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Effect of humic substances on nutrient uptake and yield of soybean

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Abstract
A field experiment was conducted during *Kharif*, 2017 at Agronomy field unit, College of Agriculture, Shivamogga to study the effect of humic substances on nutrient uptake and yield of soybean **[***Glycine max* (L.) Merrill]. The experiment consisted of 10 treatments replicated thrice was laid out in Complete Randomized Block Design. Soybean variety used in this experiment was JS-335. Application of vermicompost on FYM 'N' equivalent basis was found superior by recording significantly higher grain yield (1957 kg ha^{-1})and nutrients uptake (N, P and K) by grain, haulm, total plant uptake and less soil available nutrient status after harvest of the crop. Among humic substances treatments, soil application of humic substances @ 5 kg ha^{-1} at sowing + foliar application of humic substances extracted from vermicompost (0.2%) at 40 DAS resulted in significantly higher grain yield (1741 kg ha^{-1}) and nutrient uptake of N, P, K by grain, haulm, total uptake and less available nutrient status after harvest of the crop when compared to control.

Keywords: Soybean, humic acid substances, vermicompost, nutrient uptake, grain yield

Introduction

Soybean [*Glycine max* (L.) Merrill] is an important grain legume crop contains about 40 to 44 per cent protein and 20 per cent oil. Agronomically, it is classified under oilseed crops. Due to its high nutritional quality, higher productivity and its industrial importance there is lot of scope for its cultivation in India. Being a leguminous crop, soybean is also capable of with stand moisture stress and helps in improving the soil fertility and productivity. In India soybean is growing in an area of 11.67 m ha with production and productivity of 8.5 m t and 737 kg ha^{-1}, respectively (Anon., 2014) [1]. However, the productivity and quality of soybean was very low compared to advanced countries due to poor management practices in general and crop nutrition in particular. Ignorance attitude towards the importance of organic waste cycling and continuous application of soluble acidic based N, P and K fertilizers with an assumption that they could stimulate plant growth without organics or humic substances to the soil has caused many serious social and ecological problems. Indiscriminative application of chemicals and fertilizers has lead to soil, air, food and water pollution is one of the most important environmental and social concerns throughout the world especially in developing countries. Soil organic matter has beneficial effects not only on soil quality, but also has positive effects on crop productivity and quality of the produce. In addition, organic matters could reduce the application of industrial fertilizers in long run. It is the need of the hour to reconsider the approaches for fertilization techniques by giving priority to organic manure/amendments. However, Use of bulky organic manures has been considered as a burden by the farmers as it requires large number of laborer for transportation and application. So it is necessary to go for organic end products like humic substances for better soil condition, higher input use efficiency and enhanced productivity of crops.

Extraction of humic substances from bulky organic manures and their use may help to solve many problems associated with use of bulky organic manures. Humic substances like humus, humate, humin, humic acid and fulvic acid, will play a vital role in soil fertility and plant nutrition. They help to break soil compactness, assists in transferring micronutrients from the soil to the plant, enhances water retention, increases seed germination rates and stimulates development of microbial populations in soil. They also indirectly involved in improvement of soil properties such as aggregation, aeration, permeability, water holding capacity, serves as an effective adsorption and retention complex for inorganic plant nutrients and there by enhance the micronutrients uptake transport and availability (Tan, 2003) [17] and it exhibits auxin-like effects. As a result of these, application of humic substances stimulates plant growth and consequently yield and quality by acting on various mechanisms such as cell respiration, membrane permeability, photosynthesis, protein synthesis, water and nutrient uptake, enzyme activities (Nardi *et al*., 2002 and Chen *et al*., 2001) [11, 4].

In this context, an attempt has been made to extract the humic substances from vermicompost available at the site and to see its effects applied either alone as foliar/ soil or through both soil and foliar applications along with commercial available humic substances for comparison.

Material and Methods

A field experiment was carried out during *Kharif* 2017 at College of Agriculture, University of Agricultural and Horticultural Sciences, Navile, Shivamogga falls under Southern Transitional Zone of Karnataka to study the effect of humic substances on nutrient uptake, yield of soybean and soil available nutrient status after harvest of the crop. The location is situated between 14°0'N to 14º1'N latitude and 75°40' E to 75º42' E longitude and at an altitude of 650 meter above mean sea level. The soil of the experimental site is red sandy loam in texture with acidic pH (5.93), low in organic carbon (0.42 %) and available N (232 kg ha^{-1}), medium in available potassium (232.65 kg ha^{-1}), whereas, high in available phosphorous (58.45 kg ha^{-1}). The experiment consisted of 10 treatments *viz*., Control means no humic substance application (T_1), soil application of humic substances (humic and fulvic acid) @ 2.5 kg ha^{-1} (T_2) and 5 kg ha^{-1} (T_3), foliar application of humic substances extracted from vermicompost (0.2%) at 40 DAS (T_4), Foliar application of commercial humic substances (0.2%) at 40 DAS(T_5), T2+T4(T6), T2+T5(T7), T3+T4(T8), T3+T5(T9) and vermicompost on FYM 'N' equivalent basis *i.e*., 2.76 t ha^{-1} (T10) replicated thrice was laid out in Complete Randomized Block Design. A common dose of nitrogen, phosphorous and potassium @ 25:60:25 kg ha^{-1} was applied entirely as basal in the form of urea, DAP and muriate of potash, respectively. Two weeks prior to sowing, recommended quantity of FYM was applied uniformly to all the treatments except for T_{10} where in vermicompost on FYM 'N' equivalent basis in place of FYM was used. All cultural practices were carried out as per package of practices except treatment imposition. The Soybean variety JS-335 was used as test crop. Seeds were sown on 2nd August 2017 after treating with suitable rhizobium culture and harvested at physiological maturity. Grain and haulm samples collected from each plot at the time of harvest were dried at 60 ^{0}C in hot air oven were grounded separately in a Willey Mill using a grinder fitted with stainless steel blades to pass through 40 mesh sieve for further analysis of nutrient content. The grounded material was collected in butter paper bags and later the samples were analyzed for N, P and K content. Nitrogen content in plant samples (haulm and grain) was determined by modified Kjeldhal's method as described by Jackson (1973) [8]. For estimation of P and K content a powdered plant sample of 0.5 g was pre-digested with five ml of concentrated HNO_3 and again digested with a di-acid mixture (HNO_3: $HClO_4$ in 10:4 ratio). Volume of the digest was made up to 100 ml with distilled water and preserved for P and K analysis (Jackson, 1973) [8]. Later the phosphorus content in plant sample (haulm as well as grain) was determined by taking a known volume of the digested samples by adopting the Vanadomolybdo phosphoric yellow color method as described by Jackson (1973) [8]. The total potassium content of the di-acid digested plant and grain samples were estimated by atomizing the digested and diluted sample to a calibrated flame photometer under suitable measuring conditions as described by Jackson (1973) [8]. Nitrogen, phosphorus and potassium uptake was calculated for haulm, grain and total plant for each treatment separately using the formula given below and expressed in kg ha^{-1}.

$$\text{Nutrient uptake} = \frac{\text{Nutrient concentration (\%)} \times \text{Weight of dry matter (kg ha}^{-1}\text{)}}{100}$$

Statistical analysis of data was done as per the methodology suggested by Gomez and Gomez (1984) [7].

Results and Discussion

Grain yield of soybean

The grain yield of soybean varied significantly among humic acid treatments and their method of application (Table 1). The highest grain yield of soybean (1957 kg ha^{-1}) was realized with application of vermicompost on FYM 'N' equivalent basis (2.76 t ha^{-1}) which was significantly superior over rest of the treatments. There was an improvement in yield to an extent of 45.80 per cent due to vermicompost application on 'N' equivalent basis. Higher grain yield with application of vermicompost was due to better release of plant nutrients and their uptake (Table 2). Similar results were obtained by Edwards (1995) [5] and (Tomati *et al*. 1990) [19] in radish and lettuce crops.

Among humic acid treatments, the highest soybean grain yield of 1741 kg ha^{-1}was obtained with soil application of humic substances @ 5 kg ha^{-1} at sowing + foliar spray of humic substances extracted from vermicompost (0.2%) at 40 DAS closely followed by 1676 kg ha^{-1} soil application of humic substances @ 5 kg ha^{-1} + foliar application of commercial humic substances (0.2 %) at 40 DAS) which were on par with each other. The enhancement of yield in these treatments was to an extent of 29.76 and 24.89 per cent, respectively, over control (1342 kg ha^{-1}). Similarly, Nanda Kumar (2004) [10] reported 50.41 and 53.84 per cent yield improvement in rice, respectively, in clay and sandy loamy soils due to application of humic acid @ 20 kg ha^{-1}along with 100 per cent NPK. However, the control (POP) and the treatment receiving humic acid substances as foliar (T_4 and T_5) at 40 DAS have recorded significantly lower grain yield compared to those treatments which received either soil or foliar application of humic substances. The order of merit for methods of humic acid applications with respect to grain yield is soil + foliar>Soil >foliar. The variation in grain yield among treatments was mainly attributed to extent of nutrients taken up by the crop. Nutrient uptake by the crop is directly related to the yield (Table 2).

Similarly, the highest haulm yield of soybean (3134 kg ha^{-1}) was realized with application of vermicompost on FYM 'N' equivalent basis which was significantly superior over rest of the treatments (Table 1).

Among humic acid treatments, the highest haulm yield (2862 kg ha^{-1}) was obtained with soil application of humic substances @ 5 kg ha^{-1} at sowing + foliar spray of humic substances extracted from vermicompost (0.2%) at 40 DAS closely followed by soil application of humic substances @ of 5 kg ha^{-1} at sowing + foliar application of commercial humic substances (0.2%) at 40 DAS which are on par with each other. However, the control (POP) and the treatment receiving humic substances as foliar applications (0.2%) at 40 DAS have recorded significantly lower haulm yield compared to those treatments which received through soil and foliar application of humic substances. Higher haulm yield was due to enhancement of growth parameters attributed from higher activity of growth promoting hormones and also it might be due to higher dry matter accumulation in vegetative parts. Similar findings of increased haulm and grain yield in groundnut with humic acid application were reported by Thenmozhi *et al.* (2004) [18] and Talavia *et al.* (2007) [16] with combined application RDF + humic acid @ 20 kg ha^{-1}.

Nutrient uptake by soybean crop

Nitrogen uptake

Significantly higher nitrogen uptake in grain (135.6 kg ha^{-1}) and total plant uptake (208.4 kg ha^{-1}) was noticed with application of vermicompost on FYM 'N' equivalent basis at harvest over rest of the treatments, while significantly higher nitrogen uptake by haulm was noticed with soil application of humic acid substances @ 5 kg ha^{-1} at sowing + foliar application of humic substances extracted from vermicompost (0.2.%) at 40 DAS (71.0 kg ha^{-1}) which were significantly superior over rest of the treatments. However, these two treatments are statistically on par with regard to nitrogen uptake by grain, haulm and total plant. Among humic substances treatments soil application of humic acid substances @ 5 kg ha^{-1} at sowing + foliar application of humic substances extracted from vermicompost (0.2%) at 40 DAS excelled over others by recording significantly higher total nitrogen uptake (193.7 kg ha^{-1}). The control treatment (no application of humic substances) registered the least nitrogen uptake (85.9, 49.2 and 135.1 kg ha^{-1}, respectively by grain, haulm and total plant).

Among humic substances treatments, soil application of humic substances @ 5 kg ha^{-1} at sowing + foliar application humic substances extracted from vermicompost (0. 2%) at 40 DAS resulted in significantly higher grain (122.6 kg ha^{-1}) and total plant uptake (193.7 kg ha^{-1}) of nitrogen over other humic acid treatments, except T_9 (117.3, 182.7 kg ha^{-1}, respectively) which are on par. While, higher haulm uptake (71.0 kg ha^{-1}) of nitrogen was noticed with soil application of humic substances @ 5 kg ha^{-1} at sowing + foliar application of commercial humic acid (0.2%) at 40 DAS.

Higher nitrogen uptake was attributed to higher total dry matter (Table 1) and nutrient content since the uptake is the product of nutrient content and dry matter. The higher nitrogen uptake by soybean due to application of humic substances might be due to increased lateral root emergence and production of smaller but more ramified secondary roots coupled with improved cell permeability and better availability of nutrients in the soil solution (Sumathi and Rao, 2007; Bhandari *et al.*, 2000) [15, 2]. Enhanced microbial activity due to humic substances particularly of ammonifiers and nitrifiers will consistently supply nitrogen resulting in improved dry matter accumulation and nutrient content due to humic substances. The results are in line with the findings of Eyheraguibel *et al*. (2008) [6].

Phosphorus Uptake

The phosphorus uptake of soybean as influenced by humic substances indicated that vermicompost application on FYM 'N' equivalent basis statistically out crossed rest of the treatments by recording highest grain (12.9 kg ha^{-1}), haulm (9.4 kg ha^{-1}) and total plant uptake (22.3 kg ha^{-1}) of phosphorus.

Among humic substance treatments, T_5 recorded higher uptake of phosphorus (11.3, 8.3 and 19.6 kg ha^{-1} by grain, haulm and total plant, respectively) which are on par with T_9 and T_6 and with respect to grain and total uptake with T_9 for haulm and significantly superior over others. The least uptake was found with control (7.4, 4.4 and 11.8 kg ha^{-1} by grain, haulm and total plant, respectively) Higher phosphorus uptake is attributed to higher total dry matter plant^{-1} (Table 1) and nutrient content in plant parts including grain. In those treatments where crop received humic substances both through soil and foliar were significantly superior over treatments which received either through soil / foliar alone.

Potassium Uptake

The maximum potassium uptake was realized with vermicompost applied on FYM 'N' equivalent basis grain (18.8 kg ha^{-1}), haulm (92.8 kg ha^{-1}) and total (111.6 kg ha^{-1}) which was statistically excelled over remaining treatments. Among humic acid treatments soil application of humic substances @ 5 kg ha^{-1} + foliar application humic substances extracted from vermicompost (0.2%) at 40 DAS recorded significantly higher grain uptake of potassium (16.4 kg ha^{-1}) haulm (83.9 kg ha^{-1}) and total uptake (100.3 kg ha^{-1}) except T_9 and T_6 which are statistically on par.

The treatments involving both soil and foliar methods of humic acid were found statistically superior over sole application of either soil or foliar applications irrespective of levels. The least potassium uptake was noticed with control plot (Table 2).

Humic substances known to play a definite role in liberating fixed K because of their chelating power apart from the priming effect of solubilizing native i.e. fixed and non-exchangeable form of K. The enhanced microbial activity due to humic acid application would also pave way for increased availability of K by reducing its fixation in the soil and dissolution of fixed K. (Schnitzer and Khan, 1972) [12]. Further, better root metabolism and enzyme activity due to soil + foliar application of humic substances might have caused for higher nutrient uptake (Table 2). The results are in conformity with the findings of Cacco *et al.* (2000) [3]. Improved nutrient availability (Virgine and Singaram, 2005) [20] increased nutrient content in plants (Sharif *et al*., 2006) [14] high micronutrients (Kadam *et al*., 2010) [9], lesser leaching of nutrients (Selim *et al.*, 2010) [13] are some of the causes for higher nutrient uptake by the crop due to humic acid application.

Soil available nutrient status

The soil analysis for available nutrients status after the harvest of the soybean crop as influenced by humic substances indicate that the maximum amount of available nutrients was noticed in control plot (223.63, 99.16 and 277.4 kg ha^{-1} of nitrogen, phosphorus and potassium, respectively) closely followed by foliar application of commercial humic substances (0.2 %) at 40 DAS for nitrogen (215.25 kg ha^{-1}) and potassium (275 kg ha^{-1}) and soil application of humic acid substances @ 2.5 kg ha^{-1} at sowing + foliar application of commercial humic acid (0.2 %) at 40 DAS for phosphorus (98.04 kg ha^{-1}) which are statistically on par.

The least available nitrogen, phosphorus and potassium in soil after the crop harvest were observed in the treatment where vermicompost was applied on FYM 'N' equivalent basis (132, 88.62 and 237 kg ha^{-1}). All the humic acid treatments registered significantly lower available soil nitrogen after harvest of the crop over control. The treatments which received humic substances through foliar (T_4 and T_5) > soil (T_2 and T_3) > soil +foliar (T_6, T_7, T_8, and T_9) are in the order of merit with respect to available nutrient status. Lower available nutrient status in soil after the harvest of the crop in plots which received vermicompost (T_1) and humic substances both through soil + foliar was mainly due to the higher nutrient uptake (Table 2) and higher total dry matter per plant (Table 1) and vice versa in control plot. The construe of the data indicate that the highest total dry matter per plant was realized with application of vermicompost on FYM 'N' equivalent basis throughout the crop period 23.85 g at harvest). This was closely followed by soil application of humic substances @ 5 kg ha^{-1} at sowing + foliar application of humic substances extracted from vermicompost (0.2%) at 40 DAS and soil application of humic substances @ 5 kg ha^{-1} at sowing + foliar application of commercial humic substances (0.2 %) at 40 DAS which are statistically on par. Significantly least total dry matter (15.62 g plant^{-1}) was obtained with control (no humic substances application). The plots which received humic substances only through foliar spray was found inferior to soil applications. However, the plots receiving humic substances for both soil as well foliar was found better than those received either from soil or foliar alone.

Table 1: Yield and total dry matter of plant as influenced by humic substances at varied levels and method of application

Treatments	Grain yield (kg ha^{-1})	Haulm yield (kg ha^{-1})	Total dry matter at harvest g plant^{-1}
T_1: Control (POP)	1342	2210	15.62
T_2: Soil application of humic substances (humic and fulvic acid) @ 2.5 kg ha^{-1}	1520	2499	20.82
T_3: Soil application of humic substances (humic and fulvic acid) @ 5 kg ha^{-1}	1611	2699	21.76
T_4: Foliar application of humic substances (humic and fulvic acid) extracted from vermicompost (0.2 %) at 40DAS	1485	2434	19.59
T_5: Foliar application of commercial humic substances (humic and fulvic acid) 0.2 % at 40 DAS	1424	2246	18.45
T_6: T_2+T_4	1664	2669	22.71
T_7: T_2+T_5	1656	2732	22.43
T_8: T_3+T_4	1741	2862	23.75
T_9: T_3+T_5	1676	2772	22.99
T_{10}: Vermicompost on FYM 'N' equivalent basis (2.76 t ha^{-1})	1957	3134	23.85
S.Em. ±	46.59	104.5	0.79
C.D.(P=0.05)	138.41	310.6	2.35

Table 2: Plant uptake of major nutrients at harvest as influenced by humic substances at varied levels and method of application.

Treatments	Plant uptake (kg ha^{-1})								
	N			P			K		
	Grain	Haulm	Total	Grain	Haulm	Total	Grain	Haulm	Total
T_1: Control (POP)	85.9	49.2	135.1	7.4	4.4	11.8	9.8	61.8	71.6
T_2: Soil application of humic substances (humic and fulvic acid) @ 2.5 kg ha^{-1}	104.6	55.9	157.2	9.0	6.0	14.9	12.6	71.4	84.0
T_3: Soil application of humic substances (humic and fulvic acid) @ 5 kg ha^{-1}	109.0	60.4	169.5	9.5	6.7	16.2	13.8	77.7	91.5
T_4: Foliar application of humic substances (humic and fulvic acid) extracted from vermicompost (0.2 %) at 40DAS	96.9	57.9	154.8	8.4	5.6	14.0	11.9	69.0	80.9
T_5: Foliar application of commercial humic substances (humic and fulvic acid) 0.2 % at 40 DAS	97.9	44.0	141.8	8.0	4.9	12.9	10.7	63.3	73.9
T_6: T_2+T_4	112.7	48.6	161.3	10.2	7.2	17.4	15.1	77.7	92.8
T_7: T_2+T_5	112.2	57.5	169.6	9.9	7.1	17.1	14.7	79.1	93.8
T_8: T_3+T_4	122.6	71.0	193.7	11.3	8.3	19.6	16.4	83.9	100.3
T_9: T_3+T_5	117.3	65.4	182.7	10.6	7.8	18.3	15.6	80.9	96.4
T_{10}: Vermicompost on FYM 'N' equivalent basis (2.76 t ha^{-1})	135.6	70.8	208.4	12.9	9.4	22.3	18.8	92.8	111.6
S.Em. ±	2.19	1.79	3.46	0.33	0.31	0.62	0.47	1.57	1.86
C.D.(P=0.05)	6.50	5.32	10.29	0.97	0.92	1.85	1.39	4.67	5.53

Table 3: Major nutrients content of soil after harvest of crop as influenced by humic substances at varied levels and method of application.

Treatments	Major nutrients (kg ha^{-1})		
	Soil N	Soil P_2O_5	Soil K_2O
T_1: Control (POP)	223.63	99.16	277
T_2: Soil application of humic substances (humic and fulvic acid) @ 2.5 kg ha^{-1}	196.00	96.00	265
T_3: Soil application of humic substances (humic and fulvic acid) @ 5 kg ha^{-1}	180.63	94.70	257
T_4: Foliar application of humic substances (humic and fulvic acid) extracted from vermicompost (0.2 %) at 40 DAS	199.00	96.91	268
T_5: Foliar application of commercial humic substances (humic and fulvic acid) 0.2 % at 40 DAS	215.25	98.04	275
T_6: T_2+T_4	190.88	93.59	256

T_7: T_2+T_5	180.50	93.90	255
T_8: T_3+T_4	150.38	91.31	249
T_9: T_3+T_5	164.13	92.64	253
T_{10}: Vermicompost on FYM 'N' equivalent basis (2.76 t ha^{-1})	132.00	88.62	237
S.Em. ±	3.92	1.98	5.43
C.D.(P=0.05)	11.65	5.87	16.13

Conclusion

From the study, it can be inferred that application of vermicompost on FYM 'N' equivalent basis resulted in higher grain and haulm yield of soybean and also higher nutrient uptake. Further, the crop receiving soil application of humic substances (humic and fulvic acid) @ 5 kg ha^{-1} at sowing + foliar application of humic substances (humic and fulvic acid) extracted from vermicompost (0.2%) at 40 DAS helps for better nutrient uptake by the crop and to obtain higher yield in soybean.

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