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Short communication

Arbuscular mycorrhizal fungi in Australian stormwater biofilters

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ABSTRACT

Stormwater biofilters are important tools for managing runoff in urban watersheds. To the authors' knowledge, there have been no accounts examining the presence of mycorrhizal fungi in biofilters. This plant–fungi relationship is an important interaction in most terrestrial ecosystems, playing a role in nutrient dynamics, water cycling, and soil organic matter decomposition. The presence of mycorrhiza in biofilters could have implications for nutrient and metal uptake in plants, and thus enhance removal of target pollutants. Additionally, the establishment, growth, and survivability of plants could be enhanced when roots are colonized by mycorrhizae. The aim of this study was to determine the extent of colonization by arbuscular mycorrhizal fungi in biofilters of varying ages in three Australian cities: Melbourne, Perth, and Sydney. The 32 biofilters surveyed supported 56 plant species, with dominant species belonging to the Cyperaceae, Iridaceae, Juncaceae, Onagraceae, Poaceae, and Xanthorrhoeaceae families. Mycorrhizal associations were identified from 4 of the 11 most dominant plant species from 9 different biofilters, but relatively low percentages of mycorrhizal colonization (3–25% colonization) were observed in biofilter plant roots. Mycorrhizal colonization was not related to biofilter age. These results demonstrate that mycorrhizal fungi colonize plant roots growing in biofilters. These findings provide useful evidence of the presence of mycorrhizal fungi in stormwater biofilters that support subsequent investigation into their roles in these systems.

1. Introduction

Stormwater biofilters are ecologically engineered treatment systems composed of engineered filter media planted with species adapted to live in both wet and dry conditions. Managing urban stormwater runoff using biofiltration can provide multiple types of ecosystem services (e.g., carbon sequestration, water quality improvement, urban heat mitigation, provision of biodiversity, etc.) (Grant et al., 2012; Hatt et al., 2009; Lundy and Wade, 2011; Wong and Brown, 2009). Despite extensive research demonstrating their effectiveness with respect to hydraulic and pollutant removal (Bratieres et al., 2008; Davis, 2007; Davis et al., 2001, 2006; Hsieh and Davis, 2005) and the importance of plant species selection (Barrett et al., 2013; Bratieres et al., 2008; Payne et al., 2014; Read et al., 2008), particularly for nutrient removal, the provision of biodiversity and existence of specific plant–soil biological relationships (e.g., mycorrhizal colonization of biofilter plant roots) by green infrastructure systems (a.k.a., Water Sensitive Urban Design, Sustainable Urban Drainage Systems, Low Impact Development, etc.) are rarely studied.

Arbuscular mycorrhizal fungi (AMF) symbiotically grow with host plants by providing water and nutrients to plant roots in exchange for energy. AMF have hyphae that access crevices too small for plant roots, delivering nutrients to the plant root cortex via specialized organs called arbuscules (Brundrett, 2009). AMF are associated with more than two thirds of terrestrial plant families (Wang and Qiu, 2006) and provide plants with increased access to soil water (Duan et al., 1996) and growth-limiting nutrients (Smith and Read, 2008), resistance to soil pathogens (Newsham et al., 1995), and tolerance to heavy metals (Hildebrandt et al., 2007). Mycorrhizal colonization of plants in stormwater biofilters could therefore increase removal of nutrients and metals and plant survivability during prolonged dry periods. Since water retention capacity of typical filter media is low (Payne et al., 2015) in biofilters, AMF could provide access to interstitial water in the filter media that plant roots could not reach. This could be particularly important in areas with prolonged dry periods, such as Perth, WA, or in systems designed to exfiltrate to the underlying layers (i.e., no submerged zone or liner in place to retain moisture).

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John et al. (2014) evaluated the presence of mycorrhizae in green roof plants and provided guidance for selecting species with stronger mycorrhizal associations. Others have investigated the use of AMF inocula to improve heavy metal uptake in polluted soils; some studies indicate AMF-colonized plants had increased heavy metal uptake (Liao et al., 2003; Whitfield et al., 2003) while others indicate decreased heavy metal uptake or no effect of AMF (Weissenhorn et al., 1995; Wu et al., 2007), suggesting the relationship between AMF and heavy metal uptake cannot be generalized (Weissenhorn et al., 1995). AMF have been detected in stormwater biofilter experimental columns, colonizing roots of *Melaleuca ericifolia* (Bratieres et al., 2008), but no information is available on studies presenting field observations of mycorrhizae in stormwater biofilters.

Soils and/or growth media are typically inoculated with mycorrhizae for the purposes of improving crop yields (Jeffries and Rhodes, 1987; Menge, 1983; Sharifi et al., 2007), establishment and productivity of plants used in horticulture (Azcón-Aguilar and Barea, 1997; Maronek et al., 1981), and restoration of terrestrial ecosystems (Danielson, 1985; Miller and Jastrow, 1992; Turnau and Haselwandter, 2002; Zhang et al., 2012). Stormwater biofilters, consisting of engineered soil planted with shrubs and grasses, are essentially terrestrial ecosystems with disturbed soils; Miller and Jastrow (1992) discuss the use of mycorrhizae inocula to restore soil health and promote plant growth following disturbance. Consequently, the benefits of mycorrhizae to establish plants in newly constructed biofilters could be significant (John et al., 2016). Plant cover in recently constructed systems depends largely on design parameters and varies from plants sparsely to completely covering the ground surface. However, it is unknown whether mycorrhizal colonization of biofilter plant roots occurs at all or persists over time.

This study aims to observe the presence of mycorrhizae in stormwater biofilters in Australia to determine whether mycorrhizal colonization of biofilter plant roots is affected by regional climate and biofilter age. Biofiltration has been a popular strategy to promote urban water sustainability in Australia for the past decade. Many systems have been installed in Australian cities, particularly in Melbourne, Victoria during and following The Millennium Drought under the 10,000 Rain Gardens project (Melbourne Water, 2013). For this reason, Australian cities provide a large number of biofilters of differing ages in relatively close proximity. Differences in rainfall between cities also provide opportunities to compare plants growing in biofilters located in different climatic conditions. Evidence of mycorrhizal colonization of biofilter plant roots could inform optimization studies whereby plant species that are found to be mycorrhizal in existing biofilters could be used to test the effects of their presence on biofilter performance and drought tolerance of plants.

2. Methods

2.1. Biofilter selection

In each city, biofilters were chosen from a list of biofilters compiled from published accounts and personal communications with municipal officials. Biofilters were selected to represent a range of ages (2–14 years), but maintain consistent design specifications.

2.1.1. Rainfall data

Mean annual rainfall (MAR) for each site was determined using the average annual precipitation measured at the closest rain gauge operated by the Australian Government's Bureau of Meteorology for the period of time between the year of construction of the biofilter to the sampling date. When data were not available for that time period, rainfall data from the next closest rain gauge, which was never more than 10 km from the biofilter, was used.

2.1.2. Biofilter location descriptions

2.1.2.1. Melbourne. On average, the 12 sampled biofilter sites in Melbourne, Victoria received MAR of 767 mm (Bureau of Meteorology, 2015) during the time between biofilter construction and sampling. Seasonally, rainfall was greater in winter months and lower in summer months; average monthly rainfall ranged from about 47 mm in January to 65 mm in October (Bureau of Meteorology, 2015). The selected study sites ranged in age (period of time between construction and date of sampling in October 2014) from 1.5 to 12 years. Median biofilter age and area were 3.4 years and 24 m^2, respectively.

2.1.2.2. Perth. On average, the 11 sampled biofilter sites in Perth, Western Australia received MAR of 738 mm (Bureau of Meteorology, 2015) during the time between biofilter construction and sampling. Typical of Mediterranean climates, rainfall was very low in summer months, with most rainfall occurring during winter months; average monthly rainfall ranged from about 10 mm in January to 160 mm in June (Bureau of Meteorology, 2015). The selected sites ranged in age (period of time between construction and date of sampling in November 2014) from 1.5 to 9 years. Median biofilter age and area were 5.5 years and 200 m^2, respectively.

2.1.2.3. Sydney. On average, the 9 sampled biofilter sites in Sydney, New South Wales received MAR of 1316 mm (Bureau of Meteorology, 2015) during the time between biofilter construction and sampling. Although more rainfall occurred in winter months than in summer, rainfall was relatively abundant throughout the year, with average monthly rainfall ranging from 70–80 mm in September–December to 130 mm in June (Bureau of Meteorology, 2015). These sites ranged in age (period of time between construction and date of sampling in November 2014) from 1.8 to 14 years. Median biofilter age and area were 5.3 years and 42 m^2, respectively.

2.2. Plant survey and mycorrhizal colonization of plant roots

We surveyed plant communities in each biofilter to determine dominant plant species by identifying plants to genera and species (where possible) in the field and visually estimating cover for the entire site. We collected photo vouchers for species we could not positively identify in the field. We used compared these photo vouchers to images on an online Australian plant guide (ANPSA, 2015) to identify plants to genera and species (where possible). For sites larger than 250 m^2, we randomly placed one 0.25-m^2 quadrat for every ~125 m^2 of biofilter, with the mean cover in the quadrats used to estimate plant cover.

Plant roots were collected from the dominant plant species at each site. For each dominant plant species at any site, one sample was composited from filter media cores (cores) collected adjacent to 3–4 different individual plants of the same species. Cores were collected by driving a 2.5-cm diameter chromium–molybdenum steel soil probe to rooting depth (10–30 cm below soil surface) at the base of individual plants that were isolated (i.e., not surrounded by other plant species). Holes made by probes were filled in with fine sand and existing surrounding material. Root samples were stored at 4 °C for less than 24 h before filter media was hand-washed from roots through a 600-μm sieve.

Subsamples (0.1–0.2 g dry weight) of washed roots were placed in a 10% (w/v) KOH solution in 20-mL scintillation vials and cleared in a water bath at 80 °C for 1–12 h, until visibly transparent (Vierheilig et al., 1998). Cleared roots were stained using the ink and vinegar method based on Vierheilig et al. (1998); the 5%