

EXPLORING THE MICROBIAL ACTIVITIES IN THE RHIZOSPHERES OF EXOTIC BAMBOOS

Abhaya Garg^a, Solomon Das^b, Y.P. Singh^c, Pawan Sharma^d

^{a,b,c}Forest Research Institute, Dehradun, India; ^dCSWCRTI, Chandigarh, India

Email: abhay_garg11@yahoo.co.in

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Abstract: We need to believe that our atmosphere is changing- uneven climate patterns will draw us to the cliff of alarming situation of global warming. With this shift of climatic patterns, it has influenced the above and below ground entities of ecosystem. The objectives of this study were to quantify mycorrhizal association and glomalin content in seven exotic bamboo species raised in bambusetum of Forest Research Institute, Dehradun. The study further addresses activities of soil microbial community in terms of moisture, respiration, enzymes (dehydrogenase and phosphatase), carbon, aggregation and their inter-relationship besides their possible role in carbon sequestration in relation to bamboo-mycorrhizae. The study observed that *Melocanna baccifera* (40.91 μ g/gm/hr) recorded significantly maximum soil respiration. The dehydrogenase activity was measured highest of 92.95 μ g/25ml/g/24hr in *Dendrocalamus giganteus* while lowest of 12.61 μ g/25ml/g/24hr was quantified for *M. baccifera*. The maximum acid phosphatase activity was recorded in *D. giganteus* (18.914mg/g/hr). The alkaline phosphatase activity was recorded highest in *Cephalostachyum pergracile* (0.1502mg/g/hr) while lowest was registered in *Bambusa multiplex* (0.0432mg/g/hr). The highest microbial biomass carbon was quantified in *Bambusa polymorpha* (518.97mg/kg) and lowest was in *D. giganteus* (102.89mg/kg). Maximum root colonization was found in *Bambusa tulda* (59.05%) with maximum spores were counted in the soil collected from the root zone of *D. giganteus* (52.56/ml) and the lowest spore count was recorded in *B. multiplex* (13.22/ml). The maximum value of glomalin content was recorded in *C. pergracile* (84.09 μ g/ml) and minimum was found in *B. multiplex* (48.24 μ g/ml). The study explored the potential of soil microbes and mycorrhizae along with these exotic bamboos in mitigating the elevated CO₂, which probably becomes a suitable candidate in sequestering the carbon dioxide.

Keywords: Bamboo, Glomalin, Soil enzymes.

Postal Address: H-4, 9/198, Abhishek Residency II, Sector-3, Rajendra Nagar, Sahibabad, Ghaziabad-201005

INTRODUCTION

Bamboos are the giant grasses belonging to the family Poaceae and the subfamily Bambusoideae. There are approximately 1,250 species in 90 genera (Scurlock et al., 2000; Dura and Hiura, 2006; Lobovikov et al., 2007) ranging in size from the dwarf species, growing only 10 cm high, to the giant canes, towering well above many tree heights in temperate forests. The bamboo physiology is responsible for high biomass and substantial carbon removals from the atmosphere. By accumulating organic matter and counteracting erosion, bamboos have reversed soil degradation in exploited landscapes (Christanty et al., 1997; Zhaohua and Yang, 2004; Singh et al., 2006; Hooda et al., 2007; Marsh and Smith, 2007; Mohamed et al., 2007). Bamboo stands, as natural rafts or tightly woven root mats, offer shelter against earthquakes, floods and tsunamis. In addition to these environmental benefits, bamboos provide raw material for about 1,500 known commercial products (Scurlock et al., 2000). Generally, plants invest a lot of energy in root exudation, which depends on light intensity, temperature, type of plant, nutritional state of plants, stress factors, microbial activity in the rhizosphere and type of soil. Most of the important commercial bamboo species have a coarse surface spreading root system. This provides a suitable environment for the microbial association in relation to nutrients acquisition. Bhattacharya et al. (1999) studied mycorrhizal dependency, phosphorous utilization efficiency and the relevance of mycorrhiza for bamboo cultivation in laterite wasteland. Due to the high diversity of chemical influences in the rhizosphere of different plants, roots drive specific selections of

microbes out of the almost infinite pool of soil microbial diversity. Ample evidence exists which clearly demonstrates the selection of microbes by the roots of plants. The roots provide a specific microhabitat for the proliferation of a particular subset of soil microbes (Hartman *et al.* 2008). In the present study, we took seven bamboo species *Dendrocalamus giganteus* Munro, *Bambusa polymorpha* Munro, *Bambusa tulda* Roxb., *Bambusa nutans* Wall. Ex Munro, *Bambusa multiplex* (Lour) Schult., *Cephalostachyum pergracile* Munro and *Melocanna baccifera* (Roxb.) Kurz., which were different in their age, genus and species. The objectives of this study were to quantify mycorrhizal association and glomalin content in seven exotic bamboo species raised in the bambusetum of the Forest Research Institute, Dehradun, India. The study further addresses activities of soil microbial community in terms of moisture, respiration, enzymes (dehydrogenase and phosphatase), carbon, micro and macro aggregation and their inter-relationship, besides their possible role in carbon sequestration in relation to bamboo-mycorrhizae.

MATERIALS AND METHODS

The study area includes compartment no. 3 of the Bambusetum of the Forest Research Institute, Dehradun, where all seven bamboo species (*Bambusa multiplex* (Lour) Schult., *Bambusa nutans* Wall. Ex Munro, *Bambusa polymorpha* Munro, *Bambusa tulda* Roxb., *Cephalostachyum pergracile* Munro, *Dendrocalamus giganteus* Munro and *Melocanna baccifera* (Roxb.) Kurz are situated. It lies between the latitude of 30°-30° 32'N and longitude of 77° 43'-78° 24'E. The site receives an annual rainfall of about 2,000 mm; most of which is concentrated during the monsoon season (June- September). Annual temperatures range from 0.2-36.9°C. Samplings were done in the month of April, 2010. Surface layers were scraped and drilled to 30 cm deep to take soil samples. Four soil samples were taken from each plant. Sugar gradient centrifugation method (Gerdemann and Nicholson, 1963; Daniel's and Skipper, 1982) was employed for collection of spores from the soil samples. Grid line - Intersect method (Giovanetti and Mosse, 1980) was used for the measurement of root colonization. Soil samples were air dried, sieved through a 2mm sieve and analyzed. Soil texture was determined by Hydrometer method using the USDA textural classification chart. Soil pH (Schofield and Taylor, 1955), soil moisture (Gardner, 1986) and organic carbon (Walkley and Black, 1934) were quantified. Total nitrogen of the soil samples were determined by regular Kjeldal method (Bremner, 1965), available P by Bray's-I (Bray and Krutz, 1945) and Bray's-II method (Olsen *et al.*, 1954) and available K by ammonium acetate extraction and flame photometric determination (Table1). Microbial activity was assessed using parameters such as soil respiration; dehydrogenase activity (Lenhard, 1956); phosphatase activity (Tabatabai and Bremner, 1969); microbial biomass (Powlson & Jenkinson, 1976); soil aggregation (Elliot and Cambardella, 1991) and glomalin was extracted using Wright and Upadhyaya (1996) methodology.

Statistical Analysis: The statistical analysis was carried out in the Directorate of Statistics, Indian Council of Forestry Research and Education, Dehradun. Different characters of field survey were analyzed by One-Way ANOVA using Genstat 5.0. The difference between the treatments was tested at 5 percent level of significance. Correlation analysis was also performed between different physico-chemical and biological parameters using STATISTICA 7.

Table 1. Physico-chemical analysis of soil collected from root zone of different exotic bamboos of FRI bambusetum

Soil character	Bamboo species						
	<i>Bambusa multiplex</i>	<i>Bambusa nutans</i>	<i>Bambusa polymorpha</i>	<i>Bambusa tulda</i>	<i>Cephalostachyum pergracile</i>	<i>Dendrocalamus giganteus</i>	<i>Melocanna baccifera</i>
Partical size distribution							
Sand (%)	69.5	69	67.65	54.85	62.1	68.1	70.7
Silt (%)	17	19.25	17.65	25.2	21.9	18.1	17.6
Clay (%)	13.5	11.8	14.7	19.9	16	13.8	11.7
Textural Class	Loam	Loam	Loam	Silty loam	Loam	Loam	Loam
Soil reaction (pH)	5.82	5.48	5.65	5.62	6.86	6	6.94
Electrical conductivity (desi simon/m)	0.12	0.18	0.19	0.19	0.18	0.17	0.26
Organic C (%)	2.12	3.72	2.71	2.72	4.33	3.55	4.86
Organic matter (%)	3.65	6.41	4.67	4.69	7.46	6.12	8.38
Nitrogen (%)	0.19	0.31	0.25	0.25	0.36	0.31	0.41
Available P (ppm)	2.4	11.2	13.6	18.4	20.8	6.4	10.4
Available K (ppm)	54	81	90	236.5	120	99	170.5

RESULTS AND DISCUSSIONS

The various physical and biological soil microbial parameters of the seven exotic species of bamboo of FRI bambusetum were quantified. These parameters include soil moisture, respiration, enzymes, carbon and aggregation in relation to the terrestrial carbon sequestration. The root colonization and spore count of mycorrhizae were also studied besides glomalin. There are many processes related to land-atmosphere carbon exchanges that soil microbes and their metabolic activity can influence.

Soil Moisture: Soil moisture values showed the overlapping trends as different bamboo species had at par values (Table2). The maximum soil moisture was recorded in *C. pergracile*, which was at par with *B. nutans* and *D. giganteus*. On the other extreme, minimum value was recorded in *B. Polymorpha*, which was at par with *B. multiplex* and *M. baccifera*. Statistically at par grouping were also observed between *B. tulda* and *B. nutans*.

Soil Respiration: Soil collected from the root zone of *M. baccifera* registered significantly more and highest respiration followed by *D. giganteus*. *B. multiplex* recorded the lowest soil respiration that was significantly different than rest of the 6 species (Table2). The respiration of the soil under 3 species of *Bambusa*, *B. nutans*, *B. polymorpha* and *B. tulda* was at par along with *C. pergracile*. Soil respiration represents the integrated CO₂ flux of root respiration and heterotrophic respiration including mycorrhizal respiration (Vicca et al., 2009). Colonization by abuscular mycorrhizal fungi (AMF) can enhance below

ground respiration rates (Langley et al., 2005), although this was not observed in all studies (Cavagnaro et al., 2008). The findings of the present investigation also showed the similar observations as the soil respiration of different bamboo species were positively related with mycorrhizal spore population. The soil respiration in the root zone of *Bambusa* species was lower and significantly at par within genus as compared to the other genera (*Cephalostachyum*, *Dendrocalamus* and *Melocanna*) of bamboo. Soil respiration was found to be highly correlated to all bamboo species and glomalin content of their soil.

Dehydrogenase Activity: The dehydrogenase activity was statistically exclusive, for most of the species of bamboos except *M. baccifera* and *B. nutans* which were at par (Table2). It was measured highest in *D. gigantius*, which was significantly more than all other species. Two more species i.e. *B. tulda* and *C. pergracile* also had high dehydrogenase activity that was mutually exclusive statistically. The lowest dehydrogenase activity was quantified for *M. Baccifera*, which was at par with *B. nutans*. Soil Dehydrogenase activity was found to be correlated with the moisture percentage. Thus, it may be concluded that moisture content of the soil influences the dehydrogenase activity. The dehydrogenase activity within all *Bambusa* sp. as well as among 4 genera of bamboo was significantly different.

Acid Phosphatase Activity: The maximum acid phosphatase activity was recorded in *D. gigantius*, which was significantly higher than other six bamboo species (Table2). The minimum activity was registered in *M. baccifera* that was at par with *B. multiplex* and *C. pergracile*. All the four species of *Bambusa*, *B. multiplex*, *B. polymorpha*, *B. nutans* and *B. tulda* were significantly different from each other.

Alkaline Phosphatase Activity: The alkaline phosphatase had significant differences among the bamboo species (Table2). It was recorded highest in *C. pergracile* while lowest was registered in *B. multiplex*. Two more species i.e. *B. polymorpha* and *D. gigantius* also had reasonably high alkaline phosphatase activity next to *C. pergracile* but mutually exclusive. The activity of acid and alkaline phosphatases was found to correlate with organic matter. Like this observation, it was found similar in other various studies (Guan 1989; Jordan and Kremer, 1994; Aon and Colaneri, 2001). Alkaline phosphatase activity was found to be correlated with microbial biomass carbon, confirming its microbial origin. Irrespective of genera and species of all the seven exotic species of bamboo, the alkaline phosphatase activity and microbial carbon were significantly different, while acid phosphatase activity was significantly different only among all *Bambusa* species. Alkaline phosphatase activity was also found to be positively correlated with available P, while acid phosphatase was negatively correlated with it. It may be because of the reason that acid phosphatase activity increases when there is deficiency of P in the soil (Nakas et al., 1987; Chrost, 1991), however, the root zone soil of these species were not P deficient leading to negative relationship.

Table 2. Soil enzymes and microbial activities of soil collected from root zone of different exotic bamboos of FRI bambusetum

Species	Soil moisture (%)	Soil respiration ($\mu\text{g/gm/hr}$)	Dehydrogenase activity ($\mu\text{g}/25\text{ml/g}/24\text{hrs}$)	Acid Phosphatase activity (mg/g/hr)	Alkaline Phosphatase activity (mg/g/hr)	Macroaggregate (%)	Microaggregate (%)	Microbial biomass carbon (mg/kg)	Root colonization (%)	Spore count (no./ml of soil)	Glomalin content ($\mu\text{g/ml}$)
<i>Bambusa multiplex</i>	3.49	17.26	20.29	0.15	0.04	40.13	40.02	148.54	34.77	13.22	48.24
<i>Bambusa nutans</i>	4.39	21.39	13.64	1.36	0.06	45.13	35.67	208.14	27.62	13.91	73.76
<i>Bambusa polymorpha</i>	3.38	22.61	18.08	1.19	0.14	40.48	39.93	518.97	31.20	21.71	69.86
<i>Bambusa tulda</i>	4.20	22.61	68.58	1.62	0.06	48.90	32.31	325.80	58.21	32.34	59.08
<i>Cephalostachyum pergracile</i>	4.81	21.39	43.15	0.15	0.15	47.10	35.98	414.53	41.49	23.36	84.09
<i>Dendrocalamus giganteus</i>	4.69	29.69	92.95	18.91	0.10	33.17	35.19	102.89	41.57	52.36	62.97
<i>Melocanna baccifera</i>	3.50	40.91	12.61	0.11	0.08	47.73	35.35	266.64	47.26	32.10	83.05
S.E.M.	0.44	2.95	1.90	0.008	2.87×10^{-7}	2.98	3.26	4.35	5.43	2.76	0.22
C.D. (LSD) (5%)	0.46	2.47	1.98	0.128	0.001	2.48	2.59	3.0	3.35	2.39	0.67

Soil Aggregation: The percent macroaggregates were higher than microaggregates in all the bamboo species except *D. giganteus* (Table 2). Macroaggregates showed the overlapping trends over bamboo species while it was reasonably discrete in case of microaggregates. The maximum macroaggregate percentage was registered by *B. tulda* that was at par with *C. pergracile* and *M. baccifera*. While minimum was recorded in *D. giganteus*, which was significantly lower from all other species. Maximum microaggregate percentage was recorded in *B. multiplex* and minimum was registered in *B. tulda* that was significantly less than other bamboos. Microaggregate percentages of *B. nutans*, *C. pergracile*, *D. giganteus* and *M. baccifera* recorded at par values. Microaggregates for *B. multiplex* and *B. polymorpha* were also at par.

Microbial Biomass Carbon: The variations in soil microbial biomass carbon of the bamboo species were statistically significant and discrete (Table 2). The highest microbial biomass carbon was quantified in *B. polymorpha* and lowest was in *D. giganteus*. The microbial biomass carbon had ascending trend as follow: *D. giganteus* < *B. multiplex* < *B. nutans* < *M. baccifera* < *B. tulda* < *C. pergracile* < *B. polymorpha*.

Root Colonization: Minimum root colonization was recorded in *B. nutans*, while maximum was found in *B. tulda* (Table 2). Except *B. tulda*, other members of *Bambusa* had lower root colonization among 4 genera of exotic bamboos. *M. baccifera* was next to *B. tulda* in root colonization, which was significantly different from other species. The values of root colonization in *C. pergracile* and *D. giganteus* were at par.

Spore Count: The lowest spore count was recorded in *B. multiplex*, which was at par with *B. nutans* (Table 2). Maximum spores were counted in the soil collected from the root zone of *D. giganteus*. *M. baccifera* and *B. tulda* were next to the *D. giganteus* and mutually at par. *B. polymorpha* and *C. pergracile* had statistically similar spores in their root zone.

Glomalin Content: Significant differences were recorded in the glomalin content among the exotic bamboo species (Table 2). The maximum value was recorded in *C. pergracile* and minimum was found in *B. multiplex*.

Soil pH: All the soils were on slightly acidic side. In soil of seven species, *B. multiplex*, *B. nutans*, *B. polymorpha* and *B. tulda* were more acidic than remaining three (*C. pergracile*, *D. giganteus* and *M. baccifera*; Table 2).

Organic Carbon: Percentage organic carbon of *M. baccifera* was maximum followed by *C. pergracile* and *B. multiplex* had the minimum. *B. polymorpha* and *B. tulda* were mutually at par.

Soil Nitrogen: Percent nitrogen in soil was found maximum in *M. baccifera* followed by *C. pergracile* with lowest recorded in *B. multiplex*.

Available Soil Phosphorous: Soil from *C. pergracile* had the maximum soil P followed by *B. tulda* with *B. polymorpha* had the minimum soil P.

Available Soil Potassium: Soil from *B. tulda* had the maximum soil P followed by *M. baccifera* with *B. multiplex* had the minimum soil K.

Correlation between soil enzymes and parameters: Irrespective of different bamboo species, a significant correlation was seen between acid phosphatase and soil dehydrogenase also with macroaggregates. The mycorrhizal spore count was strongly correlated with dehydrogenase and acid phosphatase enzyme. Organic matter, organic content and soil nitrogen was found highly correlated with Glomalin. The available soil P was found to be highly negatively correlated with spore count of soil and significantly correlated with silt soil texture. Also, the soil potassium was found to be highly correlated with arbuscular root colonization and negatively correlated with microaggregation of soil. Soil texture might have influenced the microbial biomass carbon as it is positively related to this parameter. The percent macroaggregates showed positive correlation with various parameters like silt and clay percentage, pH, EC, available P and K, organic matter, microbial biomass carbon, root colonization and glomalin content. It may be inference that these parameters influenced soil aggregation through macroaggregates. In contrast, percent microaggregate showed positive correlation only with the sand fraction of soil texture. The foregoing correlations highlighted the fact that fungal activity influences the macroaggregate formation as earlier reported by Tisdall and Oades (1982) and Schutter and Dick

(2002). *B. tulda* had maximum percent macroaggregate but, in contrast, it had minimum percent microaggregate. The macroaggregates may relate to high mycorrhizal root colonization of *B. tulda* roots. On the other hand, two recent studies related to these exotic bamboos hinted that the bacterial load (e.g. fluorescent pseudomonads) in the root zone of *B. tulda* was quite low resulting in poor macroaggregate formation (Rawat, 2010). Similar observations were recorded by Tisdall and Oades (1982) and Schutter and Dick (2002). Except *B. tulda* other species of *Bambusa* had lower percent macroaggregate as the root colonization of these species was also recorded to be reasonably low. Glomalin content in all the exotic bamboo species was significantly different. There was no correlation between glomalin content with root colonization, thus it may be concluded that mycorrhizal root colonization may not influence the glomalin content as also concluded by Lovelock et al. (2004) that production rates of glomalin are not always correlated with AM abundance in soil and the two variables can be decoupled at short time scales (Lutgen et al., 2003). The positive correlation of this soil fungal origin glycoprotein with the macroaggregate may be responsible for macroaggregate stabilization.

CONCLUSION

An understanding of soil microbial ecology is central to our ability to assess terrestrial carbon cycle–climate change. Constant rise in global temperature has altered the carbon cycle to the greater extent. Continuous soil degradation and deforestation has contributed in elevated atmospheric CO₂. Present study has revealed that the role of arbuscular mycorrhizae and other microbes activities in maintaining the soil carbon to high extent in association with bamboo roots. The strong correlation between glomalin and organic matter, nitrogen, microbial biomass carbon and macroaggregate reveals the very nature of glomalin as glue, which promotes soil aggregation, which in turn, holds the nutrients and soil carbon in more stable form for longer period. Hence, this study explores the potential of soil microbes and mycorrhizae along with these exotic bamboos in mitigating the elevated CO₂, which probably becomes a suitable candidate in sequestering the carbon dioxide. However, the complexity of the soil microbial community and its many roles, coupled with the myriad of ways that climate and other global changes can affect soil microbes, hampers our ability to draw firm conclusions on this topic.

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