# Occurrence of Arbuscular Mycorrhizal Herbs Decreases Selectively in Communities Dominated by Invasive Tree *Acer negundo*

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**Abstract**—We tested whether one of the consequences predicted for alien plant invasion by the mutualism disruption hypothesis was true in the case of the ash-leaved maple *Acer negundo* L. The study aimed to determine whether the occurrences of mycorrhizal and nonmycorrhizal herbs varied similarly or differently in communities with varying degrees of *A. negundo* dominance. The analysis included the results of 78 vegetation descriptions carried out in Belarusian Polesia, the Middle Volga region, and the Middle Urals. Communities with or without *A. negundo* dominance were described in each region. The mycorrhizal status of plant species was determined using the FungalRoot Database. Species that are more likely to form arbuscular mycorrhiza were found to occur less frequently in *A. negundo* thickets. On the contrary, a higher probability of the nonmycorrhizal status was associated with a lower frequency of detection in *A. negundo* thickets. Therefore, the occurrence of arbuscular mycorrhizal herbs was found to selectively decrease in communities dominated by *A. negundo*.

## INTRODUCTION

Hypotheses about the transformation of soil conditions by alien plants or the specificity of plant-soil feedbacks are some of the frequently discussed to explain the success of alien plants. In essence, the hypotheses suppose that alien plants change the chemical or biological properties of soils in the invaded communities, forming selectively favorable conditions for their own development and unfavorable conditions for native plants. A degraded mutualism (or mutualism disruption) hypothesis is of particular interest [1−3]. According to the hypothesis, when invasive plants depend to a lesser extent on mycorrhizal fungi as compared with native plants, local fungal communities change to facilitate invasions of nonlocal nonmycorrhizal species and to prevent recovery of native mycorrhizal species. A meta-analysis confirmed the hypothesis [3]. Degradation or disruption of mycorrhizal connections possibly triggers a positive feedback system, hence increasing the invasion processes. Invasion of European *Alliaria petiolata* in North America provided an illustrative example of how inhibition of local mycorrhizal fungi ensures successful invasion [4, 5].

We have studied various components of plant−soil feedbacks in the case of invasive ash-leaved maple *Acer negundo*. In particular, our aim was to understand whether invasion of local communities by *A. negundo* changes the efficiency of mycorrhiza formation in native plants. The aim of this work was to determine whether the occurrences of mycorrhizal and nonmycorrhizal herbs varied similarly or differently in communities with different degrees of *A. negundo* dominance. If *A. negundo* affects the fungal communities of arbuscular mycorrhizas or the mycorrhiza formation in plants growing in its thickets, the proportion of herb species may be shifted consequently towards a higher abundance of nonmycorrhizal species.

## MATERIALS AND METHODS

**Regions.** Field material was collected in three geographic regions: Belarusian Polesia, the Middle Volga region, and the Middle Urals. The study was carried out in the broadleaved forest zone of Belarusian Polesia (Gomel and its surroundings) in 2021, the forest-

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steppe zone of the Middle Volga region (Ulyanovsk and its surroundings) in 2023, and the southern taiga subzone of the boreal forest zone (Yekaterinburg and its surroundings) in 2018 and 2019. Moderate continental climate is characteristic of all regions. According to a classification [6], all investigated regions have cold climate with a warm summer season, which is short in the Middle Urals. The regions are similar in the prevalent types and intensity of the anthropogenic impact on ecosystems.

**General observation design and community relevés.** In all regions, paired plant communities dominated by *A. negundo* in one of them (designated "An+") were compared to others without its dominance ("An−"). We selected the paired communities in similar conditions within the same landscape elements; no more than 600 m apart; and, wherever possible, in similar habitat type, the extent of disturbance, moisture conditions, and canopy cover. Therefore, conditions of the An− sample plots were considered as an analogue of An+ sample plots conditions if there was no *A. negundo* invasion. The final dataset included 78 relevés (39 relevés of An+ communities and 39 relevés of An− communities): 24 relevés from Belarusian Polesia (12 relevés for each of the An+ and An− treatments), 30 relevés from the Middle Volga region (15 relevés for each of the An+ and An− treatments), and 24 relevés from the Middle Urals (12 relevés for each of the An+ and An− treatments).

Dominant tree species of An− communities were *Acer platanoides*, *Alnus glutinosa*, *Fraxinus excelsior*, *Pinus sylvestris*, *Populus tremula*, *Quercus robur*, and *Robinia pseudoacacia* in Belarusian Polesia; *Acer platanoides*, *Betula pendula*, *Fraxinus excelsior*, *Populus alba*, *Populus balsamifera*, *Quercus robur*, and *Salix euxina* in the Middle Volga region; and *Prunus padus*, *Pinus sylvestris*, *Quercus robur*, *Sorbus aucuparia*, *Tilia cordata*, *Malus baccata*, *Salix alba*, *Salix fragilis*, and *Ulmus laevis* in the Middle Urals.

To describe a community, we recorded all plant species growing within 400 m$^2$ and forming the herb−shrub layer. The total dataset included records of 338 plant species of the herb−shrub layer.

**Selectivity of species occurrence in the An+ and An− community.** Out of the 338 species, we selected the species that appeared in at least 10% of the 78 relevés (i.e., in eight or more relevés). A total of 68 species were selected. For each of the species, the selectivity $S$ of occurrence in communities with *A. negundo* dominance was determined as a proportion of species records from communities with *A. negundo* dominance. Values $S = 0$ indicated the species was never found in *A. negundo* thickets; values $S = 1$ indicated the species was totally confined to communities dominated by *A. negundo*.

**Mycorrhizal status of plant species** was determined using the FungalRoot Database [7]. The database accumulates the results of tests of mycorrhiza presence for 14.5 thousand plant taxa with the number of tests from 1 to 129 for each taxon. FungalRoot data were used in studies of various designs, including allelopathy studies of alien plants [8]. The mycorrhizal status of a plant species (M) was defined as the proportion of tests that detected mycorrhiza formation in the species in the total number of tests for mycorrhiza formation of the species. Within this approach, the $M$ parameter had a probabilistic interpretation: $M = 0$ indicates that mycorrhiza was never found for the species, $M = 0.33$—mycorrhiza was found in a third of tests, $M = 1.00$—mycorrhiza was always found.

**Data analyses.** Pearson's ($r$) and Spearman's ($r_S$) correlation coefficients were calculated.

## RESULTS AND DISCUSSION

Out of the 68 plant species selected, four (*Alliaria petiolata*, *Moehringia trinervia*, *Catolobus pendulus*, and *Silene latifolia* subsp. *alba*) were never found to form a mycorrhiza ($M = 0$). The other 64 species were more or less likely to form arbuscular mycorrhiza. Species with other mycorrhiza types were absent. The $M$ value was equal to 1 in 19 species; i.e. a mycorrhiza was always detected. In 45 species, the probability of forming a mycorrhiza varied ($M = 0.20−0.97$).

Out of the 68 species, 35 were found in each of the three regions: Belarusian Polesia, the Middle Volga region, and the Middle Urals. Other 26 species were found in two regions; 7 species were detected in only one region. Hereby, species with broad geographical distribution prevailed in the dataset. Each of the 68 species was at least once observed both in a community dominated by *A. negundo* and in a community without *A. negundo*. Therefore, the dataset did not include any species only associated with communities dominated by *A. negundo* or absolutely avoided such communities.

Compared with the control communities without *A. negundo*, communities dominated by *A. negundo* were found a lower frequency of species always or often forming a mycorrhiza. The first group ($M = 1.00$) included *Agrostis gigantea*, *Carum carvi*, *Sonchus oleraceus*, *Trifolium medium*, *Trifolium repens*, and *Vicia cracca*. The other group ($M = 0.85−0.96$) included *Achillea millefolium*, *Bromus inermis*, *Calamagrostis epigejos*, *Erigeron canadensis*, *Deschampsia cespitosa*, *Lolium pratense*, *Lysimachia vulgaris*, *Poa compressa*, and *Tussilago farfara*. *Dryopteris carthusiana* and *Sisymbrium officinale* also tended to occur more often in communities dominated by *A. negundo*; their probabilities to have a mycorrhiza were low ($M = 0.33−0.50$).

Species with high $S$ values (i.e., species associated mostly with communities dominated by *A. negundo*)
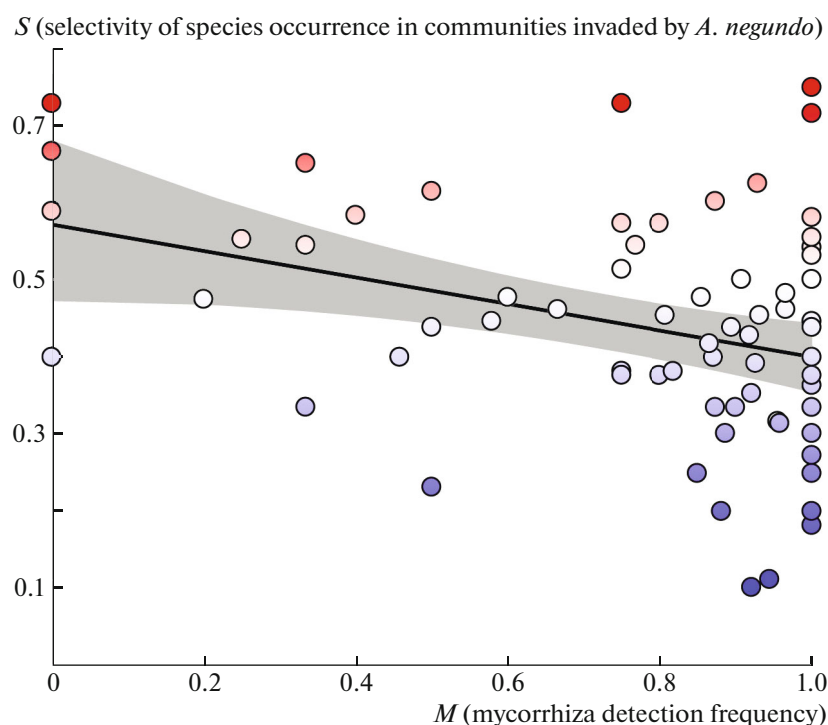
**Fig. 1.** Relationship between mycorrhiza detection frequency of species according to the FungalRoot Database ($M$) and selectivity of species occurrence in communities invaded by *A. negundo* ($S$). The 95% confidence interval is filled gray. A red component of markers' filling indicates the increased species occurrence in communities invaded by *A. negundo;* a blue component of markers' filling indicates the preferential avoidance of communities dominated by *A. negundo*.

varied in soil nutrition mechanisms and included nonmycorrhizal plants ($M = 0$: *Catolobus pendulus* and *Silene latifolia* subsp. *alba*), plants with intermediate probability of mycorrhiza formation ($M = 0.33-0.75$: *Chelidonium majus* and *Leonurus quinquelobatus*), and obligate mycorrhizal plants ($M = 1.00$: *Erigeron annuus* and *Pastinaca sativa*).

A correlation between the probability to find a mycorrhiza ($M$) and the selective occurrence of a species in communities dominated by *A. negundo* ($S$) was negative and statistically significant ($r = -0.34$, $P = 0.0047$, $n = 68$; Fig. 1). Because the assumptions to use parametric methods were not met, the association between $M$ and $S$ was additionally tested using Spearman's correlation coefficient: $r_S = -0.29$, $P = 0.0164$. Therefore, a higher probability for a species to form an arbuscular mycorrhiza was associated with a lower probability for the species to be detected in *A. negundo* thickets. Oppositely, a higher probability of the nonmycorrhizal status was associated with a higher probability for the species to be detected in *A. negundo* thickets. The findings indicated that the occurrence of arbuscular mycorrhizal herbs decreased selectively in communities dominated by *A. negundo*.

Therefore, the occurrence of arbuscular mycorrhizal herbs was mostly decreased in communities dominated by *A. negundo*. Nonmycorrhizal herbs did not demonstrate negative trends, or those herbs were more frequent in *A. negundo* thickets than in other communities. This pattern corresponds to the idea of nonmycorrhizal plants increasing their occurrence in communities invaded by alien plants, as predicted by the degraded mutualism (mutualism disruption) hypothesis [1−3]. This pattern is also consistent with the observed phenomenon of decreased mycorrhiza formation in plants grown in greenhouse experiments in soil from communities invaded by *A. negundo* [9].

The claims of the selective decrease in the arbuscular mycorrhizal herbs' occurrence in invaded communities could only be stated hypothetically. It can be supposed that the mycorrhizal fungi abundance in soil decreases under dense *A. negundo* canopy. It should be taken into account, although *A. negundo* can form arbuscular mycorrhiza both in native [10] and secondary [11−13] ranges. A certain background amount of mycorrhizal fungi can therefore be preserved in *A. negundo* thickets, and soil "sterilization" probably does not occur, in contrast to what has been observed in case of *Alliaria petiolata* invasion [4, 5]. The composition, rather than abundance, of fungi may be altered in *A. negundo* thickets, as it has been demonstrated for other invasions [14, 15].

The growth and metabolism of mycorrhizal fungi strongly depend on the amount of photosynthetically

fixed carbon (carbohydrates) supplied by plants. A decrease in carbon supply can reduce the viability of mycorrhizal fungi. A greater shading can be formed by *A. negundo* compared with native trees [16], and this has been discussed as a mechanism of the *A. negundo* effect [17]. Shading reduces the symbiosis efficiency for plants [18]. Nonmycorrhizal plants, independent of fungi may therefore get selective advantage over mycorrhizal plants in conditions with low light availability.

A direct relationship does not necessarily occur between transformation of communities and habitats by *A. negundo* invasion and a selective decrease in occurrence of arbuscular mycorrhizal herbs. Nonmycorrhizal and mycorrhizal types of soil nutrition are components of wide adaptive syndromes. The syndromes are described as ecological strategy that affect the ecological and physiological features and life forms of species, reproduction features etc. The nonmycorrhizal status is often associated with a short lifespan and a ruderal ecological strategy. Therefore, it is possible, the decrease in the occurrence of arbuscular mycorrhizal herbs may be the result of the elimination of species under *A. negundo* canopy based on some other features.

## CONCLUSIONS

Mycorrhiza formation was analyzed as a functional trait in a way similar to one used in analyses of functional diversity. The probabilistic approach to identifying the mycorrhizal status on the basis of a large set of published data made it possible to obtain the methodically homogeneous quantitative characteristics that are suitable for a formal statistical analysis. In addition, the approach allowed us to avoid coarsening of data, which is often used in similar studies due to the isolation of discrete species groups (nonmycorrhizal, low mycorrhizal, obligate mycorrhizal, etc.). Using the approach in the case of the invasive tree *A. negundo*, we obtained the results that support the degraded mutualism (mutualism disruption) hypothesis. An increase in the occurrence of nonmycorrhizal herbs in communities invaded by *A. negundo* is possible to interpret as a simplification of the plant community structure.

## FUNDING

## ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This work does not contain any studies involving human and animal subjects.

## CONFLICT OF INTEREST

The authors of this work declare that they have no conflicts of interest.

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