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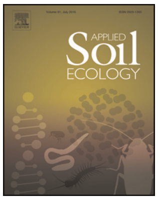

Alder trees enhance crop productivity and soil microbial biomass in tea plantations

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ABSTRACT

Monoculture farming systems lead to soils depleted of nutrients and diminished microbial functional diversity, disrupting processes crucial to maintaining soil health. The planting of trees in these monoculture systems is one way to improve soil nutrition and biodiversity. Therefore, the objective was how planting the N-fixing tree *Alnus nepalensis* (7 years old), into monoculture tea (*Camellia sinensis* var., *assamica*) plantations (32 years old), influences the soil fungal and bacterial communities, and how this impacts on tea productivity. Soil samples (0–15, 15–30, 30–60 cm depths) were collected from plantations of monoculture tea and tea interplanted with *A. nepalensis* trees. The samples were analyzed for basic soil properties and nutrients. Phospholipid fatty-acid analyses were conducted on the soil samples to determine the microbial functional groups and biomass of bacterial and fungal communities. Biomass of soil fungi and bacteria were 41% and 10% higher in the tea + *A. nepalensis* sites than in the tea monoculture sites, respectively. These higher levels were recorded despite there being no changes in the diversity of the soil fungi and bacteria, or the soil nutrition, between the different sites. Tea productivity increased between 52% and 72%, and is attributed to the increases in the soil community biomass. Ectomycorrhizal biomass, as well as Gram-positive, Gram-negative, and actinomycetes bacterial biomass, all increased ranging from 10% to 83%. These groups of organisms have been shown to contain plant growth promoting characteristics, contributing toward increased crop productivity. We provide clear evidence that *A. nepalensis* in tea plantations promotes the growth and development of the soil microbial communities and that this impacts on above ground productivity. This study highlights the benefits of introducing N-fixing tree species, such as *A. nepalensis*, into monoculture systems, and how this relates to soil health and harvest yield.

1. Introduction

Soil plays a crucial role in all terrestrial ecosystems, providing substrate, nutrition, water and a reservoir of organisms upon which the plants rely on for survival. An integral component of soils, and a component which has an important influence on above ground productivity, is the soil biota (Barrios, 2007). The soil microbial community is the largest constituent of soil biota and plays key roles in several ecological processes, such as organic matter decomposition, nutrient acquisition and cycling, and soil formation and aggregation (Zhou and Thompson, 2002). Soil microbes also contribute to plant productivity through the formation of symbiotic relationships (Bainard et al., 2013). Nitrogen-fixing bacteria and mycorrhizal fungi are the most widely known and studied of these symbiotic groups; they have a strong influence on plant productivity through the enhancement of nutrient acquisition and transport (van der Heijden et al., 2006). There are however, many other soil microbial groups that perform vital functions in maintaining above and below ground functions (Gui et al., 2012).

Amongst these microbial groups, soil fungi and Gram positive and negative bacteria play key roles in plant growth and production, eliciting both positive and negative above ground vegetation growth responses (Welbaum et al., 2004). Soil fungi contribute toward soil nutrient cycling, as well as influencing plant

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community composition through the development of symbiotic relationships and pathogenic infections (Bashan et al., 2004; Kernaghan, 2005; Liu et al., 2013; Moore et al., 2004). Gram negative and gram positive bacteria have a wide range of functions within the soil environment, including both free-living, associative and symbiotic N-fixation, antibiotic production, siderophore (iron chelating compounds) production, and sulfur oxidizing capabilities (Bashan et al., 2004; Kishore et al., 2005; Neeno-Eckwall et al., 2001; Welbaum et al., 2004).

Besides bacteria and fungi, actinomycetes, such as *Frankia*, are known to form symbiotic relationships with actinorhizal plants, developing root nodules, where N-fixation takes place (Wall, 2000). Alder trees, such as the Himalayan alder (*Alnus nepalensis*), are well known actinorhizal plants. *A. nepalensis* occurs naturally throughout the eastern Himalayan region and is a fast growing pioneer species. It is widely used in land restoration, reforestation projects, and has a long traditional use as an intercropping tree species (Carlson and Dawson, 1985; Chand et al., 1994; Goldman, 1961; Li et al., 2006).

Agroforestry systems are known to improve soil nutrient availability, soil microbial diversity and above ground productivity (Barrios et al., 2012; Li et al., 2013). More specifically, numerous studies have indicated that planting *Alnus* trees in agricultural settings have positive effects on plant growth, crop production and soil health (Binkley, 1983; Sharma et al., 2009; Vanlalhluna and Sahoo, 2009; Das et al., 2010).

The use of *A. nepalensis* as a shade tree in tea plantations is gaining popularity in Asia (Guo et al., 2006). Tea (*Camellia sinensis*) plantations dominate much of the agricultural landscape in Asia and usually occur as monoculture systems, impacting negatively on local biodiversity and soil health (Bainard et al., 2013). Thus, the incorporation of N-fixing trees into these monoculture systems is highly advantageous in terms of soil health and crop production, both of which have been shown to be negatively affected in monoculture stands (Guihua, 1996; Wang and Li, 2003). The use agroforestry systems for tea production, exemplified by the traditional practice of planting *C. sinensis* into existing forest systems, is an emerging practice in many tea-growing regions, such as northern Thailand and southwestern China (Sysouphanthong et al., 2010). However, the primary method of tea production remains monoculture systems, which are easier to manage and more economically viable for large-scale agriculture, albeit more detrimental to the environment.

Despite the large numbers of studies highlighting the benefits of using agroforestry and incorporating trees into the agricultural landscapes, few studies have focused on the changes brought about in the soil microbial community and the subsequent role that this plays in contributing toward healthier soils and crop production. Thus, the primary objective of this study was to elucidate how soil microbial communities, in particular bacteria and fungi, are influenced by the incorporation of the N-fixing tree *A. nepalensis* into monoculture plantations, and how this impacts on crop productivity.

2. Methods and materials

2.1. Site description and field experiment

This study was conducted in Changning County, Yunnan Province, China. Three tea (*C.sinensis*) plantation sites (Dazhuangwan (T), Xiaoluopo (X) and Ertaipo (E)) were selected. The geographical information for each site is given in Table S1. The three study sites were comparable in terms of climate, soil type, plant age, planting density, and management practices (pruning, weeding). Slope was comparable between the plots within each site. The climate is classified as temperate humid, with an annual rainfall of *ca.* 1268 mm, most of which falls between May and October. The mean annual temperature is approximately 15 °C and the maximum and minimum temperatures are 24.7 °C and 7.7 °C, respectively. The detailed meteorological data in 2012 is given in Fig. 1S according to China Meteorological Data Sharing Service System (2014).

Each study site consisted of a paired comparison between the agroforestry plots, that is tea plants + *A. nepalensis* trees, and the control plots, represented by a tea monoculture. The age (7 years) and planting density (*ca.* 660 trees ha^{-1}) of the *A. nepalensis* trees was similar in the agroforestry plots at all the study sites. The *A. nepalensis* trees were planted into the tea plantation by replacing

Table 1
The soil properties of soils taken from either monoculture (tea) or agroforestry (tea + alder) plots ($n = 3$), from three soil depths (0–15, 15–30, 30–60 cm).

	Sites	0–15 cm			15–30 cm			30–60 cm		
		PLFA	SE	*p* value	PLFA	SE	*p* value	PLFA	SE	*p* value
Organic matter ($g\,kg^{-1}$)	Tea	48.98	2.69	0.245	43.32	2.73	0.933	37.98	3.2	0.22
	Tea + alder	54.88	4.08		43.69	3.29		32.26	3.13	
Total N ($g\,kg^{-1}$)	Tea	2.01	0.13	0.614	1.73	0.13	0.961	1.59	0.14	0.221
	Tea + alder	2.1	0.12		1.72	0.16		1.34	0.14	
Total P ($g\,kg^{-1}$)	Tea	0.98	0.11	0.012*	1.98	0.96	0.18	1.02	0.16	0.083
	Tea + alder	0.65	0.05		0.63	0.08		0.64	0.13	
Total K ($g\,kg^{-1}$)	Tea	12.03	0.31	0.632	11.68	0.39	0.332	11.73	0.21	0.076
	Tea + alder	12.37	0.62		12.42	0.63		12.67	0.45	
Available N ($mg\,kg^{-1}$)	Tea	172.04	9.24	0.116	163.85	9.99	0.793	162.54	13.57	0.184
	Tea + alder	197.21	12.02		159.66	12.06		132.87	16.52	
Available P ($mg\,kg^{-1}$)	Tea	10.67	2.19	0.173	11.01	2.53	0.052	10.48	3.6	0.462
	Tea + alder	7.28	0.91		5.16	1.15		6.84	3.23	
Available K ($mg\,kg^{-1}$)	Tea	116.64	33.27	0.205	72.01	21.97	0.265	51.4	13.73	0.721
	Tea + alder	68.68	14.44		44.09	10.03		42.76	19.42	
pH	Tea	4.94	0.13	0.736	5.33	0.31	0.182	5.1	0.14	0.054
	Tea + alder	5	0.14		4.89	0.05		4.74	0.1	

* $p < 0.05$.