Acta Oecologica 77 (2016) 128–135

Contents lists available at ScienceDirect

Acta Oecologica

journal homepage: www.elsevier.com/locate/actoec

Original article

Dispersal of arbuscular mycorrhizal fungi and plants during succession

David García de León*, Mari Moora, Maarja Öpik, Teele Jairus, Lena Neuenkamp, Martti Vasar, C. Guillermo Bueno, Maret Gerz, John Davison, Martin Zobel

Institute of Ecology and Earth Sciences, Department of Botany, University of Tartu, Estonia

ARTICLE INFO

Article history:
Received 26 May 2016
Received in revised form
6 October 2016
Accepted 10 October 2016

Keywords:
Arbuscular mycorrhiza
Cultured fungi
Dispersal ability
Functional traits
Plant-fungi interaction
Phylogenetic distance

ABSTRACT

Arbuscular mycorrhizal (AM) fungi are important root symbionts that enhance plant nutrient uptake and tolerance to pathogens and drought. While the role of plant dispersal in shaping successional vegetation is well studied, there is very little information about the dispersal abilities of AM fungi. We conducted a trap-box experiment in a recently abandoned quarry at 10 different distances from the quarry edge (i.e. the potential propagule source) over eleven months to assess the short term, within-year, arrival of plant and AM fungal assemblages and hence their dispersal abilities. Using DNA based techniques we identified AM fungal taxa and analyzed their phylogenetic diversity. Plant diversity was determined by transporting trap soil to a greenhouse and identifying emerging seedlings. We recorded 30 AM fungal taxa. These contained a high proportion of ruderal AM fungi (30% of taxa, 79% of sequences) but the richness and abundance of AM fungi were not related to the distance from the presumed propagule source. The number of sequences of AM fungi decreased over time. Twenty seven plant species (30% of them ruderal) were recorded from the soil seed traps. Plant diversity decreased with distance from the propagule source and increased over time. Our data show that AM fungi with ruderal traits can be fast colonizers of early successional habitats.

1. Introduction

The diversity and composition of plant communities is to a significant degree shaped by dispersal limitation (Cadotte, 2006; Myers and Harms, 2009; Zobel, 2016). Dispersal limitation also influences the process of plant community succession. The species composition of early successional plant communities with sparsely distributed plant individuals has been shown to reflect seed rain composition (Fraaije et al., 2015), while sampling of seed rain indicates that the quantity and diversity of arriving seeds decrease with distance from seed sources (Diacon-Bolli et al., 2013). Consequently, the composition of successional vegetation depends on the proximity of potential seed sources (Tischew et al., 2014; Torrez et al., 2016).

In recent years there has been increasing interest in the role of microbial organisms in shaping the structure and dynamics of plant communities (Bardgett and van der Putten, 2014). Arbuscular mycorrhizal (AM) fungi (Phylum Glomeromycota) are an ancient group of root symbionts that associate with more than 80% of plants in terrestrial ecosystems (Smith and Read, 2008). AM fungi enhance plant nutrient uptake and tolerance to abiotic and biotic stresses such as drought and pathogens. These fungi may have multiple impacts on plant community composition and dynamics. For instance, microcosm (Vogelsang et al., 2006) and field (Yang et al., 2014) experiments show that manipulation of AM fungi results in changes to plant community composition and diversity, suggesting that particular fungal taxa may play important roles for plant communities. While there is considerable information about plant dispersal and its role in driving plant community succession, much less is known about the dispersal of AM fungi.

AM fungi were traditionally viewed as poor dispersers due to their relatively large spores (0.01–1 mm). However, AM fungi have three types of propagule-soil-borne spores, mycelial fragments and colonised root pieces- and multiple potential dispersal vectors, including wind, water and animals (Smith and Read, 2008). There is empirical evidence of dispersal of AM fungal propagules by wind (Egan et al., 2014; Warner et al., 1987), invertebrates (Gange, 1993; Harinikumar and Bagyaraj, 1994; Warner et al., 1987), rodents (Janos et al., 1995; Mangan and Adler, 2002; Vernes and Dunn, 2009), large mammals (Lekberg et al., 2011) and birds (Nielsen et al., 2016). Many AM fungal taxa have recently been found to exhibit cosmopolitan distributions (Davison et al., 2015),

* Corresponding author. Lai 40, Tartu, 51005, Estonia.
E-mail address: david.garciadeleon@ut.ee (D. García de León).

http://dx.doi.org/10.1016/j.actao.2016.10.006
1146-609X/

suggesting that they may be relatively efficient dispersers at large spatial scales, at least over long time periods. Evidence showing the presence of AM fungi in recently exposed substrate also indicates an ability to arrive from neighbouring habitats relatively quickly (in less than a year; Dodd et al., 2002). However, there is little quantitative information about which AM fungal taxa colonize early successional sites and how the speed of AM fungal colonization compares to that of plants.

It is well known that different plant functional types replace each other during succession. For instance, early successional stages are characterized by a prevalence of ruderal species with good dispersal ability (e.g. Caccianiga et al., 2006; Lavorel et al., 1999). In principle, it is possible to consider different types of strategies among AM fungi. AM fungal isolates, species and families exhibit functional differences, e.g. in their ability to produce spores that facilitate long distance dispersal (Chagnon et al., 2013; Van Der Heijden and Scheublin, 2007). Only AM fungal taxa that produce spores in the so-called trap cultures can be multiplied with the current methods used to bring AM fungi to culture; this characteristic (cultured vs uncultured) has been used as a proxy for certain life history traits related to ease of sporulation. Cultured AM fungal taxa are expected to be better colonizers than uncultured taxa, and to represent ruderal strategists (Chagnon et al., 2013; Ohsowski et al., 2014; van der Heijden et al., 2008). Because ruderal AM fungi more often occur in human impacted habitats (Ohsowski et al., 2014), a predominance of such taxa in early successional habitats could be expected. Indeed, Nielsen et al. (2016) showed that AM fungal lineages with a predominantly ruderal strategy rapidly colonized a barren substrate (artificial island). However, there are no direct measurements documenting the arrival of particular AM fungal taxa on formerly un-colonized substrate.

We used traps filled with natural sterilized substrate and measured dispersal of AM fungi and plants in a limestone quarry within a matrix of calcareous grassland. We recorded propagules arriving at different distances from the quarry edge (the potential propagule source). We asked whether the arrival of AM fungal and plant propagules depends on distance from the propagule source - surrounding calcareous grasslands, within-year arrival helps to understand AM fungal diversity changes, and whether the abundance and richness of colonizing plant species and AM fungal taxa was similar. We expected distance and time to affect the diversity of arriving propagules, and ruderal taxonomic groups to be dominant at the early successional sites.

2. Materials and methods

2.1. Study site

The study was conducted in a limestone quarry at Koguva (58°36′- 58°37′ N, 23°05′ - 23°06′ E) on the Estonian island of Muhu in the eastern Baltic Sea. The climate of the study area is mild-maritime, with 500–700 mm annual precipitation and 17 °C and −5 °C mean temperatures in July and January, respectively (Jaagus, 1999). The quarry is surrounded by a matrix of alvar grasslands - floristically diverse calcareous grasslands on thin soil (Pärtel et al., 1999). The AM fungal community in the surrounding grasslands is representative of the global and European diversity of AM fungi, according to Maarj*AM* database (García de León et al. 2016; Öpik et al., 2010). The quarry was actively used until the beginning of 2014, at which point the substrate of the area largely consisted of gravel and bare rock, and there were no established plants besides single specimens of *Taraxacum* spp. (unknown mycorrhizal status), and *Juncus* spp. (nonmycorrhizal). As abandonment of the area occurred shortly before the start of this study, it is unlikely that the quarry area represented a net source of mycorrhizal inoculum. The size of the gravel pit was approximately 1100 × 400 m. There was no visible soil arrival (e.g. due to water erosion) from the neighborhood; and soil formation appeared to be a local process driven by weathering of the parent material. Because only a relatively thin layer of gravel was removed from the pit, the topography was only moderately changed (i.e. depressed).

2.2. Sampling and sample processing

Soil from mature alvar grasslands was collected, pooled, mixed and sterilized by gamma-irradiation (20 h at 1 kGy). One hundred trap-boxes (190 × 165 × 60 mm) filled with sterilized soil were then embedded in the quarry substrate during the second week of June 2014. Trap-boxes included drainage to reduce the impact of precipitation on collected AM fungi and seeds. Pairs of adjacent trap-boxes, 30 cm apart, were established along five transects (each 36 m long, with 4 m intervals between pairs of boxes) starting five meters from the quarry edge (Fig. 1). To understand short term, within-year, propagule arrival, trap-boxes were collected at two times: one trap-box of the pair in the last week of October 2014 and the other in the last week of April 2015. Any plants growing in trap-boxes in the field were recorded when the traps were collected. We recorded whether or not pots were flooded at the time of sampling. Soil for DNA extraction was collected, dried with silica gel and stored airtight at room temperature. For samples gathered in October, DNA was extracted in December 3-17, 2014. For samples gathered from April, DNA was extracted in May 11-12, 2015. Five grams of soil were used for DNA extraction and 454-pyrosequencing following the approach described in Gazol et al. (2016). The remaining material per sample was spread out in 3–5 mm thick layer on medium oven-foil trays, filled with a 1:2 mixture of sand and commercially available soil (BiolanMust Muld, Biolan Oy, Kauttua, Finland) following the methodology of Metsoja et al. (2014) from May 21st to October 31st, 2015. The trays (n = 99) were placed in ambient conditions and kept moist. The positions of trays were randomised weekly to ensure homogeneous light and temperature conditions. To prevent colonization by propagules of nearby plant species, the trays were protected with a gauze. All seedlings were counted and removed after identification. As we did not treat the samples in any way besides watering, it is possible that not all seeds germinated; however, this represents more realistically which seedlings may actually emerge under field conditions. Five control trays with pure substrate were placed randomly among the trays containing sample material. Counts of emerging seedlings were used as estimates of the arrival of plant diaspores into the field trap-boxes.

2.3. Bioinformatics

Sequence reads were included in analyses only if they carried the correct forward primer and a barcode. Retained sequences were subjected to quality filtering and trimming: (1) barcode and primer sequences were removed, (2) sequences <170 bp were discarded, (3) sequences >520 bp were trimmed at the end to remove the AML2 primer, (4) sequence ends were truncated if the average quality score for 50 consecutive nucleotides was <20, (5) sequences where the average quality of remaining nucleotides was ≤25 were discarded, and (6) retained sequences were subjected to a chimera check using USEARCH (Edgar, 2010) and 38 chimeras were removed. Remaining sequences were subjected to a BLAST search against Maarj*AM* database for AM fungal identification (Öpik et al., 2010; version October 2015; 352 VT).

A BLAST match was considered a hit with a ≥97% sequence similarity threshold, the alignment length covering ≥95% of the shorter of the query (pyrosequencing read) and subject (reference