



## Improving Soil Microbiological Properties in Typic Ustochrepts through Tree-based Cropping System

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Anthropogenic interference in natural ecosystem due to intensive agriculture has led to deterioration of soil fertility, productivity and quality. The soil microbiological properties are very important means of assessing soil health and productivity of agricultural ecosystems. The purpose of this study was to investigate the impact of tree-based cropping systems on soil microbiological activity in Shiwalik region of lower Himalayas. A study was conducted in a mixed watershed comprising of agroforestry, agrohorticulture and maize-wheat cropping systems. The soil samples were collected from three cropping systems and were analyzed for soil microbiological and basic physicochemical properties. The soil microbiological properties significantly improved in agroforestry and agrihorticultural systems compared to maize-wheat cropping systems. Soil respiration rate and polysaccharide carbon content were also higher in soils under tree-based cropping system (agroforestry and agrihorticulture) than in soils under maize-wheat system. The soil microbiological properties were positively correlated with soil organic carbon, available N and Olsen P. The higher soil microbiological activity under tree-based cropping system as compared with sole agriculture shows the propitious role of trees for the ecosystem functioning in north-west region of India.

**Key words:** Tree-based cropping system, microbiological properties, enzyme activity, respiration rate, sole agriculture, polysaccharide carbon

Rapid population growth in the lower Himalayan region of north-west India has led to the conversion of natural forest and grassland into cultivated lands. This historical incompatible land use change eventually has led to environmental degradation. The prevalence of intensive agriculture *i.e.* sole agricultural production has altered the below-ground ecosystem, plant cover and biomass often leading to loss of biodiversity and depletion of soil organic matter (Beheshti *et al.* 2012). On the other hand, tree-based cropping system, growing of multipurpose trees along with agricultural crops is a better practice for the maintenance of soil fertility as compared to sole agriculture. The cropping systems involving trees favour the soil organic carbon (SOC) sequestration because of their increased woody biomass, extensive roots and enormous amount of litter fall (Dhaliwal *et al.* 2017). Growing evidence suggests that tree species have significant impacts on soil physicochemical properties, substrate quality and on the soil microbiological properties (Ushio *et al.* 2010).

The microbiological properties, characteristics of the microbial community, are very sensitive to the environmental conditions and are efficient functional indicators of soil health. Among microbiological properties, enzymatic activity is important biological indicators of soil quality because of their quick response to management practices and the metabolic activity of soil microorganisms, which influence soil fertility, nutrient cycles and transformations (Nannipieri *et al.* 2002). Chief constituent of these reactions are soil enzymes, which also insinuate the change in soil quality with respect to environmental and anthropogenic factors. As enzymes response swiftly to environmental signals and therefore the use of soil enzyme activities as soil quality indicator will reveal biological status of the soil (Melero *et al.* 2008). Specifically, dehydrogenase activity in soils provides correlative information on the biological activity; phosphatase activity is thought to be directly related to the level of phosphorus (P) in the soil; urease activity generally correlates with soil organic carbon (SOC) content due to its existence as a complex with organic constituents. The measurement

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of soil enzymes activities in the ecosystem would help quantify and evaluate specific biological processes in soils. Soil respiration provides an overall potential of microbial activity and is considered as a sensitive bio-indicator of soil quality (Islam and Weil 2000).

The influence of tree-based cropping systems on soil microbiological properties in lower Himalayas is not known. For the maintenance of soil fertility and ecosystem functioning in sustainable manner, it is very important to study the effect of tree-based cropping system on key indicators of soil health *viz.*, SOC and biological activity. We hypothesized that different cropping systems can influence microbial and enzyme activity in soil, which in turn, will affect organic matter turnover in soil. The aim of this study was to evaluate the changes in soil microbiological properties under different tree-based cropping systems in comparison to sole agriculture (maize-wheat).

## Materials and Methods

### *Study site, climate and soil characteristics*

The study was conducted at 31°31'46.53" N latitude and 75°55'12" E longitude in Shiwalik region of lower Himalayas, at an altitude of 336 m above the mean sea level, with hot and semi-arid climate. The average annual rainfall in the region is 800-1000 mm, about 80% of which is received during the months of July to September, July being the wettest month. The mean maximum temperature varies from 19 °C in January to 41 °C in June, and the mean minimum temperature varies from 6 °C in January to 27 °C in June. The soils were sandy loam in texture; slightly alkaline and non-saline (Table 1). The study was conducted in a mixed watershed comprising of three different cropping systems *viz.*, agroforestry (40% of total area), agrihorticulture (20% of total area) and maize-wheat (40% of total area). These cultivated lands were either levelled or gently sloping (0.5-2.0%

slope steepness). Maize (*Zea mays*) was grown during summer season (June-October) and wheat (*Triticum aestivum*) during winter season (November-April). The agroforestry system comprised of poplar (*Populus deltoides*) trees intercropped with sorghum fodder (*Sorghum bicolor*) during summer, and wheat during winter season. The agrihorticulture system consisted of guava (*Psidium guajava*) trees intercropped with sorghum and wheat as described above. The maize, sorghum and wheat crops were cultivated as per recommended practices of Punjab Agricultural University, Ludhiana (Anonymous 2013a,b). The poplar trees were planted at a spacing of 6.5 m × 4.3 m and guava trees were planted at a spacing of 6 m × 5 m. The tree plantations were of six years in age at the time of sampling (2013).

### *Soil sampling*

The bulk soil samples were collected from the watershed at three sites for each cropping system (agroforestry, agrihorticulture and maize-wheat) and for two depths *i.e.* from surface (0-15 cm) and sub-surface (15-30 cm) soil layer. At each site, the samples were randomly collected from six places and mixed to prepare a composite sample. The soil samples were stored in refrigerator at 4 °C for future estimation of microbial properties, while those were ground to pass through 2-mm sieve after air drying for determination of soil physicochemical parameters. The samples were collected after clearing the land surface of the accumulated leaf litter under agroforestry and agrihorticulture. In all the cropping systems the soil sampling was carried out during the off-season.

### *Chemical analysis*

The soil pH was determined in 1:2 soil:water suspensions using an Elico-glass electrode pH meter (Jackson 1973). The electrical conductivity (EC) was determined by a conductivity bridge after equilibrating

**Table 1.** Physicochemical properties of soil under different cropping systems

| Properties                         | 0-15 cm     |              |                  | 15-30 cm    |              |                  |
|------------------------------------|-------------|--------------|------------------|-------------|--------------|------------------|
|                                    | Maize-wheat | Agroforestry | Agrihorticulture | Maize-wheat | Agroforestry | Agrihorticulture |
| Sand (%)                           | 80.8        | 78.1         | 80.0             | 76.6        | 74.9         | 77.7             |
| Silt (%)                           | 11.8        | 8.1          | 14.2             | 10.8        | 9.3          | 11.3             |
| Clay (%)                           | 7.4         | 13.8         | 5.8              | 12.6        | 15.8         | 11.0             |
| pH (1:2)                           | 8.46        | 8.42         | 8.37             | 8.48        | 8.44         | 8.40             |
| EC (dS m <sup>-1</sup> )           | 0.20        | 0.29         | 0.26             | 0.18        | 0.25         | 0.22             |
| SOC (%)                            | 0.48        | 0.54         | 0.53             | 0.36        | 0.27         | 0.24             |
| Available N (kg ha <sup>-1</sup> ) | 141         | 149          | 148              | 119         | 126          | 124              |
| Olsen P (kg ha <sup>-1</sup> )     | 34.8        | 23.0         | 32.1             | 28.6        | 18.9         | 24.8             |

for 30 min in the soil:water suspension used for pH determination. The SOC was determined by Walkley and Black rapid titration method (Walkley and Black 1934). The available P was determined by Olsen's extraction method (Olsen *et al.* 1954). The available N was estimated by alkaline permanganate method given by Subbiah and Asija (1965).

#### Microbiological analysis

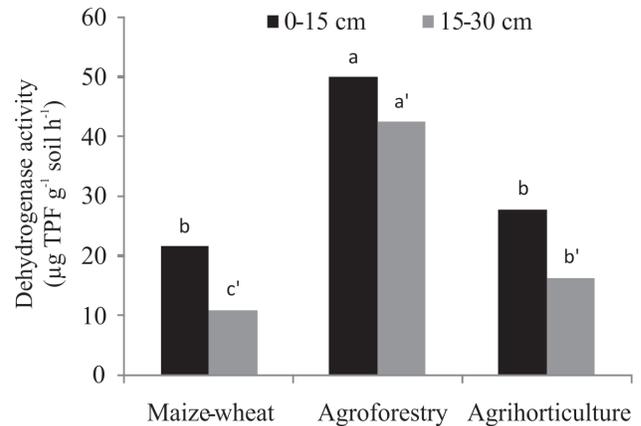
The dehydrogenase activity (DHA) was assayed as per the method described by Tabatabai (1982). The acid and alkaline phosphatase was assayed by adopting the standard procedure of Tabatabai and Bremnar (1969). The urease activity was assayed by method of Douglas and Bremner (1970). Soil respiration rate was measured by using alkaline trap method (Anderson 1982). The acid-extractable polysaccharides were analyzed with the technique modified from Whistler and Wolfrom (1962) by Lowe (1993).

The data so obtained for different soil enzymes, soil respiration rate and polysaccharide carbon content were compared using least square difference (LSD) at 5% level of significance calculated from analysis of variance (ANOVA) using software SAS 9.3 in a completely randomized design (CRD).

## Results and Discussion

#### Dehydrogenase activity

The DHA participates in electron transport system of oxygen metabolism so it reflects the extent of oxidative activity of soil microorganisms and is a good indicator of microbial activity (Nannipieri *et al.* 2002) in soils. The DHA was significantly ( $P < 0.05$ ) higher in soils under agroforestry ( $49.9 \mu\text{g TPF g}^{-1} \text{h}^{-1}$ ) than in soils under agrihorticulture ( $27.8 \mu\text{g TPF g}^{-1} \text{h}^{-1}$ ) and maize-wheat ( $21.6 \mu\text{g TPF g}^{-1} \text{h}^{-1}$ ) in surface soil layer (Fig. 1). The higher DHA activity under agroforestry system may be due to the high substrate availability in these soils. The higher leaf biomass of poplar trees (about  $3\text{-}4 \text{ t ha}^{-1} \text{ yr}^{-1}$ ) due to leaf litter fall facilitates greater carbon inputs to the soil. The different biochemical constituents of roots also influence DHA activity. The DHA decreases with the depth; the decrease was more in maize-wheat (49.3%) than in agrihorticulture (41.7%) and was lowest in agroforestry system (15.0%) in the sub-surface soil layer. The decline in activities with depth seems to be due to the decrease in oxidizable SOC fraction as many of the microbiological properties including the DHA have been reported to be higher in



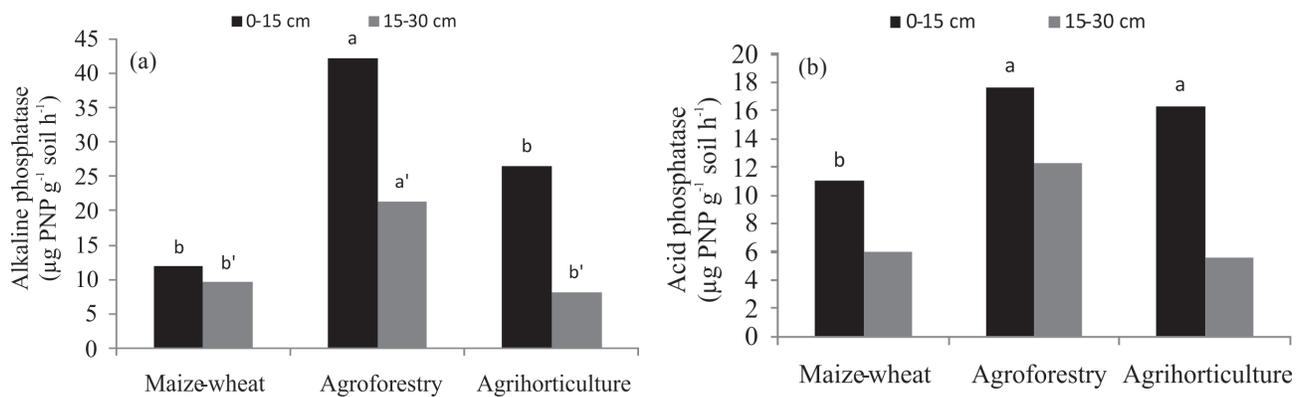
**Fig. 1.** Dehydrogenase activity in surface (0-15 cm) and sub-surface (15-30 cm) soil layers under different cropping systems. For a given depth the values indicated by the same letter are not significantly different

surface layer and considerably decrease with increase in soil depth (Melero *et al.* 2008).

#### Alkaline and acid phosphatase activity

Soil phosphatase play a major role in the mineralization processes of organic P substrates and their activity can be influenced by soil properties and cropping systems (Sarapatka 2003). The alkaline phosphatase activity in the surface (0-15 cm) soil layer was significantly higher in soils under agroforestry ( $42.2 \mu\text{g PNP g}^{-1} \text{ soil h}^{-1}$ ) and decreased by 37.2 and 71.8 per cent in soils under agrihorticulture and maize-wheat, respectively (Fig. 2a). The higher enzyme activity in agroforestry system could be due to higher organic carbon content in the soils, because of addition of enormous amount of leaf litter fall which may be easily decomposable and facilitate enzyme activity (Table 2). The present results for alkaline phosphatase were in consistence with the findings of Ushio *et al.* (2010) and Singh *et al.* (2014) who reported positive impact of litter quality on the alkaline phosphatase activity under forest soil as compared to agrihorticultural systems. The increase in activity of hydrolase enzymes might be due to higher levels of intracellular and/or extracellular enzymes, immobilized by recalcitrant humic moieties. In the sub-surface layer enzyme activity decreased and it was lowest ( $8.2 \mu\text{g PNP g}^{-1} \text{ soil h}^{-1}$ ) in agrihorticulture system which could be due to the reason that the reactions of horticultural litter may be acidic.

The acid phosphatase activity in surface soil layer was significantly higher in tree-based cropping system in comparison to sole agriculture (Fig. 2b).

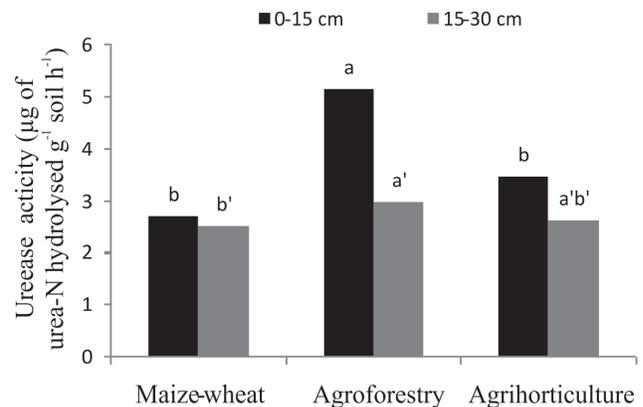


**Fig. 2.** Alkaline phosphatase (a) and acid phosphatase (b) in surface (0-15 cm) and sub-surface (15-30 cm) soil layers under different cropping systems. For a given depth the values indicated by the same letter are not significantly different.

The increased activity of acid phosphatase under tree-based cropping systems tends to increase with high organic matter content and due to high P stress (Li *et al.* 2002). As P content was highest ( $34.8 \text{ kg ha}^{-1}$ ) in soils under maize-wheat followed by agrihorticulture ( $32.2 \text{ kg ha}^{-1}$ ) and was lowest ( $23.0 \text{ kg ha}^{-1}$ ) in soils under agroforestry (Table 1). Trees have evolved many morphological and enzymatic adaptations to tolerate low phosphate availability as and when there is a signal indicating P deficiency in the soil, acid phosphatase secretion from plant roots is increased to enhance the solubilization and remobilization of phosphate, thus influencing the ability of the plant to cope with P-deficient conditions. Phosphatase activity decreased with soil depth and corresponded to the distribution of microorganisms in the soil profiles and organic carbon content. In the sub-surface soil layer, there was no significant difference in acid phosphatase activity under different cropping systems.

#### Urease activity

The urease activity was significantly higher in agroforestry ( $5.2 \mu\text{g}$  of urea-N hydrolysed  $\text{g}^{-1} \text{h}^{-1}$ ) than in agrihorticulture ( $3.5 \mu\text{g}$  of urea-N hydrolysed  $\text{g}^{-1} \text{h}^{-1}$ ) and lowest ( $2.7 \mu\text{g}$  of urea-N hydrolysed  $\text{g}^{-1} \text{h}^{-1}$ ) in soils under maize-wheat (Fig. 3). The variation in urease activity may be due to the organic carbon concentration. Agroforestry system improved the quality of soil organic matter because soil polysaccharide content increased in agroforestry system that relate to C turnover as well as urease activity. Moreover, higher available N in agroforestry system may be attributed to lesser losses of N through immobilization, volatilization, denitrification and leaching (Table 1) (Malhi *et al.* 2001), whereas in maize-wheat cropping system more disturbance of soil

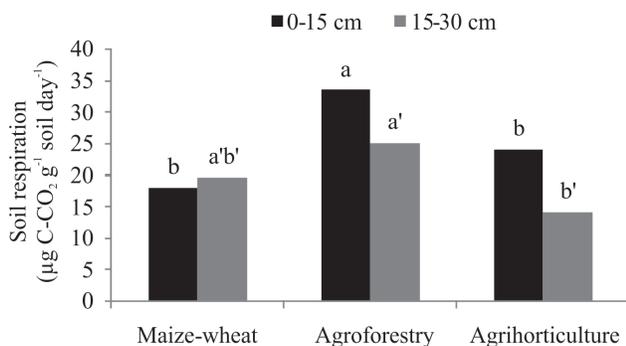


**Fig. 3.** Urease activity in surface (0-15 cm) and sub-surface (15-30 cm) soil layers under different cropping systems. For a given depth the values indicated by the same letter are not significantly different

might lead to more losses of N through different mechanisms. The results showed that urease activity was significantly affected by different soil management systems. It is reported that urease activity declined with increase in soil depth and maximum activity is generally restricted in the surface soil layer (Velmourougane 2013).

#### Soil respiration rate and polysaccharide carbon

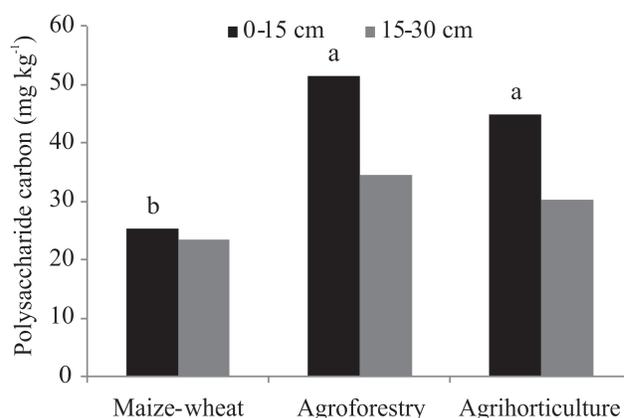
The soil respiration rate was 86.8 and 34.3 per cent higher under agroforestry and agrihorticulture as compared to maize-wheat system (Fig. 4). Soil respiration rate was higher under tree-based cropping system as compared to sole agriculture (Joergensen 2010) as it depends on the quantity and quality of detritus supplied to the soil, and the overall rate of root respiration. Soil respiration is the production of  $\text{CO}_2$  by the metabolizing organisms within the soil. It



**Fig. 4.** Soil respiration in surface (0-15 cm) and sub-surface (15-30 cm) soil layers under different cropping systems. For a given depth the values indicated by the same letter are not significantly different

estimates the overall biological activity in the litter-soil layer and is known to be affected by the availability of substrates, nutrients, moisture and temperature. In tree-based cropping systems, organic C is contributed by root decomposition and crop residue incorporation (Gale and Camberdella 2000) whereas in maize-wheat system due to tillage practice, loss of soil C and pulverization of root and crop residues led to dilution of organic C in soil. The variation in organic C content might be due to the distribution of organic matter within the profile, which is influenced by tillage. This pattern supports our study that the variation in organic C content under different cropping system influenced by the availability of carbon substrates for microorganisms, plant root densities and activities and microbial population.

There was a significant difference in the polysaccharide C content in surface soil layer under different cropping systems. It was higher in tree-based cropping system in comparison to sole agriculture (102.4% higher in agroforestry and 76.8% higher in agrihorticulture than in maize-wheat) (Fig. 5). Soil polysaccharide is an important source of energy for microorganisms; therefore agroforestry soil is



**Fig. 5.** Polysaccharide carbon in surface (0-15 cm) and sub-surface (15-30 cm) soil layers under different cropping systems. For a given depth the values indicated by the same letter are not significantly different

suggested to promote biological activity in the soil. This was supported by higher DHA and ratio of polysaccharide to SOC in agroforestry as compared with agrihorticulture and maize-wheat soils. Moreover, there was more litter fall under tree-based cropping system also increased the polysaccharide content as compared to sole agricultural crops. There was no significant difference in the polysaccharide carbon in the sub-surface layer but the amount of polysaccharide carbon was highest in agroforestry soils followed by agrihorticulture and lowest in soils under maize-wheat.

#### Correlation study

The microbiological properties assayed in the study were significantly and positively correlated with SOC and P under different cropping system (Table 2). The values of correlation coefficient ( $r$ ) ranged from 0.979 to 0.625. The polysaccharide C and acid phosphatase, have higher  $r$  value with SOC. Polysaccharide C has highest correlation coefficient ( $r = 0.979$ ) with soil respiration.

**Table 2.** Correlation coefficients of microbial properties under different cropping systems in surface soil layer

| Parameter        | SOC     | DHA     | Polysach-C | Soil respiration | Olsen-P | Alk-Phos |
|------------------|---------|---------|------------|------------------|---------|----------|
| DHA              | 0.715*  |         |            |                  |         |          |
| Polysach-C       | 0.825** | 0.811** |            |                  |         |          |
| Soil respiration | 0.762** | 0.811** | 0.979**    |                  |         |          |
| Olsen-P          | 0.625   | 0.974** | 0.775*     | 0.788*           |         |          |
| Alk-phos         | 0.674** | 0.729** | 0.790*     | 0.880*           | 0.743** |          |
| Acid-phos        | 0.826** | 0.769** | 0.947**    | 0.959**          | 0.737*  | 0.851**  |

\* Correlation is significant at the  $P < 0.05$  level; \*\* Correlation is significant at the  $P < 0.01$  level (Polysach-C - Polysaccharide carbon, SR- Soil respiration, Alk-P- Alkaline phosphatase)

## Conclusions

The results indicated that different cropping systems have intense impact on soil microbiological properties. Soils under agroforestry based cropping system showed invariably highest value of all the microbiological properties as compared to sole agriculture. Further, among the tree-based cropping, agroforestry system was more efficient than the agrihorticulture system. The tree-based cropping system could be regarded as a promising management practices for better soil health especially in Shiwalik region where soils were less fertile due to soil erosion.

## References

- Anonymous (2013a) Package of practices for crops of Punjab-Kharif, Punjab Agricultural University Ludhiana, pp. 24-29.
- Anonymous (2013b) Package of practices for crops of Punjab-Rabi. Punjab Agricultural University Ludhiana, pp. 1-18.
- Anderson, J.P.E. (1982) *Soil respiration: Chemical and Microbiological Properties*. In *Methods of Soil Analysis* (R.H. Miller and D.R. Keeney, Eds.) Madison, Wisconsin, USA, pp. 831-871.
- Beheshti, A., Raiesi, F. and Golchin, A. (2012) Soil properties, C fractions and their dynamics in land use conversion from native forests to croplands in northern Iran. *Agriculture, Ecosystems and Environment* **148**, 121-133.
- Dhaliwal, J., Kukal, S.S. and Sharma, S. (2017) Soil organic carbon stock in relation to aggregate size and stability under tree-based cropping systems in Typic Ustochrepts. *Agroforestry Systems* doi: 10.1007/s10457-017-0103-8.
- Douglas, L.A. and Bremner, J.M. (1970) Extraction and colorimetric determination of urea in soils. *Soil Science Society of America Journal* **34**, 859-862.
- Gale, W.J. and Cambardella, C.A. (2002) Carbon dynamics of surface residue- and root-derived organic matter under simulated no-till. *Soil Science Society of America Journal* **64**, 190-195.
- Islam, K.R. and Weil, R.R. (2000) Land use effects on soil quality in a tropical forest ecosystem of Bangladesh. *Agriculture, Ecosystems and Environment* **79**, 9-16.
- Jackson, M.L. (1973) *Soil Chemical Analysis*. Prentice Hall of India (Pvt.) Ltd., New Delhi.
- Joergensen, R.G. (2010) Organic matter and micro-organisms in tropical soils. *Soil Biology* **21**, 17-44.
- Li, D., Zhu, H., Liu, K., Liu, X., Leggewie, G., Udvardis, M. and Wang, D. (2002) Purple acid phosphatases of *Arabidopsis thaliana* comparative analysis and differential regulation by phosphate deprivation. *Journal of Biological Chemistry* **277**, 27772-27781.
- Lowe, L.E. (1993) Total and labile polysaccharide analysis of soils. In *Soil Sampling and Methods of Analysis* (M.R. Carter, Ed.), CSSS Boca Raton, Florida, pp. 373-376.
- Malhi, S.S., Grant C.A., Johnston, A.M. and Gill, K.S. (2001) Nitrogen fertilization management for no-till cereal production in the Canadian Great Plains: a review. *Soil and Tillage Research* **60**, 101-122.
- Melero, S., Vanderlinden, K., Ruiz, J.C. and Madejon, E. (2008) Long-term effect on soil biochemical status of a Vertisol under conservation tillage system in semi-arid Mediterranean conditions. *European Journal of Soil Biology* **44**, 437-442.
- Nannipieri, P., Kandeler, E. and Ruggiero, P. (2002) Enzyme activities and microbiological and biochemical processes in soil. In *Enzymes in Environment* (R.G. Burns and R.P. Dick, Eds.), Marcel Dekker, New York, pp.1-33.
- Olsen, S., Cole, C., Watanabe, F. and Dean, L. (1954) Estimation of available phosphorus in soils by extraction with sodium bicarbonate. *United States Department of Agriculture Circular No. 939*.
- Piper, C.S. (1950) *Soil and Plant Analysis*. International Science Publishers, New York.
- Sarapatka, B. (2003) Phosphatase activities (ACP, ALP) in agroecosystem soils. *Dissertation*, Swedish University of Agricultural Sciences.
- Singh, R.D., Kumar, A., Patra, A.K., Sahu, S.K., Khan, M.A. and Bhole, B.S. (2014) Impact of different land use management on soil enzyme activities and bacterial genetic finger prints of north-western Himalayas. *Current World Environment* **9**, 728-740.
- Subbiah, B.V. and Asija, G.L. (1956) A rapid procedure for the determination of available nitrogen in soils. *Current Science* **25**, 259-260.
- Tabatabai, M.A. (1982) Soil enzyme. In *Methods of Soil Analysis*. Part 2 (A.L. Page Eds.) ASA Madison, WI, pp. 903-948.
- Tabatabai, M.A. and Bremner, J.M. (1969) Use of p-nitrophenyl phosphate for assay of soil phosphatase activity. *Soil Biology and Biochemistry* **1**, 301-307.
- Ushio, B.M., Kitayama, K. and Teri, C.B. (2010) Tree species effects on soil enzyme activities through

- effects on soil physicochemical and microbial properties in a tropical montane forest on Mt. Kinabalu, Borneo. *Pedobiologia* **53**, 227-233.
- Velmourougane, K., Venugopalan, M.V., Bhattacharyya, T., Sarkar, D., Pal, D.K., Sahu, A., Chandran, P., Ray, S.K., Mandal, C., Nair, K.M., Prasad, J., Singh, R.S. and Tiwary, P. (2013) Urease activity in various agro-ecological sub-regions of black soil regions of India. *Proceedings of the National Academy of Sciences, India Section B: Biological Sciences* **83**, 513-524.
- Walkley, A. and Black, I.A. (1934) An examination of the Degtjareff method for determining soil organic matter, and a proposed modification of the chromic acid titration method. *Soil Science* **37**, 29-38.
- Whistler, B.L. and Wolfrom, M.L. (1962) *Methods in Carbohydrate Chemistry* I. London, Academic Press.