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Effect of Organic Manures and Humic Acids on Nitrogen Concentration and Yield of Chickpea (JG-16)

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Abstract
A pot experiment was conducted to study the effect on nitrogen concentration and yield (dry matter and no. of pods pot^{-1}) of chickpea (JG-16) grown in a *Typic Haploumbrepts* soil amended with different N sources (*viz.*, urea, FYM, compost, vermicompost, N-enriched compost and the humic acids extracted from the corresponding organic manures) @ equivalent to the recommended dose of 20 kg N ha^{-1}. Results revealed that significantly higher amount of N and dry matter yield were accumulated in plants grown in soil amended with different N sources over control at different stages of crop growth. Application of organic manures enhanced the N concentration, dry matter yield and no. of pods plant^{-1} of chickpea as compared to the plants grown in the corresponding humic acid amended soils. Among the organic manures, significantly higher amount of N concentration was found in plant grown in compost while dry matter yield and no. of pods pot^{-1}was found significantly higher in FYM treated soils. Agronomic effectiveness of organic manures are more than the corresponding humic acids.

Keywords: Organic manure, humic acid, nitrogen concentration, yield, chickpea

Introduction
Nitrogen (N) occupies a conspicuous place in plant metabolism. All vital processes in plants are associated with protein, of which nitrogen is an essential constituent. It directly influences the amino acid composition of protein and thereby nutritional quality of the economic produce. Consequently to get more crop production, nitrogen application is indispensable and unavoidable. Nitrogen plays a key role in agriculture by increasing crop yield (Sonmez and Bozkurt, 2006; Ahmad *et al*., 2008) [42, 1]. Nitrogen not only enhances the yield but also increases photosynthetic processes, as well as net assimilation rate. Nitrogen is showing a fundamental role in enhancing the productivity of crops.

Understanding the role of nitrogen and its metabolic interaction in crops is an important step toward increasing productivity without compromise on quality. Nitrogen can be applied to soil either through natural sources or fertilizers. Natural source, whether it is from soil organic matter or added organic matter is of utmost importance with respect to maintenance of soil fertility as a whole and controlled release of nitrogen during cropping season. It is critical to understand what happens to organic nitrogen after soil application in order to use it efficiently for crop production. Its release from organic matter depends upon its source and state of decomposability (Khalil *et al*., 2001; Sajjad *et al*., 2003; Nourbakhsh and Dick, 2005; Mubarak *et al*., 2010; Baitilwake *et al*., 2012) [22, 31, 29, 27, 7]. Helgason *et al*. (2007) [18] reported that less than 5% of organic N content of composted manures was mineralized over 425 days suggesting that little of the organic N becomes available in the year of application. However, faster mineralization of manures was also reported with an initial rapid N release at 0-30 days, phase of constant release at 45-55 days, decline phase in N release at 70-90 days and sharp increase in N release at 120 days by Azeez and Averbeke (2010) [6] and Baitilwake *et al*. (2012) [7]. Dry matter production and N uptake applied with organic manures showed higher than the chemical fertilizer (Kodashuma *et al*., 2006; Sonmez and Bozkurt, 2006; Helgason *et al*., 2007 and Lehrsch and Kincaid, 2007) [23, 42, 18, 24].

Humic acid is the major constituent of humic substances and it is an integral part of organic matter and one of the key component of terrestrial ecosystem (Paul and Saha, 2016) [30]. Nitrogen content of the humic acid varies from 1-5% (Flaig, 1975) [16] depending upon its origin and sources of organic matter. Nitrogen present in humus can serve as a slow release

Nitrogen fertilizer (Flaig and Sochtig, 1967; Schnitzer and Khan, 1972; Filip and Kubat, 2003 and Nguyen *et al*., 2004a) [15, 36, 14, 28]. However, reports on more susceptible nature of humic acid to mineralization than organic manures were given by Satisha and Devarajan (2011) [35] and Devi and Saha (2017) [10]. Natural organic substances such as humic and fulvic acids play an essential role in ensuring soil fertility and plant nutrition. Application of humic acids was found to increase the total amount of nitrogen in plant (Govindasamy and Chandrasekaran, 1992; Sharif *et al*., 2002a and Sharif *et al*., 2004) [17, 40, 39]. Humic acids in small amounts act as specific sensitizing agents increasing the permeability of the cell membrane and resulting in an increased uptake of nutrients by the plants in large amounts (Senn and Kingman, 1973) [38]. Immobilization of N due to application of organic manures and humic acid was reported by Mohanty *et al.* (2011) [26] and Bird *et al*. (2002) [8]. So, understanding the effect of organic amendments on the agronomic efficiency of crops is a pre-requisite for managing nitrogen input for a given soil.

Considering the above points, the present investigation was undertaken to study the effect of organic manures and manure derived humic acids on N concentration and yield (dry matter and no. of pods pot^{-1}) of chickpea (JG-16) grown in a *Typic Haploumbrepts* soil.

Materials and Method

Composite soil samples (0 - 15 cm depth) were collected from the Central Agricultural University Research farm, Andro, Imphal East, Manipur, India following the standard procedure as described by Jackson (1973) [19]. The soil belongs to *Typic Haploumbrepts* (Sen *et al*., 2006) [37] having the general characteristic presented in table 1. The composite soil samples were air dried at room temperature in shade and passed through a 2 - mm sieve.

Four different organic manures *viz.* FYM, compost, vermicompost and N-enriched compost were prepared using paddy straw as raw material. Thermocol box was used as composting bin and holes were made in the box for the proper aeration so that there can be proper supply of oxygen. For preparing FYM, mixture of 4-5 cm long cut urine soaked paddy straw and cow dung slurry was put in the box, plastered with mud and left for 135 days. Four kg of partially decomposed cut paddy straw were placed layer by layer with cow dung slurry alternately in a thermocol box and vermiworm was added @ 4-5 matured worm kg^{-1} substrate for vermicompost preparation. Same process was followed without vermiworm for making compost. For preparing N-enriched compost urea was added @ 20g kg^{-1} raw material. Turning was done at 15 days interval. Compost, vermicompost and N - enriched compost were fully matured at 110 days.

Humic acid used in the present experiment was extracted from the corresponding organic manures using the method described by Stevenson (1996) [59]. Twenty gram (20 g) of air dried, ground and passed through 2mm sieve compost were placed in 250 ml conical flask and 200 ml of 0.5 N NaOH solution were added, shaken for 12 hours on a mechanical shaker and transferred to a Buchner funnel using whatman No 42 filter paper. Concentrated HCl was added dropwise and stirred with a glass rod until cloudiness appeared in the solution and let it sit overnight to complete the precipitation of humic acid. The solution were then centrifuged at 5000 rpm for 20 min. The humic acid was collected on the filter paper and dialyzed overnight to make ion salt free. The humic acid were then dried in an oven, ground and used for the experiment.

Total nitrogen content of the organic manures and their corresponding humic acid (HA) are FYM - 0.5208%, compost - 0.8008%, vermicompost - 1.0808%, N- enriched compost - 1.4864%, HA extracted from FYM - 2.142%, HA extracted from compost - 0.938%, HA extracted from vermicompost - 1.834% and HA extracted from N- enriched compost - 2.8%, respectively.

Four kg soils were filled in one hundred eighty (180) plastic pots. Each pot was sown with 5 seeds of chickpea (JG-16) and thinned out after 15 days of germination maintaining one plant per pot. Recommended doses of phosphorus and potassium @ 40 and 20 kg ha^{-1} were applied as basal in the form of DAP and MOP. Recommended dose of nitrogen @ 20 kg ha^{-1} was added through urea, different organic manures (FYM, compost, vermicompost, N-enriched compost) and humic acids derived from the corresponding organic manures maintaining different sets of treatments. Equivalent weight based on the recommended dose of nitrogen taken for different organic N sources were: FYM - 6.86 g pot^{-1}, compost - 4.46 g pot^{-1}, vermicompost - 3.31 g pot^{-1}, N-enriched compost - 3.29 g pot^{-1}, HA extracted from FYM - 1.67 g pot^{-1}, HA extracted from compost - 3.81 g pot^{-1}, HA extracted from vermicompost -1.95 g pot^{-1} and HA extracted from N- enriched compost - 1.28 g pot^{-1}. Urea, organic manures and humic acids were mixed thoroughly with the soil. Soils of each treatment were moistened to 60% of water holding capacity (WHC) and kept in a net house throughout the experiment. The loss of moisture was replenished by the periodic addition of sterile distilled water on every alternate day by difference in weight.

The treatments used in the study were: T_1= (control); T_2=RDN through urea; T_3=RDN through FYM; T_4= RDN through compost; T_5= RDN through VC; T_6 =RDN through N enriched compost; T_7= RDN through HA (FYM); T_8=RDN through HA (Compost); T_9=RDN through HA (VC) and T_{10}=RDN through HA (N enriched compost)

Plant samples were periodically collected on 0th, 20th, 40th, 60th, 80th days after sowing seeds and at harvest for determination of nitrogen content and recording dry matter yield. Crop yield (no. of pods pot^{-1}) of chickpea (JG-16) was also recorded at harvest. The experiment was carried out under CRD with 10 treatments replicated thrice.

Soil parameters like soil texture (hydrometer method), pH (1:2.5 soil: water suspension using glass electrode systronic pH meter), EC (1:2.5 soil: water suspension using systronic direct reading conductivity meter), organic carbon (Walkley and Black`s rapid titration method), cation exchange capacity (leaching with 1N NH_4OAc), available N (alkaline potassium permanganate), P (Bray and Kurtz No.1) and K (flame photometer) were analysed as described by Jackson (1973) [19]. Plant N concentration was estimated by Kjeldahl method (Jackson, 1973) [19].

Table 1: General characteristics of the soil used in the experiment

Soil characteristics	Results
i. Textural class	Clay
ii. Sand (%)	5.78
iii. Silt (%)	22.56
iv. Clay (%)	72.79
v. Maximum water holding capacity (%)	44.71
vi. pH (1:2.5 soil: water ratio)	5.26
vii. EC (1:2.5 soil: water ratio, dsm^{-1})	0.15
viii. CEC [$cmol(p^+)kg^{-1}$]	9.40
ix. Organic carbon (%)	1.53
x. Total nitrogen (mg kg^{-1})	1191.13
xi. Available nitrogen (mg kg^{-1})	84.00
xii. Available phosphorus (mg kg^{-1})	16.79
xiii. Available potassium (mg kg^{-1})	17.50

3. Results and Discussion

3.1 N concentration of chickpea grown in soil amended with organic manures and humic acids

Data on changes in the amount of N concentration in plant grown in soil amended with organic manures and humic acid derived from the manures are presented in Figure 1. Results showed that the trend of changes in N concentration of chickpea grown in organic manures and humic acid treated soils occurred similarly with gradual increase up to 40th day followed by a decline up to 80th and finally increased at harvest. However, N concentration in plant grown in urea treated soil showed a different trend with an increase till 40th day and then declined on 60th followed by an increase till harvest. Comparing with control, significantly higher N was accumulated in plants grown in soil amended with different N sources at different stages of crop growth. This is at par with the findings of Sharif *et al*. (2002a) [40]; Sharif *et al.* (2002b) [41]; Sharif *et al.* (2004) [39]; Sarir *et al*. (2005a) [34] and Sarir *et al*. (2005b) [33]. Further study revealed that comparatively higher amount was observed in organic manures treated soil than corresponding humic acid treated soil on 60th, 80th days after sowing seeds and at harvest. Reports on ineffectiveness of humic substances in promoting plant N accumulation were given earlier by Fernandez *et al*. (1996) [12]. Significantly higher amount of N concentration was found in plants grown in soils treated with FYM followed by vermicompost and compost grown plants on 60th and 80th day, respectively. However, at the time of harvest N content in chickpea grown in compost treated soil was significantly higher which is at par with that grown in FYM amended soil.

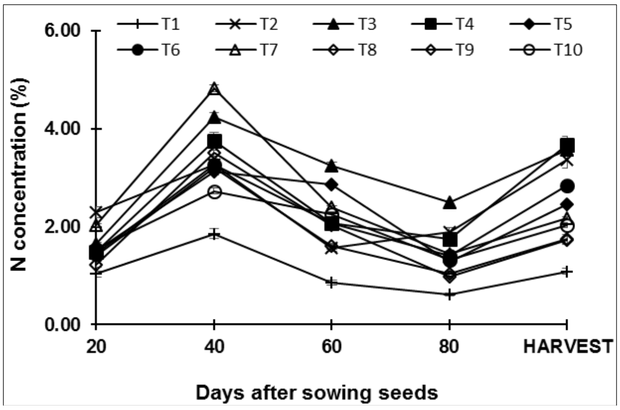

Fig 1: N concentration of chickpea grown in soil amended with organic manures and humic acids (error bars show the standard deviation from the mean)

3.2 Dry matter yield of chickpea grown in soil amended with organic manures and humic acids

There was an increasing trend of dry matter yield up to 80th day with slight decline at harvest (Figure 2). In general, comparatively higher dry matter yield of chickpea was observed in soil treated with urea, organic manures and humic acid over control at different growth stages. Enhanced agronomic effectiveness of organic sources of N was reflected in increased dry matter yield (Arancon *et al*., 2003; Khadija *et al*., 2004 ; Defline *et al*., 2005 and Mohajerani *et al*., 2016) [4, 21, 9, 25]. Critical examination of data revealed that comparatively higher dry matter yield of chickpea was found in plant grown in compost treated soil which is at par with the plants grown in N- enriched and FYM treated soils at 80th days after sowing seeds. However, at the time of harvest plants grown in FYM treated soils showed significantly higher dry matter yield which is followed by compost and vermicompost grown plants. Application of organic manures enhanced dry matter yield of chickpea significantly as compared to the plants grown in the corresponding humic acid amended soils.

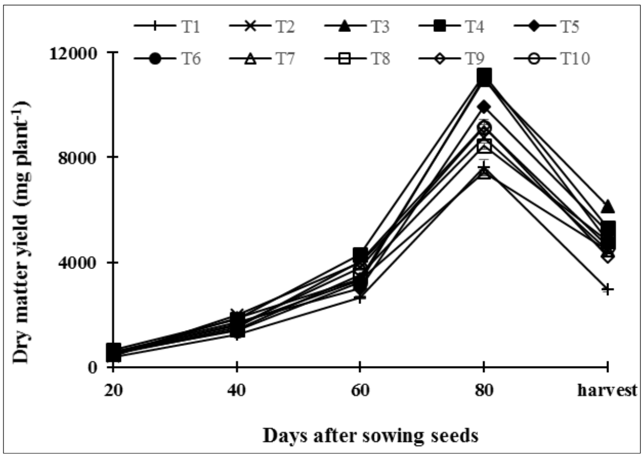

Fig 2: Dry matter yield (mg kg^{-1}) of chickpea grown in soil amended with organic manures and humic acids (error bars show the standard deviation from the mean)

3.3. Yield (Number of Pods plant $^{-1}$) of chickpea grown in soil amended with organic manures and humic acids

Data revealed that chickpea grown in soil amended with different N sources give significantly higher no. of pods plant^{-1} over the untreated control (Figure 3). Reports on higher pod yield due to organic N application was also revealed earlier by Eghball and Power (1999) [11]; Santhy *et al*. (2001) [32]; Sharif *et al.* (2002b) [41]; Arshad *et al.* (2004) [5]; Sharif *et al.* (2004) [39]; Sarir *et al*. (2005b) [33]; Sonmez and Bozkurt (2006) [42]; Ahmad *et al*. (2008) [1]; Ferrara and Brunetti (2008) [13]; Akhtar *et al*. (2014) [2]; Ali and Mindari (2016) [3]; Mohajerani *et al*. (2016) [25]; Kahraman (2017) [20]; Yadav *et al*. (2017) [44]. Further study pointed out that significantly higher no. of pods was observed in chickpea grown in urea treated soil followed by FYM addition. Detailed study revealed that number of pods plant^{-1} was significantly higher in chickpea grown in organic manures as compared to the corresponding humic acid extracted from organic manures. Sharif *et al*. (2002a) [40] also revealed no significant effect of humic acid application on crop yield.

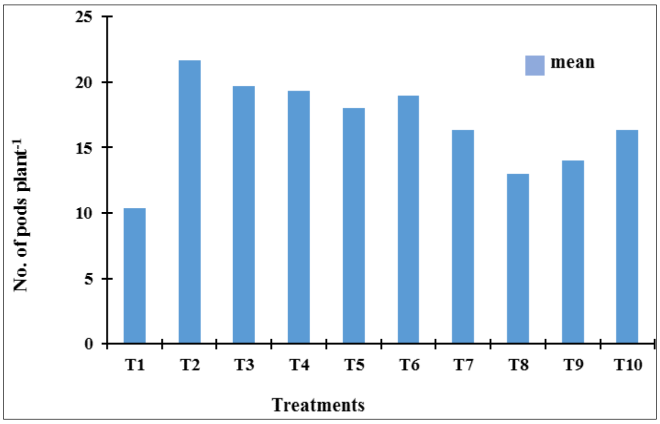

Fig 3: No. of Pods plant^{-1} of chickpea grown in soil amended with organic manures and humic acids

Conclusion
Significantly higher N was accumulated in plants grown in soil amended with different N sources over control at different crop growth stages. Application of organic manures enhanced the N concentration, dry matter yield and no. of pods plant^{-1} of chickpea as compared to the plants grown in the corresponding humic acid amended soils. This shows higher agronomic efficiency of organic manures comparing with the corresponding humic acids. Among the organic manures, significantly higher amount of N concentration was found in plants grown in compost while dry matter yield and no. of pods pot^{-1}was found significantly higher in FYM treated soils.

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