Short Communication
Annals of Plant and Soil Research 24(4):676-678 (2022)
https://doi.org/10.47815/apsr.2022.10228

Enzyme activities and microbial population status of soil as influenced by humic acid under doses of nitrogen in Spinach rhizosphere

AKHILA NAND DUBEY[1], PRIYANKAR RAHA [2] AND SHIVAM SINGH[3*]

Krishi Vigyan Kendra, Khagaria, Bihar Agricultural University, Sabour

Received: May, 2022; Revised accepted, August, 2022

In recent years, soil amendment with humus to increase fertility of soil with low organic matter content resulted to increase microbial density, enzyme activities as well as to stimulate the growth of plants. Dehydrogenase activity is an oxidoreductase enzyme that has been used as a measure of microbial activity because of an intracellular enzyme. Other phosphate is hydrolase enzyme, which is associated with nutrient mineralization. It is a sensitive indicator of changes in characteristics with soil organic matter content and quality (Veum et al, 2014). The experiment presentedin this paper was designed to study the effect of soil amendment with humic acid on enzyme activities and microbial activities, because they are indicators of microbial activity and nutrients mineralization in soil.

The lignite coal was collected from Matasukh Coal Mines, Nagaur, Rajasthan and HAwas extracted by 0.5 N KOH method from lignite coal. A pot experiment was conducted in net house at the Department of Soil Science & Agricultural Chemistry, Banaras Hindu University, Varanasi (Uttar Pradesh), (25°16´10´´N and 82°59´9´´E), during 2017-18, with factorial complete randomized design. The surface soil (0-15 cm depth) was collected from Gangetic plain of alluvial soil (Inceptisol) before monsoon season. Initially, the soil was clay loam in texture. The WHC, pH and EC was 45.5%, 7.9, 0.30 dSm^{-1}, respectively. Organic Carbon (4.97mgkg^{-1}), available nitrogen (220 kg ha^{-1}), available phosphorous (25 kg ha^{-1}) and exchangeable potassium (171 kg ha^{-1}) were analyzed as standard procedure. Each earthen pot was filled with 10 kg soil with six levels of HA [*i.e*. 0.0 (H_0), 0.5 (H_1) 0.75 (H_2) 1.5 (H_3) 2.5 (H_4) and 5.0 (H_5) mg HA kg^{-1} soil] and four doses of N [0.0 (N_0), 18.50 (N_1), 27.80 (N_2) and 37.0 (N_3) mg kg^{-1} soil]. According to treatments, the recommended dose of N (80 kg ha^{-1}) along withP_2O_5: K_2O (50: 50 kg ha^{-1}) were applied for spinach (variety “All Green”) in each pot. Dehydrogenase and alkalinephosphatase enzyme activity were measured in soil as per the Casida et *al.*, (1964) and Tabatabai and Bremner (1969), respectively. The microbial population in soil was enumerated by serial dilution platetechnique (Veum et al, 2014) and using various medium *i.e*.nutrient agar, Kenknight’s agarmedium (Allen, 1957) and Martin rose bengal (Martin *et al,* 1950) for bacteria, actinomycetes and fungi, respectively.

Enzyme activity (dehydrogenase and alkaline phosphatase) in soil increased significantly with the application levels of humic acids and nitrogen (Table 1). Dehydrogenase and alkaline phosphatase enzyme activities in soil were recorded significantly highest in H_5 (101.58mg of TPF formed g^{-1} soil 24 hr^{-1}and70.42m mol P-nitro phenol released g^{-1} fresh soil) as compared to other treatments. The application of higher dose of nitrogen (37.00 mg kg^{-1} soil) recorded significantly highest dehydrogenase and alkaline phosphatase enzyme (81.14mg of TPF formed g^{-1} soil 24 hr^{-1}and54.00m mol P-nitro phenol released g^{-1} fresh soil) in all the treated pots. The enhancement of dehydrogenase and alkaline phosphatase activities due to application of H_5 were 22.6% and 46.1%, respectively in soil over the control (Figure 1.a). Soil enzymes are the biological catalysts for certain reactions, and these reactions depend on various factorsviz - kinetics and kinematics of nutrient release-retention pattern, variety of germplasm etc. Incorporation of soil organic matter as HA might stimulate the enzymatic activity of soil (Rohman et al, 2019).The soil microbial population increased with the application of humic acids after harvesting of spinach (Table 1).

Corresponding author email (*): shivambuat@outlook.com
2 - Department of Soil Science & Agricultural Chemistry, BHU, Varanasi
3 – Department of Soil Science & Agricultural Chemistry, SVPUAT, Meerut

Table 1: Impact HA and nitrogenous fertilizer on enzymatic activities and microbial population in soil under spinach crop

Treatement	Dehydrogenase (mg of TPF formed g^{-1} soil 24 hr^{-1})	Alkaline phosphates (mmol P-nitro phenol released g^{-1} fresh soil)	Bacteria (x 10^6/g soil)	Fungi (x 10^4/g soil)	Actinomycetes (x 10^5/g soil)
H_0:Control	82.75±1.44^e	48.17±1.4^f	19.27±1.003^f	10.25±0.725^e	15.52±0.663^e
H_1: 0.50 mg HAkg^{-1}soil	83.83±1.71^d	52.58±1.04^e	22.15±1.491^e	12.24±0.678^d	16.92±0.620^d
H_2: 0.75 mg HAkg^{-1}soil	86.33±2.07^c	56.25±1.49^d	23.94±1.362^d	12.46±0.491^d	17.49±0.661^{cd}
H_3: 1.50 mg HAkg^{-1} soil	90.00±3.39^b	60.58±1.66^c	25.07±1.354^c	13.04±0.471^c	18.07±0.822^c
H_4: 2.50 mg HAkg^{-1} soil	91.50±2.92^b	64.33±2.01^b	26.44±1.478^b	14.53±0.853^b	19.12±0.924^b
H_5: 5.00 mg HAkg^{-1} soil	101.58±3.54^a	70.42±3.16^a	29.91±2.138^a	15.31±0.876^a	20.50±1.170^a
N_0:Control	72.43±3.10^d	46.81±2.63^d	20.90±1.074^d	11.26±0.698^d	15.98±0.384^d
N_1 :18.50 mg kg^{-1}soil	74.29±2.57^c	49.19±3.19^c	23.73±1.585^c	12.57±0.689^c	17.73±0.688^c
N_2: 27.80 mg kg^{-1}soil	78.43±3.09^b	51.33±3.56^b	25.39±1.589^b	13.68±0.726^b	18.36±1.003^b
N_3: 37.0 mg kg^{-1}soil	81.14±3.53^a	54.00±3.87^a	27.82±1.752^a	14.37±0.853^a	19.68±0.791^a

H, N represents humic acid, nitrogen level.

The microbial population (bacteria, fungi and actinomycetes) was found maximum in H_5 (29.91, 15.31, 20.50, respectively) followed by H_4, H_3, H_2 and H_1 as compared to control in harvested soil. The application of nitrogen doses followed the similar trend of HA application. It recorded the highest in N_3 (27.82×10^6/g, 14.37×10^4/g, 19.68×10^5/g soil respectively) $>N_2>N_1>N_0$. The per cent enhancements of bacteria, fungi and actinomycetes due to application of H_5 were 54.5, 50.0 and 31.5, respectively in soil over the control (Figure 1.b). Application of humic acids to soil increased the population of beneficial microorganisms in soil by serving as a source of nutrients, which is in agreement with results (Dong et al, 2009; Rohman et al, 2019).The increasing dynamics for the microbial count and enzymatic activities were obtained for HA and nitrogen application which might be due to fact that HA enriches nourishment for soil micro-organism (Rohman et al, 2019). Increase in the nitrogen lowers C:N ratio which facilitates the decomposition rate much faster (Spohn, 2015).

The response of rhizospheric microbial population and enzymes activities to humic acid and N-fertilizer were acquaintly correlated with each other. Sustained level of crop nutrition requires, achieving the potential yield and maximum nutrient use efficiency. Hence through integrated use of fertilizers and humic acid, the microbial population and soil enzymes in rhizosphere soil could be build up for the efficient utilization of nutrients.

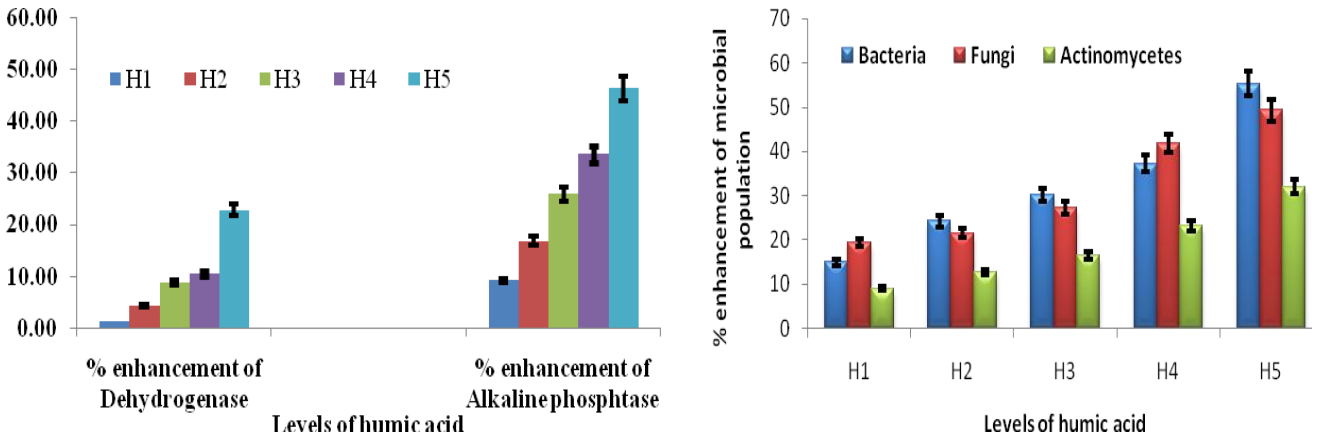

Figure 1: Impact of humic acid originated from lignite and nitrogenous fertilizer on % enhancement of enzymatic activities (a) and microbial population (b) in soil

REFERENCES

Allen O.N. (1957). Experiments in Soil Bacteriology. Burgess Publishing CO., Mineeapolis, 15, Minnesota.

Casida Jr., L.E., Klein, D.A., Santoro, T., 1964. Soil dehydrogenase activity. Soil Science. **93**, 371–376

Dong L., Córdova-Kreylos A. L., Yang J., Yuan H., Scow K. M. (2009). Humic acids buffer the effects of urea on soil ammonia oxidizers and potential nitrification. *Soil Biology and Biochemistry,* **41**(8), 1612-1621.

Martin J. P. (1950). Use of acid, rose bengal, and streptomycin in the plate method for estimating soil fungi. *Soil Science*, **69**(3): 215-232.

Rohman, A. F., Wachjar, B. A., Santosa, B. E., Abdoellah, C. S. (2019) HA and biofertilizer applications enhanced pod and cocoa bean production during the dry season at Kaliwining Plantation, Jember, East Java, *Indonesia. Journal of Tropical Crop Science* **6**(3), 153-163.

Spohn, M. (2015) Microbial respiration per unit microbial biomass depends on litter layer carbon-to-nitrogen ratio. Biogeosciences, **12**(3), 817-823.

Veum, K. S., Goyne, K. W., Kremer, R. J., Miles, R. J., & Sudduth, K. A. (2014). Biological indicators of soil quality and soil organic matter characteristics in an agricultural management continuum. *Biogeochemistry* **117**(1): 81-99.