing more slowly. In general, the litter of EcM-associated trees tend to have a lower quality than AM trees such as lower C:N and lignin:N ratios (Lin and others 2017), which may drive soil C accumulation in the short-term. In our field sites, litter in EcM stands had lower quality compared to AM stands. The EcM stands were mostly composed of American beech. However, American beech litter is less recalcitrant than many conifers (Moore and others 1999), which may explain discrepancies with other studies from coniferous EcM forests in which Gadgil effects have been observed (Fernandez and Kennedy 2016; Smith and Wan 2019). In temperate forests, AM plants tend to produce leaf litter that decomposes more rapidly *in situ* than that of EcM plants (Keller and Phillips 2019). Similarly, litter originating from AM patches decomposed more quickly than EcM litter after one year but interestingly, this was not the case after two years in our experiment. This is consistent with previous studies showing that sugar maple leaf litter tend to decomposes more quickly during the first years after senescence (McHale and others 1998; Lovett and others

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

2016), but tends to be more similar to American beech after several years (within standard error range, see Lovett and others (2016). Community of decomposers in EcM forests may be efficient at decomposing recalcitrant organic matter (Fernandez and others 2019). Contrary to the results of Midgley and others (2015) obtained from another study system, we observed HFA in EcM forests but not in AM forests. The efficiency of the microbial decomposers present in EcM soil for decomposing organic matter may be high regardless of litter type and quality. Furthermore, we found no evidence that fragmented (F) and humic (H) horizons in AM stands decomposed faster than the same horizons in EcM stands (but see Jacobs and others 2018). Taken together, these results suggest that the significant impact of initial litter chemistry on decomposition diminishes after the first year of decomposition and that microbial decomposer community may adapt to "home" substrate quality.

Reducing decomposer diversity reduces litter decomposition rate (Handa and others 2014; Li and others 2020), but this effect is context-dependent and the effect of soil fauna is variable across focal species (Makkonen and others 2012). Smaller mesh size are known to reduce the potential diversity of soil fauna that are important for decomposition processes (Hättenschwiler and others 2005). In our study, patches were mainly composed of American beech or sugar maple, and previous studies indicate that maple litter is generally preferred over beech litter by the soil fauna (Hättenschwiler and Bretscher 2001; Jacob and others 2010). However, the difference in decomposition between American beech and sugar maple seems to decrease over time (Lovett and others 2016) and to be dependent on stand type of incubation (Côté and Fyles 1994). Unlike most studies, we used litterbags that were designed to follow decomposition of the upper three organic horizons while avoiding soil trenching and tree girdling which confound the effects of roots and mycorrhizal fungi (Fernandez and Kennedy 2016). Trenching is the historical and most

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

widely used method to test the Gadgil effect, but it is known to directly affect soil drainage, increase soil moisture by impeding root water uptake and strongly disturb the system (Gadgil and Gadgil 1971; Fisher and Gosz 1986; Fernandez and Kennedy 2016). Tree girdling is the most extreme alternative as it kills trees, also preventing further research on the same site. In our experiment, the initial disturbance may have increased labile C but the persistence of this effect after two years was assumed to be rather limited. Furthermore, the observed effects of our exclusion treatment on mass loss increased between the first and second years of incubation suggesting persistent biological effects. Decreasing mesh size might have decreased soil moisture but we observed no impact on litter soluble content losses suggesting a rather low effect caused by mesh size, at least on the most labile fractions of C. It is possible that the use of litterbags with small mesh size limited the exposure to biophysical perturbations, which might hamper mass loss (Prescott 2005; Berg and McClaugherty 2014), but this was common to all treatments. To better predict soil C processes and stocks, more research may be needed to understand how interaction between mycorrhizas, soil fauna, plant inputs and variables such as soil moisture, or bulk density impact decomposition (Lin and others 2017).

Our sampling design allowed us to spatially distinguish decomposition processes in the upper three horizons and assess the fate of young to older organic matter overcoming some limits of short-term experiments. The overall net effect of mycorrhizas on decomposition was positive regardless of mycorrhizal type, but varied throughout the soil profile. Further analyses would allow us to better understand if the greater decomposition could be due to higher microbial biomass inside the 44 µm-pore mesh bags, leading to higher enzymatic activities. As expected from previous studies (e.g., Averill and others 2014; Soudzilovskaia and others 2019), C stocks were greater in EcM stands compared to neighboring AM stands in this northern temperate forest

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

even though decomposition was greater in EcM soils, and positively influenced by the broader fungal network. This indicates the potential importance of others factors such as litter quantity, soil fauna and moisture level in regulating C dynamics. The quality and composition of litter is important for short-term C release, but the microbial community, including root-associated fungi and mycorrhizal-associated organisms (Netherway and others 2020), potentially have a strong impact on a longer-term which is important for C sequestration (Cotrufo and others 2015). Overall, our study shows that forests dominated by different mycorrhizal strategies have distinct impacts in soil organic matter dynamics. The EcM forests store higher soil organic C but support microbial decomposer communities that are more efficient at degrading organic matter than those of adjacent AM forests, rejecting the Gadgil effect as a driver of C accumulation in the northern temperate forests.

Acknowledgements

We would like to thank the staff from the Station de biologie des Laurentides (SBL) of Université de Montréal for facilitating the field work. Funding, including scholarships to AC, was provided by Discovery Grants to EL (RGPIN-2014-06106, RGPIN-2019-04537) by the Natural Sciences and Engineering Research Council of Canada (NSERC) as well as a “Nouveau Chercheur” grant (2016-NC-188823) by the Fonds de recherche du Québec sur la Nature et technologies (FRQNT). AC would like to sincerely thank the following institutions for providing generous scholarships: FRQNT (Dossier 272522), Institut de recherche en biologie végétale, Centre de la science de la biodiversité du Québec, Centre d'étude de la forêt and Université de Montréal through the "Bourse d'excellence Hydro-Québec".

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

Authors' contributions

EL and AC conceived the ideas and designed methodology; AC and FC collected and analyzed the data; AC and EL interpreted the results; AC led the writing of the manuscript. All authors contributed critically to the drafts and gave final approval for publication.

Literature Cited

Allison SD, Lu Y, Weihe C, Goulden ML, Martiny AC, Treseder KK, Martiny JBH. 2013. Microbial abundance and composition influence litter decomposition response to environmental change. Ecology 94:714–25.

Austin AT, Vivanco L, González-Arzac A, Pérez LI. 2014. There's no place like home? An exploration of the mechanisms behind plant litter–decomposer affinity in terrestrial ecosystems. New Phytologist 204:307–14.

Averill C, Turner BL, Finzi AC. 2014. Mycorrhiza-mediated competition between plants and decomposers drives soil carbon storage. Nature 505:543–5.

Bahram M, Peay KG, Tedersoo L. 2015. Local-scale biogeography and spatiotemporal variability in communities of mycorrhizal fungi. New Phytol 205:1454–63.

Baldrian P. 2017. Forest microbiome: diversity, complexity and dynamics. FEMS Microbiol Rev 41:109–30.

Bardgett RD, Wardle DA. 2010. Aboveground-belowground linkages: Biotic interactions, ecosystem processes, and global change. OUP Oxford

Bélanger N, Côté B, Fyles JW, Courchesne F, Hendershot WH. 2004. Forest regrowth as the

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

505 controlling factor of soil nutrient availability 75 years after fire in a deciduous forest of Southern
506 Quebec. Plant and Soil 262:363–272.

507 Berg B, McClaugherty C. 2014. Plant Litter: Decomposition, Humus Formation, Carbon
508 Sequestration. 3rd ed. Berlin Heidelberg: Springer-Verlag.

509 Bödeker ITM, Lindahl BD, Olson Å, Clemmensen KE. 2016. Mycorrhizal and saprotrophic
510 fungal guilds compete for the same organic substrates but affect decomposition differently. Funct
511 Ecol 30:1967–78.

512 Bunn RA, Simpson DT, Bullington LS, Lekberg Y, Janos DP. 2019. Revisiting the 'direct
513 mineral cycling' hypothesis: arbuscular mycorrhizal fungi colonize leaf litter, but why? The
514 ISME Journal 13:1891.

515 Carrillo Y, Dijkstra FA, LeCain D, Pendall E. 2016. Mediation of soil C decomposition by
516 arbuscular mycorrizhal fungi in grass rhizospheres under elevated CO_2. Biogeochemistry
517 127:45–55.

518 Carteron A, Beigas M, Joly S, Turner BL, Laliberté E. 2020. Temperate forests dominated by
519 arbuscular or ectomycorrhizal fungi are characterized by strong shifts from saprotrophic to
520 mycorrhizal fungi with increasing soil depth. Microb Ecol:1–14.

521 Carvalhais N, Forkel M, Khomik M, Bellarby J, Jung M, Migliavacca M, Mu M, Saatchi S,
522 Santoro M, Thurner M, Weber U, Ahrens B, Beer C, Cescatti A, Randerson JT, Reichstein M.
523 2014. Global covariation of carbon turnover times with climate in terrestrial ecosystems. Nature
524 514:213–7.

525 Clemmensen KE, Bahr A, Ovaskainen O, Dahlberg A, Ekblad A, Wallander H, Stenlid J, Finlay

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

526 RD, Wardle DA, Lindahl BD. 2013. Roots and associated fungi drive long-term carbon
527 sequestration in boreal forest. Science 339:1615–8.

528 Clemmensen KE, Finlay RD, Dahlberg A, Stenlid J, Wardle DA, Lindahl BD. 2015. Carbon
529 sequestration is related to mycorrhizal fungal community shifts during long-term succession in
530 boreal forests. New Phytol 205:1525–36.

531 Colin G, Cooney JD, Carlsson DJ, Wiles DM. 1981. Deterioration of plastic films under soil
532 burial conditions. J Appl Polym Sci 26:509–19.

533 Côté B, Fyles JW. 1994. Leaf litter disappearance of hardwood species of southern Québec:
534 Interaction between litter quality and stand type. Écoscience 1:322–8.

535 Cotrufo MF, Soong JL, Horton AJ, Campbell EE, Haddix ML, Wall DH, Parton WJ. 2015.
536 Formation of soil organic matter via biochemical and physical pathways of litter mass loss.
537 Nature Geoscience 8:776–9.

538 Courchesne F, Côté B, Fyles JW, Hendershot WH, Biron PM, Roy AG, Turmel M-C. 2005.
539 Recent changes in soil chemistry in a forested ecosystem of southern Québec, Canada. Soil
540 Science Society of America Journal 69:1298.

541 Craig ME, Turner BL, Liang C, Clay K, Johnson DJ, Phillips RP. 2018. Tree mycorrhizal type
542 predicts within-site variability in the storage and distribution of soil organic matter. Global
543 Change Biology 24:3317–30.

544 Crowther TW, Hoogen J van den, Wan J, Mayes MA, Keiser AD, Mo L, Averill C, Maynard DS.
545 2019. The global soil community and its influence on biogeochemistry. Science 365:eaav0550.

546 Dickie IA, Koele N, Blum JD, Gleason JD, McGlone MS. 2014. Mycorrhizas in changing

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

547 ecosystems. Botany 92:149–60.

548 Dickie IA, Xu B, Koide RT. 2002. Vertical niche differentiation of ectomycorrhizal hyphae in
549 soil as shown by T-RFLP analysis. New Phytologist 156:527–35.

550 Dighton J, Thomas ED, Latter PM. 1987. Interactions between tree roots, mycorrhizas, a
551 saprotrophic fungus and the decomposition of organic substrates in a microcosm. Biol Fert Soils
552 4:145–50.

553 Dixon RK, Solomon AM, Brown S, Houghton RA, Trexier MC, Wisniewski J. 1994. Carbon
554 pools and flux of global forest ecosystems. Science 263:185–90.

555 Fernandez CW, Kennedy PG. 2016. Revisiting the 'Gadgil effect': do interguild fungal
556 interactions control carbon cycling in forest soils? New Phytol 209:1382–94.

557 Fernandez CW, See CR, Kennedy PG. 2019. Decelerated carbon cycling by ectomycorrhizal
558 fungi is controlled by substrate quality and community composition. New Phytologist 226:569–
559 82.

560 Fisher FM, Gosz JR. 1986. Effects of trenching on soil processes and properties in a New Mexico
561 mixed-conifer forest. Biol Fert Soils 2:35–42.

562 Frey SD. 2019. Mycorrhizal fungi as mediators of soil organic matter dynamics. Annu Rev Ecol
563 Evol Syst 50:237–59.

564 Gadgil RL, Gadgil PD. 1971. Mycorrhiza and litter decomposition. Nature 233:133–133.

565 Gholz HL, Wedin DA, Smitherman SM, Harmon ME, Parton WJ. 2000. Long-term dynamics of
566 pine and hardwood litter in contrasting environments: toward a global model of decomposition.

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

567 Global Change Biology 6:751–65.

568 Gui H, Hyde K, Xu J, Mortimer P. 2017. Arbuscular mycorrhiza enhance the rate of litter
569 decomposition while inhibiting soil microbial community development. Scientific Reports
570 7:42184.

571 Handa IT, Aerts R, Berendse F, Berg MP, Bruder A, Butenschoen O, Chauvet E, Gessner MO,
572 Jabiol J, Makkonen M, McKie BG, Malmqvist B, Peeters ETHM, Scheu S, Schmid B, Ruijven J
573 van, Vos VCA, Hättenschwiler S. 2014. Consequences of biodiversity loss for litter
574 decomposition across biomes. Nature 509:218–21.

575 Hättenschwiler S, Bretscher D. 2001. Isopod effects on decomposition of litter produced under
576 elevated CO_2, N deposition and different soil types. Global Change Biology 7:565–79.

577 Hättenschwiler S, Tiunov AV, Scheu S. 2005. Biodiversity and litter decomposition in terrestrial
578 ecosystems. Annual Review of Ecology, Evolution, and Systematics 36:191–218.

579 He X, Critchley C, Ng H, Bledsoe C. 2004. Reciprocal N (15NH4+ or 15NO3−) transfer between
580 nonN2-fixing Eucalyptus maculata and N2-fixing Casuarina cunninghamiana linked by the
581 ectomycorrhizal fungus Pisolithus sp. New Phytologist 163:629–40.

582 Hodge A. 2017. Chapter 8 - Accessibility of inorganic and organic nutrients for mycorrhizas. In:
583 Mycorrhizal Mediation of Soil. Elsevier. pp 129–48.
584 https://www.sciencedirect.com/science/article/pii/B9780128043127000085

585 Hodge A, Campbell CD, Fitter AH. 2001. An arbuscular mycorrhizal fungus accelerates
586 decomposition and acquires nitrogen directly from organic material. Nature 413:297–9.

587 Jacob M, Viedenz K, Polle A, Thomas FM. 2010. Leaf litter decomposition in temperate

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

588 deciduous forest stands with a decreasing fraction of beech (*Fagus sylvatica*). Oecologia
589 164:1083–94.

590 Jacobs LM, Sulman BN, Brzostek ER, Feighery JJ, Phillips RP. 2018. Interactions among
591 decaying leaf litter, root litter and soil organic matter vary with mycorrhizal type. Journal of
592 Ecology 106:502–13.

593 Johnson D, Leake JR, Read DJ. 2001. Novel in-growth core system enables functional studies of
594 grassland mycorrhizal mycelial networks. New Phytologist 152:555–62.

595 Kassambara A. 2018. ggpubr: 'ggplot2' Based Publication Ready Plots. https://CRAN.R-
596 project.org/package=ggpubr https://CRAN.R-project.org/package=ggpubr

597 Keller AB, Phillips RP. 2019. Leaf litter decay rates differ between mycorrhizal groups in
598 temperate, but not tropical, forests. New Phytologist 222:556–64.

599 Koide RT, Wu T. 2003. Ectomycorrhizas and retarded decomposition in a *Pinus resinosa*
600 plantation. New Phytologist 158:401–7.

601 Kubartová A, Ranger J, Berthelin J, Beguiristain T. 2008. Diversity and decomposing ability of
602 saprophytic fungi from temperate forest litter. Microb Ecol 58:98–107.

603 Kuzyakov Y. 2010. Priming effects: Interactions between living and dead organic matter. Soil
604 Biology and Biochemistry 42:1363–71.

605 Kyaschenko J, Clemmensen KE, Karltun E, Lindahl BD. 2017. Below-ground organic matter
606 accumulation along a boreal forest fertility gradient relates to guild interaction within fungal
607 communities. Ecol Lett 20:1546–55.

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

608 Lal R. 2005. Forest soils and carbon sequestration. Forest Ecology and Management 220:242–58.

609 Leifheit EF, Verbruggen E, Rillig MC. 2015. Arbuscular mycorrhizal fungi reduce
610 decomposition of woody plant litter while increasing soil aggregation. Soil Biology and
611 Biochemistry 81:323–8.

612 Lenth R. 2019. emmeans: Estimated Marginal Means, aka Least-Squares Means.
613 https://CRAN.R-project.org/package=emmeans https://CRAN.R-project.org/package=emmeans

614 Li Y, Veen GF (Ciska), Hol WHG, Vandenbrande S, Hannula SE, ten Hooven FC, Li Q, Liang
615 W, Bezemer TM. 2020. 'Home' and 'away' litter decomposition depends on the size fractions of
616 the soil biotic community. Soil Biology and Biochemistry 144:107783.

617 Lin D, Dou P, Yang G, Qian S, Wang H, Zhao L, Yang Y, Mi X, Ma K, Fanin N. 2020. Home-
618 field advantage of litter decomposition differs between leaves and fine roots. New Phytologist
619 227:995–1000.

620 Lin G, McCormack ML, Ma C, Guo D. 2017. Similar below-ground carbon cycling dynamics but
621 contrasting modes of nitrogen cycling between arbuscular mycorrhizal and ectomycorrhizal
622 forests. New Phytologist 213:1440–51.

623 Lindahl BD, de Boer W, Finlay RD. 2010. Disruption of root carbon transport into forest humus
624 stimulates fungal opportunists at the expense of mycorrhizal fungi. The ISME Journal 4:872–81.

625 Lindahl BD, Ihrmark K, Boberg J, Trumbore SE, Högberg P, Stenlid J, Finlay RD. 2007. Spatial
626 separation of litter decomposition and mycorrhizal nitrogen uptake in a boreal forest. New
627 Phytologist 173:611–20.

628 Lindahl BD, Tunlid A. 2015. Ectomycorrhizal fungi – potential organic matter decomposers, yet

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

not saprotrophs. New Phytol 205:1443–7.

Lovett GM, Arthur MA, Crowley KF. 2016. Effects of calcium on the rate and extent of litter decomposition in a northern hardwood forest. Ecosystems 19:87–97.

Makkonen M, Berg MP, Handa IT, Hättenschwiler S, Ruijven J van, Bodegom PM van, Aerts R. 2012. Highly consistent effects of plant litter identity and functional traits on decomposition across a latitudinal gradient. Ecology Letters 15:1033–41.

Malik RJ. 2019. No “Gadgil effect”: Temperate tree roots and soil lithology are effective predictors of wood decomposition. Forest Pathology 49:e12506.

McHale PJ, Mitchell MJ, Bowles FP. 1998. Soil warming in a northern hardwood forest: trace gas fluxes and leaf litter decomposition. Can J For Res 28:1365–72.

Midgley MG, Brzostek E, Phillips RP. 2015. Decay rates of leaf litters from arbuscular mycorrhizal trees are more sensitive to soil effects than litters from ectomycorrhizal trees. Journal of Ecology 103:1454–63.

Moore TR, Trofymow JA, Taylor B, Prescott C, Camiré C, Duschene L, Fyles J, Kozak L, Kranabetter M, Morrison I, Siltanen M, Smith S, Titus B, Visser S, Wein R, Zoltai S. 1999. Litter decomposition rates in Canadian forests. Global Change Biology 5:75–82.

Mujic AB, Durall DM, Spatafora JW, Kennedy PG. 2016. Competitive avoidance not edaphic specialization drives vertical niche partitioning among sister species of ectomycorrhizal fungi. New Phytologist 209:1174–83.

Netherway T, Bengtsson J, Krab EJ, Bahram M. 2020. Biotic interactions with mycorrhizal systems as extended nutrient acquisition strategies shaping forest soil communities and functions.

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

650 Basic and Applied Ecology:10.1016/j.baae.2020.10.002.

651 Orwin KH, Kirschbaum MUF, St John MG, Dickie IA. 2011. Organic nutrient uptake by
652 mycorrhizal fungi enhances ecosystem carbon storage: a model-based assessment. Ecology
653 Letters 14:493–502.

654 Phillips RP, Brzostek E, Midgley MG. 2013. The mycorrhizal-associated nutrient economy: a
655 new framework for predicting carbon–nutrient couplings in temperate forests. New Phytol
656 199:41–51.

657 Pinheiro J, Bates D, DebRoy S, Sarkar D, Team RC. 2012. nlme: Linear and nonlinear mixed
658 effects models. R package version 3.

659 Prescott CE. 2005. Do rates of litter decomposition tell us anything we really need to know?
660 Forest Ecology and Management 220:66–74.

661 R Core Team. 2018. R: A Language and Environment for Statistical Computing. Vienna, Austria:
662 R Foundation for Statistical Computing https://www.R-project.org/

663 Read DJ, Leake JR, Perez-Moreno J. 2004. Mycorrhizal fungi as drivers of ecosystem processes
664 in heathland and boreal forest biomes. Can J Bot 82:1243–63.

665 Rosling A, Landeweert R, Lindahl BD, Larsson K-H, Kuyper TW, Taylor AFS, Finlay RD. 2003.
666 Vertical distribution of ectomycorrhizal fungal taxa in a podzol soil profile. New Phytologist
667 159:775–83.

668 Schimel JP, Schaeffer SM. 2012. Microbial control over carbon cycling in soil. Front Microbiol
669 3:348.

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

Sietiö O-M, Santalahti M, Putkinen A, Adamczyk S, Sun H, Heinonsalo J. 2019. Restriction of plant roots in boreal forest organic soils affects the microbial community but does not change the dominance from ectomycorrhizal to saprotrophic fungi. FEMS Microbiol Ecol 95. https://academic.oup.com/femsec/article/95/9/fiz133/5554003. Last accessed 21/10/2019

Smith GR, Wan J. 2019. Resource-ratio theory predicts mycorrhizal control of litter decomposition. New Phytologist 223:1595–606.

Smith SE, Read DJ. 2008. Mycorrhizal Symbiosis. Academic Press

Soudzilovskaia NA, Bodegom PM van, Terrer C, Zelfde M van't, McCallum I, McCormack ML, Fisher JB, Brundrett MC, Sá NC de, Tedersoo L. 2019. Global mycorrhizal plant distribution linked to terrestrial carbon stocks. Nat Commun 10:1–10.

Soudzilovskaia NA, van der Heijden MGA, Cornelissen JHC, Makarov MI, Onipchenko VG, Maslov MN, Akhmetzhanova AA, van Bodegom PM. 2015. Quantitative assessment of the differential impacts of arbuscular and ectomycorrhiza on soil carbon cycling. New Phytol 208:280–93.

Steidinger BS, Crowther TW, Liang J, Nuland MEV, Werner GDA, Reich PB, Nabuurs G, de-Miguel S, Zhou M, Picard N, Herault B, Zhao X, Zhang C, Routh D, Peay KG. 2019. Climatic controls of decomposition drive the global biogeography of forest-tree symbioses. Nature 569:404.

Sterkenburg E, Clemmensen KE, Ekblad A, Finlay RD, Lindahl BD. 2018. Contrasting effects of ectomycorrhizal fungi on early and late stage decomposition in a boreal forest. The ISME Journal 12:2187–97.

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

691 Subke J-A, Voke NR, Leronni V, Garnett MH, Ineson P. 2011. Dynamics and pathways of
692 autotrophic and heterotrophic soil CO_2 efflux revealed by forest girdling. Journal of Ecology
693 99:186–93.

694 Taylor MK, Lankau RA, Wurzburger N. 2016. Mycorrhizal associations of trees have different
695 indirect effects on organic matter decomposition. J Ecol 104:1576–84.

696 Tedersoo L, Bahram M. 2019. Mycorrhizal types differ in ecophysiology and alter plant nutrition
697 and soil processes. Biological Reviews 94:1857–80.

698 Teste FP. 2008. Role of mycorrhizal networks in dry Douglas-fir forests. University of British
699 Columbia https://open.library.ubc.ca/cIRcle/collections/ubctheses/24/items/1.0066342. Last
700 accessed 21/10/2020

701 Teste FP, Karst J, Jones MD, Simard SW, Durall DM. 2006. Methods to control ectomycorrhizal
702 colonization: effectiveness of chemical and physical barriers. Mycorrhiza 17:51–65.

703 Teste FP, Simard SW, Durall DM, Guy RD, Jones MD, Schoonmaker AL. 2009. Access to
704 mycorrhizal networks and roots of trees: importance for seedling survival and resource transfer.
705 Ecology 90:2808–22.

706 Tisserant E, Malbreil M, Kuo A, Kohler A, Symeonidi A, Balestrini R, Charron P, Duensing N,
707 Frey NF dit, Gianinazzi-Pearson V, Gilbert LB, Handa Y, Herr JR, Hijri M, Koul R, Kawaguchi
708 M, Krajinski F, Lammers PJ, Masclaux FG, Murat C, Morin E, Ndikumana S, Pagni M,
709 Petitpierre D, Requena N, Rosikiewicz P, Riley R, Saito K, Clemente HS, Shapiro H, Tuinen D
710 van, Bécard G, Bonfante P, Paszkowski U, Shachar-Hill YY, Tuskan GA, Young JPW, Sanders
711 IR, Henrissat B, Rensing SA, Grigoriev IV, Corradi N, Roux C, Martin F. 2013. Genome of an

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

712 arbuscular mycorrhizal fungus provides insight into the oldest plant symbiosis. PNAS
713 110:20117–22.

714 Veen GF (Ciska), Freschet GT, Ordonez A, Wardle DA. 2015. Litter quality and environmental
715 controls of home-field advantage effects on litter decomposition. Oikos 124:187–95.

716 Verbruggen E, Pena R, Fernandez CW, Soong JL. 2017. Chapter 24 - Mycorrhizal interactions
717 with saprotrophs and impact on soil carbon storage. In: Mycorrhizal Mediation of Soil. Elsevier.
718 pp 441–60. https://www.sciencedirect.com/science/article/pii/B9780128043127000243

719 van der Wal A, Geydan TD, Kuyper TW, de Boer W. 2013. A thready affair: linking fungal
720 diversity and community dynamics to terrestrial decomposition processes. FEMS Microbiol Rev
721 37:477–94.

722 Wang Y, Li FY, Song X, Wang X, Suri G, Baoyin T. 2020. Changes in litter decomposition rate
723 of dominant plants in a semi-arid steppe across different land-use types: Soil moisture, not home-
724 field advantage, plays a dominant role. Agriculture, Ecosystems & Environment 303:107119.

725 Wickham H. 2016. ggplot2: Elegant graphics for data analysis. Springer-Verlag New York
726 https://CRAN.R-project.org/package=ggplot2

727 Wickham H, Francois R, Henry L, Müller K. 2017. dplyr: A grammar of data manipulation.
728 https://CRAN.R-project.org/package=dplyr https://CRAN.R-project.org/package=dplyr

729 Wiesmeier M, Urbanski L, Hobley E, Lang B, von Lützow M, Marin-Spiotta E, van Wesemael
730 B, Rabot E, Ließ M, Garcia-Franco N, Wollschläger U, Vogel H-J, Kögel-Knabner I. 2019. Soil
731 organic carbon storage as a key function of soils - A review of drivers and indicators at various
732 scales. Geoderma 333:149–62.

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

733 Wurzburger N, Brookshire ENJ. 2017. Experimental evidence that mycorrhizal nitrogen
734 strategies affect soil carbon. Ecology 98:1491–7.

735 Xu J, Liu S, Song S, Guo H, Tang J, Yong JWH, Ma Y, Chen X. 2018. Arbuscular mycorrhizal
736 fungi influence decomposition and the associated soil microbial community under different soil
737 phosphorus availability. Soil Biology and Biochemistry 120:181–90.

738 Zak DR, Pellitier PT, Argiroff W, Castillo B, James TY, Nave LE, Averill C, Beidler KV,
739 Bhatnagar J, Blesh J, Classen AT, Craig M, Fernandez CW, Gundersen P, Johansen R, Koide
740 RT, Lilleskov EA, Lindahl BD, Nadelhoffer KJ, Phillips RP, Tunlid A. 2019. Exploring the role
741 of ectomycorrhizal fungi in soil carbon dynamics. New Phytologist 223:33–9.

742 Zhu W, Ehrenfeld JG. 1996. The effects of mycorrhizal roots on litter decomposition, soil biota,
743 and nutrients in a spodosolic soil. Plant Soil 179:109–18.

744

745 **Conflict of Interest**

746 The authors declare that they have no conflict of interest.

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

Table legends

Table 1. Initial soil chemistry characteristics: C:N ratio, lignin:N ratio, $\delta^{15}N$, thickness and carbon stocks of the upper three horizons (litter - L, fragmented - F, humic - H) of arbuscular mycorrhizal (AM) and ectomycorrhizal (EcM) forest. Means and standard deviation are shown (*n* = 5).

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

	Horizon	AM-dominated forest	Standard deviation	EcM-dominated forest	Standard deviation
C:N ratio	L	22.71	2.53	27.36	1.81
	F	20.50	1.01	22.32	1.36
	H	18.41	0.85	20.09	1.48
lignin:N ratio	L	9.68	1.40	13.05	1.74
	F	10.70	1.69	12.57	0.99
	H	8.62	1.30	10.50	1.50
$\delta^{15}N$ (‰)	L	-3.26	0.36	-3.16	0.63
	F	-2.48	0.40	-1.96	0.48
	H	-0.72	0.59	-0.74	0.36
Thickness (cm)	L	0.71	0.22	0.81	0.30
	F	2.36	0.68	3.44	0.65
	H	4.21	0.95	7.19	2.73
Carbon stock ($kg.m^2$)	L	0.89	0.28	1.05	0.43
	F	2.68	0.76	4.24	0.86
	H	2.97	0.67	6.99	4.00

753

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

754 **Figure legends**

755

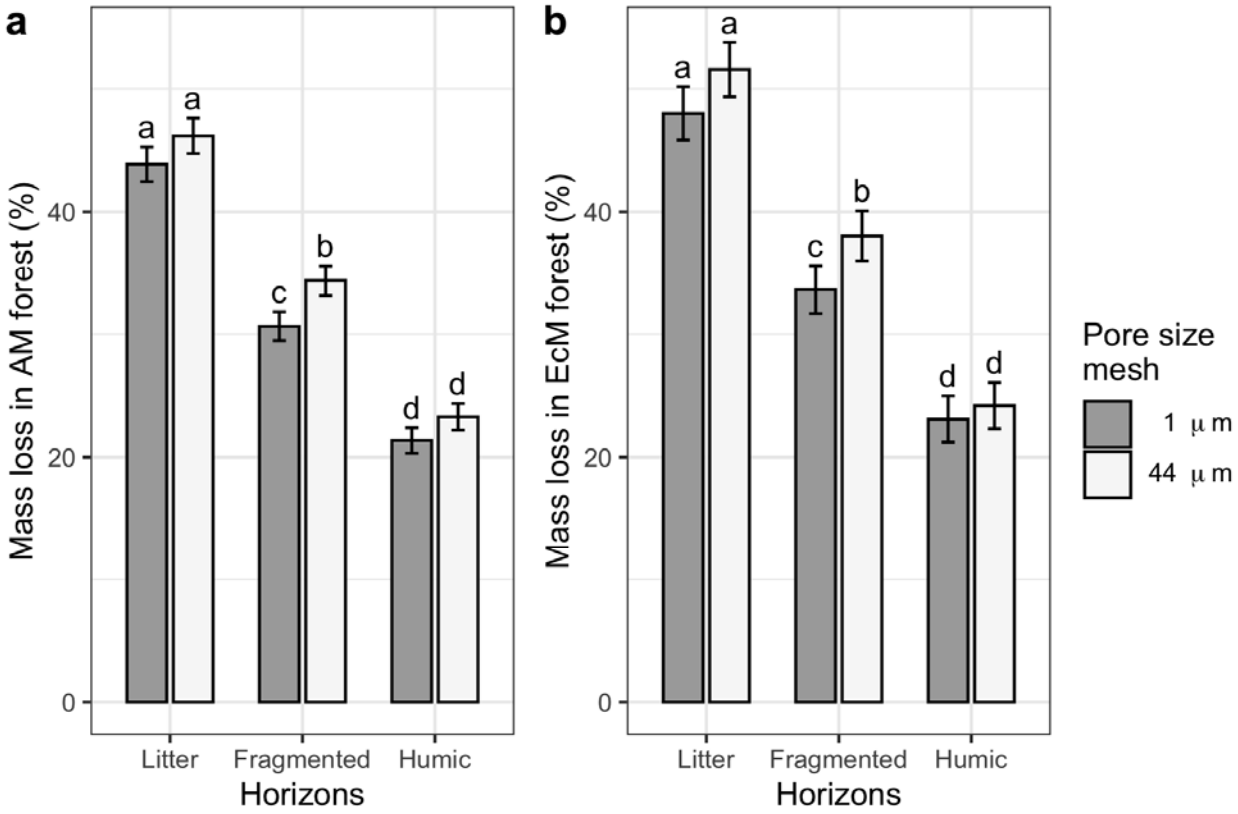
756 **Figure 1.** Percentage of mass loss of the three upper horizons incubated for two years in forests
757 dominated by (a) arbuscular mycorrhiza (AM) or (b) ectomycorrhiza (EcM) in litterbags with
758 pore mesh size of 1 μm (grey bars) and 44 μm (white bars). Means ± 1 SE are shown (n = 20).
759 Multiple comparison using Tukey's honestly significant difference post-hoc test, different letters
760 within each panel indicates significant differences (P-value < 0.05).

761

762 **Figure 2.** Percentage of mass loss after one and two years of incubation in forests dominated by
763 arbuscular mycorrhiza (AM) or ectomycorrhiza (EcM) in litterbags with pore size mesh of 1 μm
764 (top panels) and 44 μm (bottom panels) and organic matter provenance from AM and EcM.
765 Means ± 1 SE are shown (n = 30). Multiple comparison using Tukey's honestly significant
766 difference post-hoc test, different letters indicates significant differences (P-value < 0.05).

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

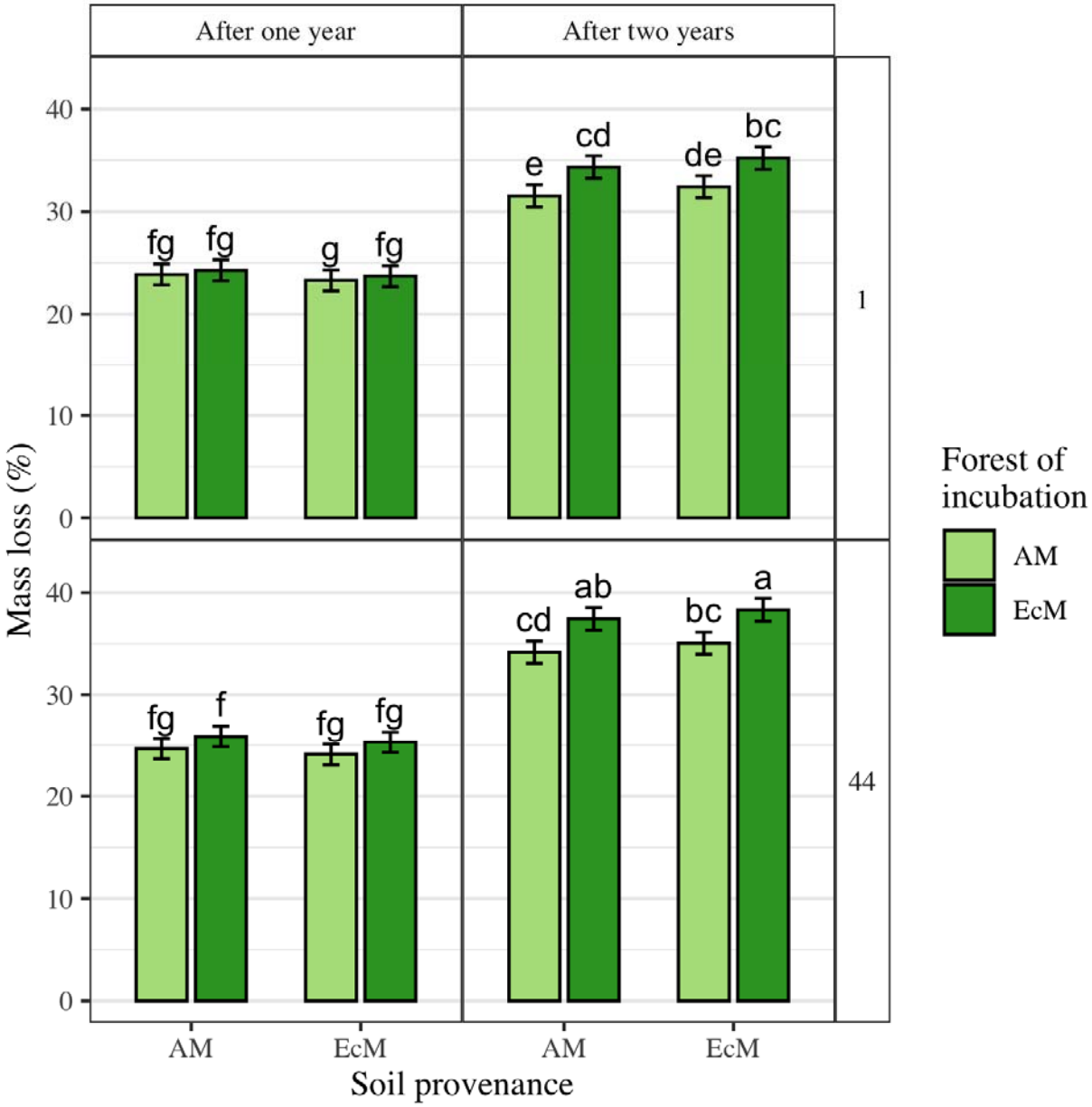
767 **Figure 1**



768

bioRxiv preprint doi: https://doi.org/10.1101/2021.02.09.430490; this version posted February 10, 2021. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY 4.0 International license.

769 **Figure 2**



770