Short communication

Earthworms (*Perionyx excavates* Perr.), cowdung, and leaf residues alter soil physico-chemical and microbial properties in a traditional agroecosystem of Arunachal Pradesh

Sumpam Tangjang[1], A. Arunachalam[2]*, K. Arunachalam[3], and D. Balasubramanian[2]

[1]*Department of Botany, Rajiv Gandhi University, Rono Hills 791 112, Itanagar, Arunachal Pradesh, India;* [2]*Division of Natural Resources Management, Indian Council of Agricultural Research, Krishi Anusandhan Bhavan II, Pusa, New Delhi 110012;* [3]*School of Environment & Natural Resources, Doon University, Kedarpur, Dehra Dun 248001, Uttarakhand.*

Received 30 May 2012; received in revised form 6 September 2012; accepted 6 September 2012.

Abstract

The effects of native earthworms (*Perionyx excavates*), cowdung, and leaf residues on soil physico-chemical properties and microbial biomass C, N and P were studied for three months in a laboratory. The soil in the boxes was amended with leaf residues (arecanut, banana, bamboo, and colocasia) and cowdung (1:1 ratio) to which a minimum of five earthworms were introduced. Soil pH, moisture content, total nitrogen content, and available phosphorous were significantly (p<0.05) higher in the treated soil. Among the treatments, earthworm population was high in the banana and bamboo residues. Nonetheless, population and biomass C, N, and P were higher in the pots treated with earthworm + cowdung + leaf residues, compared to soil with only earthworm. Overall, earthworms in conjunction with high quality plant residues would enrich the soil nutrient status vis-à-vis crop production in the humid tropics.

Keywords: Organic amendment, Tropical soils, Soil properties, Microbial population.

The hill farmers of north-eastern India, particularly Arunachal Pradesh, practice traditional farming in which chemical or organic inputs are hardly used. They maintain a low external input system, leading to low productivity and depletion of soil nutrients. Earthworms play an important role in sustaining soil productivity through their effects on soil organic matter decomposition and increasing the availability of plant nutrients (Anderson and Ingram, 1989). During vermicomposting, the earthworms release easily assimilable substances (e.g., mucus for microbiota: Brown and Doube, 2004) and the earthworm casts have a favourable nutrient content and microbiota composition. Although considerable work has been done on vermicomposting of various organic materials such as animal dung, agricultural waste, forestry wastes, and food wastes, less attention was paid to the extent to which earthworms influence the dynamics of soil microbial biomass. The present study was intended to understand how combinations of earthworm, leaf residues, and cowdung affects the soil physical and chemical properties, microbial biomass, microbial population, and earthworm population.

The laboratory experiment was conducted at the North Eastern Regional Institute of Science & Technology, Nirjuli, Arunachal Pradesh (27º60´N, 94º21´E, 118 m elevation,) in northeast India from September to November 2009. Fresh leaf residues

* Author for correspondences : Fax +91 11 25842716; Email <arun70@gmail.com>.

from four dominant plant species viz., *Areca catechu* (arecanut), *Bambusa pallida* (bamboo), *Musa glauca* (banana), and *Colocasia esculenta* (colocasia) were collected along with cowdung from the homestead gardens and the leaf residues cut into small pieces. All materials were oven-dried at 60°C for 48 h and powdered using Wiley Mill for chemical analysis. Ash and carbon content were determined by igniting 1g sample at 550ºC for 6 h in a muffle furnace (Allen et al., 1974). Total nitrogen content of different samples was estimated using Pelican semi-automatic N analyser.

To prepare a uniform feed material, 10 g plant residue was mixed with cowdung in the ratio of 1:1 on a weight basis and placed in culturing boxes (15x25x9 cm) filled with 1 kg soil collected from the nearby agriculture field. Before amending, the soils were sterilized at 180 to 200°C in a hot air oven. At least five juvenile *Perionyx excavates*, the most common and abundantly found earthworm, were introduced into each culturing box. Nine treatments (control; soil+earthworm; soil+arecanut residue + earthworm; soil+arecanut residue +cowdung + earthworm; soil + banana residue + earthworm; soil + banana residue + cowdung + earthworm; soil + bamboo residue + earthworm; soil + bamboo residue + cowdung + earthworm; soil + colocasia residue + earthworm; soil + colocasia residue + cowdung + earthworm) were used with three replications each (Table 1). The culture boxes were supplied with equal amount of water at regular intervals. Total population of earthworms was estimated by counting the number of adults, juveniles, and cocoons from each culture boxes at 15 days interval for three months. The sum of the weight of adults, juveniles, and cocoons from each culture boxes was expressed as total earthworm biomass.

After three months, soil samples were collected from each culture box, sieved through a 2 mm sieve, and divided into two parts: one part was used for determining pH, moisture content (gravimetric method), ammonium N (indophenol blue method), nitrate N (phenol disulphonic acid method), and available P (molybdenum blue method); and the other part was air-dried for determining organic C (rapid titration method) and total N (semi-micro kjeldahl method). The soil bacterial population was estimated by Waksman (1952) method using nutrient agar medium at 10^5 dilution and the fungal populations were determined by dilution plate method (Johnson and Curl, 1972) using Martin Rose Bengal agar medium at 10^3 dilutions in water. The inoculated petri-dishes were incubated at 30±1°C for 24 h for bacteria and at 25±1°C for five days for fungi. The colonies developed were counted using a digital colony counter. The final counts were expressed on per gram soil dry weight basis. Microbial biomass C, N, and P were estimated following chloroform fumigation incubation (CFT) and extracted using 0.5 M K_2SO_4 and $NaHCO_3$ respectively (Jenkinson and Powlson, 1976). Fumigation and incubation extraction method of microbial biomass was corrected to account for the added chloroform C. The data were analysed using ANOVA to test the significance level of variations in soil physico-chemical and microbial properties under different treatments.

Table 1. Initial residue quality of the materials used in the laboratory study.

Chemical properties	Cowdung	Arecanut leaf	Banana leaf	Bamboo leaf	Colocasia leaf
Carbon (%)	17.44	40.77	26.39	28.52	23.45
Nitrogen (%)	0.80	1.26	0.28	1.02	1.39
Phosphorus (%)	0.87	0.24	0.17	0.31	0.12
Lignin (%)	–	29.15	14.89	24.51	13.23
C/N	21.80	32.36	38.80	27.99	16.87
Lignin/N	–	23.13	53.18	24.03	9.52

As can be seen from Table 2, soil pH decreased following the addition of earthworms and plant residues within three months. Available nitrogen i.e., ammonium N and nitrate N, and available P, however, increased. Soil organic C was significantly (p<0.05) higher in the bamboo residues + cowdung amended soils, compared to other treatments. On the other hand, soil ammonical N and nitrate N in the banana + cowdung treatment were greater (p<0.05) than the remaining treatments. The earthworm population in the soil was particularly affected by the addition of cowdung and leaf residues. Addition of arecanut and banana residues along with cowdung increased their number by 20 and 40 to 60% respectively (Table 2). The microbial population also was higher in the pots with earthworm + cowdung + leaf residues. Fungal population and bacterial population were, however, highest in the soil treated with arecanut residue + cowdung. Microbial biomass C and P were highest for the soil + bamboo residue + cowdung + earthworm among different residue treatments. Whereas, microbial

Table 2. Soil physico-chemical and biological characteristics under different treatments after three months of incubation.

	Treatments									
Soil properties	T_0	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	T_9
Physico-chemical Properties										
Soil pH	5.97^{a}	5.83^{a}	5.87^{a}	5.75^{a}	5.65^{a}	5.49^{a}	5.51^{a}	5.53^{a}	5.52^{a}	5.37^{a}
Soil moisture (%)	34.10^{a}	23.56^{b}	26.53^{b}	29.01^{b}	30.52^{ab}	21.50^{b}	18.92^{c}	25.84^{b}	22.26^{b}	34.08^{ab}
Organic C (%)	1.05^{a}	1.47^{a}	2.45^{b}	3.26^{c}	2.48^{b}	2.89^{b}	2.11^{b}	3.59^{c}	1.47^{a}	1.45^{a}
Total N (%)	0.78^{a}	0.65^{a}	1.02^{b}	1.08^{b}	0.55^{a}	0.57^{a}	0.45^{a}	0.47^{a}	0.31^{a}	0.45^{a}
C/N	1.35^{a}	2.26^{b}	2.45^{b}	3.01^{c}	4.51^{d}	5.07^{e}	4.69^{de}	7.64^{f}	4.71^{d}	3.22^{c}
NH_4-N (µg g^{-1})	10.17^{a}	10.83^{a}	13.84^{b}	14.80^{c}	14.71^{bc}	15.62^{d}	10.41^{ab}	12.75^{e}	8.45^{f}	9.94^{g}
NO_3-N (µg g^{-1})	10.40^{a}	14.29^{b}	16.53^{c}	18.53^{d}	13.54^{ab}	23.51^{e}	20.03^{f}	21.42^{f}	15.49^{ab}	19.09^{g}
Available-P (µg g^{-1})	3.90^{a}	6.17^{b}	6.55^{b}	7.88^{c}	7.44^{c}	9.52^{d}	11.96^{e}	15.86^{f}	8.94b^{c}	11.11^{e}
Microbial Properties										
Bacteria (10^5 dil.)	102^{a}	74.99^{b}	149.92^{c}	261.11^{d}	76.84^{b}	85.20^{ab}	26.19^{e}	33.67^{e}	42.67^{f}	58.33^{g}
Fungi (10^3 dil)	7.0^{a}	35.00^{b}	70.67^{c}	108.00^{d}	53.96^{bc}	83.60^{e}	17.78^{f}	29.43^{g}	13.07^{f}	21.74^{g}
Microbial C (µg g^{-1})	84.59^{a}	460.47^{b}	561.32^{c}	609.53^{d}	324.07^{e}	286.29^{f}	655.70^{g}	714.66^{h}	52.51^{i}	185.47^{j}
Microbial N (µg g^{-1})	12.29^{a}	14.55^{b}	23.53^{c}	25.77^{c}	18.84^{d}	25.99^{c}	8.51^{e}	15.64^{ab}	17.10^{e}	12.10^{a}
Microbial P (µg g^{-1})	1.81^{a}	2.25^{b}	2.73^{b}	3.07^{c}	3.18^{c}	3.30^{c}	4.49^{d}	4.76^{d}	1.09^{a}	1.71^{a}
Microbial C/MBN	6.68^{a}	31.64^{b}	23.85^{c}	23.62^{c}	17.20^{d}	11.01^{e}	77.02^{f}	45.68^{g}	3.06^{h}	15.33^{d}
Microbial C/MBP	46.73^{a}	204.35^{b}	205.36^{b}	198.22^{c}	102.02^{d}	86.67^{e}	146.04^{f}	150.03^{g}	48.32^{a}	108.46^{c}
Microbial N/MBP	6.79^{a}	6.46^{a}	8.61^{b}	8.39^{b}	5.93^{c}	7.87^{d}	1.90^{e}	3.28^{f}	15.74^{g}	7.07^{ad}
Earthworm Biological Properties										
Population Initial (No.)	0	5	5	5	5	5	5	5	5	5
Population Final (No.)	0	5^{a}	5^{a}	6^{b}	7^{c}	8d	4^{a}	5^{a}	5^{a}	5^{a}
Mortality/natality (%)	0	0^{a}	0^{a}	20^{b}	40^{c}	60^{d}	–20^{e}	0^{a}	0^{a}	0^{a}
Weight (g earthworm^{-1})	0	0.18^{a}	0.27^{a}	0.27^{a}	0.21^{a}	0.20^{a}	0.26^{a}	0.24^{a}	0.22^{a}	0.21^{a}
Weight Initial	0	0.19^{a}	0.32^{b}	0.37^{b}	0.28^{c}	0.29^{c}	0.28^{c}	0.27^{c}	0.26^{c}	0.29^{c}
Loss/gain (%) Final	0	5.56^{a}	18.51^{b}	37.03^{c}	25.00^{d}	45.00^{e}	7.69^{f}	12.5^{g}	18.18^{b}	38.10^{c}

T_0=Control; T_1=Soil + earthworm; T_2=Soil + arecanut residue + earthworm; T_3=Soil + arecanutresidue + cowdung + earthworm; T_4=Soil + banana residue + earthworm; T_5=Soil + banana residue + cowdung + earthworm; T_6= Soil + bamboo residue + earthworm; T_7=Soil + bamboo residue + cowdung + earthworm; T_8=Soil + colocasia residue + earthworm; T_9=Soil + colocasia residue + cowdung + earthworm.

[a]Means followed by same superscript within a row are not significantly different.

N was greater in the soil + banana residue + cowdung + earthworm than in other treatments. Furthermore, main factor ANOVA showed that microbial C, N, and P were significantly (p<0.05) different among the treatments.

Soils treated with banana and bamboo residues had the highest nutrient levels, compared to the more sclerophyllous and recalcitrant arecanut residues (Table 1). All parameters except available P, ammonium N, and microbial biomass C also showed significant and positive correlation with earthworm populations (data not shown). In this study, the earthworm population in the soil was affected by incorporation of leaf residues and cowdung; in particular, banana and arecanut residue incorporation resulted in increasing their number. At the end of the experiment, the population of earthworm increased by ca. 60% in the banana residue treatments (Table 2). Weight loss and high mortality of earthworms observed in the bamboo-amended soil could be due to its physical texture and high lignin content (24.51%), implying substrate preference by earthworms.

The major influence of earthworm was to increase the amount of extractable N and decrease the amount of microbial biomass N particularly in the soil that was supplied with earthworm without any organic residues. This suggests that earthworm converts microbial-N to extractable N. Thus, crop residue management in the light of earthworm dynamics could help improve soil nutrient status whereby crop productivity could be sustained, especially on nutrient impoverished sites. Overall, application of earthworm along with different plant resides and cowdung increased nutrient supply as well as earthworm and microbial population depending on the quality of residues used and their relative proportion.

Acknowledgements

Financial support from the Indian Council of Agricultural Research, New Delhi is thankfully acknowledged. The authors also express their gratitude and thanks to the field assistant Shri M. Ingti.

References

Allen, S.E., Grimshaw, H.M., Parkinson, J.A., and Quarnby, C. 1974. *Chemical Analysis of Ecological Materials*. Blackwell Scientific, Oxford, 565p.

Anderson, J.M. and Ingram, J.S. 1989. *Tropical Soil Biology and Fertility: A Handbook of Methods.* Commonwealth Agricultural Bureau, Wallingford, 221p.

Brown, G.G. and Doube, B.M. 2004. Functional interactions between earthworms, microorganisms, organic matter, and plants. In: Edwards, C.A. (ed.), *Earthworm Ecology*, 2nd Edition, CRC Press, Boca Raton, pp 213–224.

Jenkinson, D.S. and Powlson, D.S. 1976. The effect of biocidal treatments on metabolism in soil-V.A. methods for measuring soil biomass. Soil Biol. Biochem., 8: 209–213.

Johnson, L.F. and Curl, E.L. 1972. *Methods for the Research on Ecology and Soil-borne Pathogens.* Burgess publishing & Co. Minneapolis, Minnesota, 247p.

Waksman, S.A. 1952. *Soil Microbiology.* John Wiley and Sons, New York, 356p.