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Release of Nitrogen and CO_2 evolution from Manures and Fertilizers applied in Soil: An incubation study

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Abstract: Carbon and nitrogen mineralization and urease activity were evaluated under wet and dry climate with input of manures and fertilizers. Results indicates that release of NH_4^+ and NO_3^-–N and CO_2 evolution was maximum during 7 to 21 days after incubation (DAI) with recommended dose of fertilizer and decreased later on. Release of N and C took place after 21 DAI where manures applied either with neem cake or fertilizers. The evolution of CO_2–C and total nitrogen were significantly higher in poultry manure combined with neem cake and FYM as compared to vermicompost along with neem cake. The NH_4^+–N released during the entire incubation period were higher than NO_3^-–N. Combination of neem cake, poultry manure and FYM had higher enzyme activity than other treatments. Application of commercial fertilizers registered significantly higher CO_2–C fluxes upto 35 DAI compared to manures and control. Overall effect of manures applied with neem cake or their combinations proved better for release of nitrogen. Correlation study between CO_2–C flux, total N and urease activity showed a positive relationship ($p<0.05$) with each others.

Key words: *Manures, N mineralization, carbon evolution, urease activity.*

Introduction

Fertile and well-managed soils form the basis for efficient crop production. Soil amendment could play an important role in plant nutrition in the organic agriculture movement as well as under the integrated soil fertility system (John *et al*. 2004). Additions of organic manures synchronize between the nutrient release and plant uptake and showed that slower release fertilizers can increase plant yield and reduce nutrient leaching. Since the sole use of mineral fertilizer has been reported to lack the capacity to sustain productivity under the continuous intensive cropping system found in most commercial farms (Benbi *et al.* 1998). Further Yaduvanshi (2003) and Beckman (1973) reported that the application of organic material had improved the soil physical properties, nutrient supply and crop yield over time. The application of organic materials only as source of nutrient for crop is often not effective in the short run due to their low nutrient content and the time required for decomposition/mineralization to take place naturally (Rahman *et al.* 2001). Yet, continuous application is known to yield some benefits in the long run. Organic materials such as crop residues, cattle dung, poultry manure, and their composted products like vermicompost are available in abundance in India (Bhattacharyya and Kumar 2005; Ramesh *et al.* 2005), and their use as organic amendments for improving the soil productivity (Ramesh *et al.* 2006). Therefore, combined application of two or more sources of organic manures is one of the ways to improve the nutrient supply and also to some extent synchronize the nutrient supply with crop requirement and is likely to improve the soil fertility and thereby crop performance. The turnover and mineralization of residues largely depends on soil biological processes, and its rate of decomposition by soil microorganisms ensures the recycling of nutrients

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that can be reused by plants and microbes. The duration of this phenomenon is known to be affected by the chemical characteristics of the organic materials *viz.,* C:N ratio, lignin, polyphenols, total N content and climatic variables such as moisture, temperature and relative humidity (Tian *et al.* 1995; Bayala *et al.* 2005). Thus, there is a need to determine the immobilization or mineralization time lag; this will provide information that could be used to synchronize the time of organic materials application with that of plant nutrient need. The soil microbial respiration (CO_2), soil nutrients and enzyme activities respond much more quickly to the changes in soil management practices as compared to total soil organic matter (Dick 1992; Doran *et al.* 1996). Therefore, measurement of CO_2 and N dynamics are provides a critical period for organic matter turn over and nutrients availability. Therefore, our study was set up to examine nutrient dynamics under different manures and fertilizer treatments and at specific gap period.

Materials and Methods

A 70 days incubation experiment was conducted at Department of Soil Science and Agricultural Chemistry, College of Agriculture (PJTSAU), Rajendranagar, Hyderabad, (Telangana) India, to know the mineralization pattern of nitrogen and CO_2 evolution from different organic manures and fertilizers. The soil (*Alfisol*) was collected at depth of 0-15 cm from a frequently cropped field at the college research farm. The soil had pH 8.1, EC 0.42 dS m^{-1}, Organic C 7.9 mg kg^{-1}, available N 82.9 mg N kg^{-1}, available P 5.6 mg P kg^{-1} and available K 76.1 mg K kg^{-1}. Manures, vermicompost and neem cake were collected from National Institute of Rural Development (NIRD), Rajendranagar, Hyderabad. Poultry manure was collected from AICRP on Poultry Breeding, Sri Venkataswara Veterinary University, while FYM was collected from Dairy Farm, College of Veterinary Science, Rajendranagar, Hyderabad. The physico-chemical characteristics of the organic materials are presented in Table 1.

The experiment was laid out in a CRD (completely randomized design) with each treatment being replicated thrice. The treatments details are T1–Control, T2–RDF (recommended dose of fertilizers) N, P and K (100, 50 and 50 kg ha^{-1}), T3–100% RDN through FYM, T4–100% RDN through vermicompost, T5–100% RDN through poultry manure, T6–100% RDN through neem cake, T7–50% RDN through FYM + 50% RDN through vermicompost, T8–50% RDN through FYM+ 50% RDN through poultry manure, T9–50% RDN through FYM + 50% RDN through neem cake, T10–50% RDN through vermicompost + 50% RDN through poultry manure, T11–50% RDN through vermicompost + 50% RDN through neem cake and T12–50% RDN through poultry manure + 50% RDN through neem cake.

Plastic containers having capacity of 1.5 kg were filled with 1.0 kg processed soil. Prior to filling required quantity of organic manures (FYM, poultry manure, vermicompost, neem cake) and inorganic fertilizers [urea, single super phosphate (SSP) and muriate of potash (MOP)] were mixed in soil as per treatment. Low quantity of fertilizers was dissolved in water for a treatment and equal amount of solution was distributed in respective replications to make the replication homogenous and application of manures is based on N content. The treated soil was brought to about 60% of water holding capacity (WHC) and then incubated at 25±1 °C. These plastic containers were covered with polythene sheet to prevent ammonia volatilization and evaporation losses. Soil samples were collected at 0, 7, 14, 21, 35, 50 and 70 days interval and

Table 1. Nutrient composition of different organic manures

Manures	pH	EC (dS m^{-1})	Total nutrients (g kg^{-1}) N	P	K
FYM	7.2	1.12	107	40	78
Poultry Manure	7.0	1.62	347	133	112
Vermicompost	7.5	0.35	112	40	73
Neem Cake	5.1	1.45	343	30	121

analyzed for nitrogen, urease activity and carbon mineralization by evolving CO_2 under different treatments.

A combined glass calomel electrode was used to determine the pH of aqueous suspension (1:2.5 soils: water ratio). Organic carbon was determined by Walkley and Black (1934), available nitrogen by Subbiah and Asija (1956) methods. Available potassium was extracted with 1*N* NH_4OAc and then measured by using flame photometer. Available phosphorus was extracted by Olsen method (Olsen *et al.* 1954) and then determined by colorimetric spectrophotometer. Potential soil nitrogen mineralization and nitrification were measured using laboratory incubations (Robertson *et al.*1999). Mineralization of nitrogen in exchangeable forms (NH_4 and NO_3–N) are estimated by three step, first was extraction using 2*M* KCl solution (Dorich and Nelson 1983) followed by steam distillation method using MgO–Devarda alloy after removal of NH_4–N form the same sample used to estimate NO_3^-–N by adding Devarda alloy. Urease activity (µg of NH_4^+ N released g^{-1} soil h^{-1}) analysed by Tabatabai and Bremner (1972) method with modifications as suggested by Rao (1989). Soil respiration (CO_2 evolution) was estimated by titration method. The amount of CO_2–C trapped was determined by back titration of unused NaOH with 1*N*HCl. The net CO_2-C was obtained by subtracting the value of the control from that of the treatments, using the relationship reported by (Anderson 1982).

$CO_2 = (B - V)NE$ Where B = volume of HCl used to titrate NaOH in the beaker to the end point of the control; V = volume of acid used to titrate NaOH in the beaker to the end point; N = normality of the acid; E = equivalent weight (to express data as C, E = 6, while as CO_2 = E = 22);

The mineralized CO_2-C recorded for the first week was the cumulative value obtained daily for that week.

Measured data were analyzed for analysis of variance (ANOVA) using SAS statistical package (SAS Institute 1999) to examine the effect of different manures and period on mineralization of manures. Statistical significance of each attribute was assessed using Fisher's least significant difference (LSD) at $p<0.05$ level.

Result and Discussion

CO_2 evolution

The CO_2 fluxes in the treated and control soil during the incubation period is shown (Fig.1). The CO_2 fluxes from soil with organic materials were consistently higher at all the sampling times compared with the control. Application of commercial fertilizers (T2) registered significantly higher CO_2 fluxes up to 21 days after incubation (DAI) compared to manures and control treatment. Treatments T_8 and T_{10} showed CO_2 fluxes up to 35 DAI, and T_5, T_{12} and T_9 were dominated after 35 DAI. Application of organic manures and fertilizer resulted in significantly ($p<0.05$) higher CO_2 fluxes (9.3) at 35 DAI compared to initial and later stage. Maximum CO_2 fluxes' peak was observed from organic manures treatment during the last 50 and 70 DAI. Results showed that the amendment of soil with the organic materials positively influenced C mineralization at all the sampling periods. The increased in CO_2 efflux (up to 35 DAI) was due to the priming effects of the added organic materials, which is known to occur at the active phase of organic C release (Curtin *et al.* 1998; Khalil *et al.* 2005). Treatments having manures combination had higher carbon mineralization than others. Similar results were also reported by Gupta *et al.* (2004) and Surekha *et al.* (2004) that the soil respiration rate measured as microbial activity increased significantly in manure treatments over control.

The cumulative percentage of C released from the total C added to soil by the treatments (organic materials and fertilizer) is shown in Fig. 2. Total

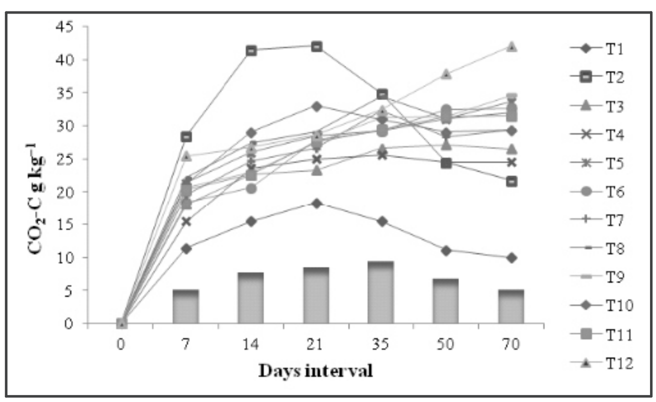

Fig. 1. Influence of manures and fertilizer on carbon mineralization from incubated soil. Bars represent LSD (0.05).

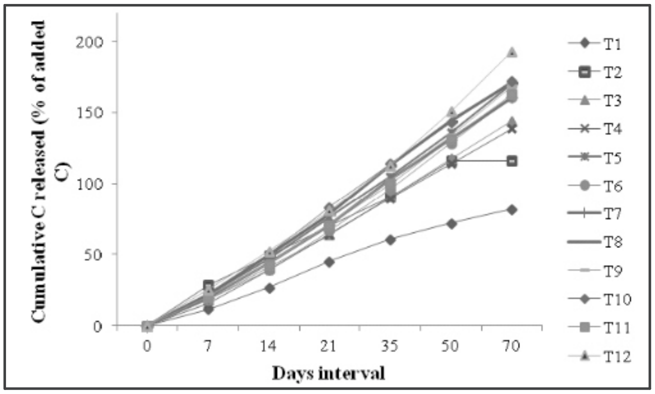

Fig. 2. Influence of manures and fertilizer on cumulative carbon mineralization from incubated soil.

C released were significantly ($p<0.05$) different at all sampling period between the control and treatment with organic material. At 50 DAI, the application of fertilizer resulted in 11.3 per cent decrease in the cumulative amount of C released compared to 35 DAI. At 50 DAI, application of poultry manure (T_{12}), FYM (T9) combination with neem cake resulted in 11.1 per cent and 8.4 per cent higher cumulative C.

The application of individual manures and fertilizer, maximum CO_2 evaluation was observed in earlier days of incubation and gradually declined. This indicates that re-moistening created a congenial environment in the soil for optimum microbial activity in the beginning without any lag period accelerated the decomposition of native soil organic matter as well as the readily available (Gaur *et al.* 1971). The lower efflux of CO_2-C from poultry manure treated soils, compared with other manures, was due to rapid mineralization of the readily hydrolysable C in the fresh materials; this is known to be low in stable compost. This resulted in further breakdown of the organic materials and mineralization of their nutrient content. The C:N ratio of soil decreased with fertilization. The organic amendments showed a greater effect in decreasing C: N ratio compared to inorganic fertilizers. A decrease in soil C: N ratio with organic amendments indicates build-up of N pool in the soil.

Nitrogen mineralization

Addition of manures and fertilizers increased NH_4^+–N and NO_3^-–N content of the soils (Fig. 3 and Fig. 4). The release nitrogen from manures and fertilizers were dependent on the manure combinations and duration. Immobilization of nitrogen was observed in manures treatments up to 21 DAI. After 21 DAI, net mineralization of N increased with the application of organic materials. The application of poultry manures and FYM with neem cake had higher net mineralized N than the sole application of the organic materials.

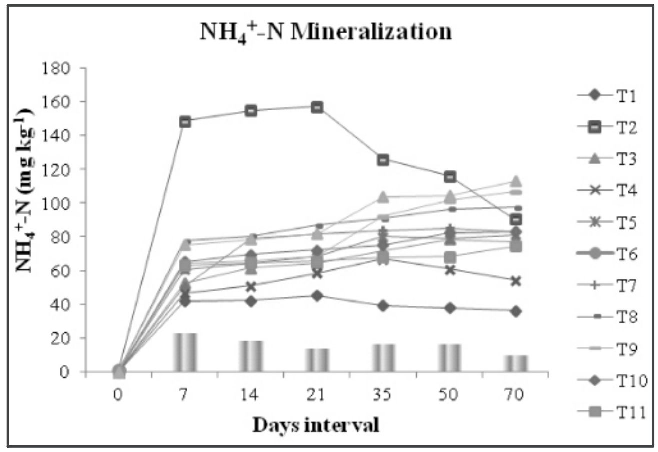

Fig. 3. Influence of manures and fertilizer on NH_4^+–N from incubated soil. Bars represent LSD (0.05).

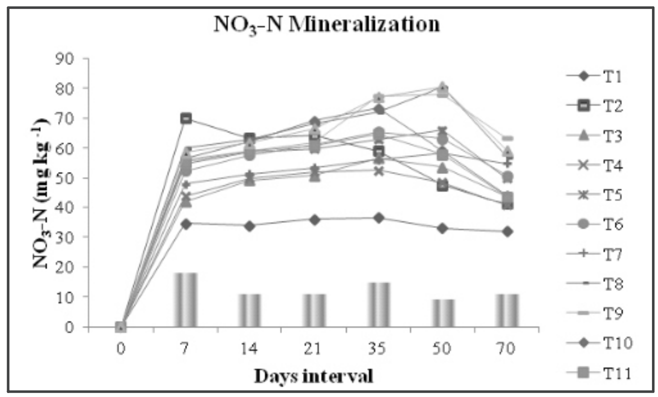

Fig. 4. Influence of manures and fertilizer on NO_3^-N from incubated soil. Bars represent LSD (0.05).

Among the treatments, T2 had maximum release of NH_4^+–N content up to 21 DAI followed by 35-70 DAI, wherein decreasing order was observed. Even though, fertilizer treatment had maximum release of NH_4^+–N content up to 50 DAI than manures and manures combination. In the succeeding weeks, NH_4^+–N content release was more in manures treatments. Among the manures combination, T8 treatment had higher release of NH_4^+–N content up to 21 DAI. The NO_3^-–N content, since 7 DAI to 50 DAI gradually increased higher in manures treatments but later on decreased. Among the treatments, T2 had dominant release of NO_3^-–N content up to 14 DAI. Treatments like T8, T10 and T12 had more release of NO_3^-–N content over others. The poultry manure combined with FYM and neem cake showed significantly higher release of NO_3^-–N content from 21 DAI to 70 DAI. NO_3^-–N levels generally increased with incubation time which was probably due to the transformation of ammonium to nitrate.

In general, NH_4^+–N content released during the entire incubation period were higher than NO_3^-–N content. In the initial period, fertilizer treatment had maximum N mineralization, but later on manure combinations showed better result. The net mineralization of N among the applied organic materials were not significantly different from one another, but substantially higher in other treatments than the control owing to N content of the applied materials. Other factors as the soil C:N ratio, soil macro fauna, soil microbial activities and soil pH have been reported to contribute to N mineralization when organic materials are added to the soil (Aulakh *et al.* 2000; Khalil *et al.* 2005).

Addition of manures and fertilizers increased NH_4^+-N in the soil. The increase was due to hydrolysis of urea and mineralization of organic nitrogen in manure treatments. In earlier periods of manure treatments, nitrogen mineralization was slow because addition of organic manures delayed the hydrolysis of urea. The N mineralized in a given quantity of soil with specific time lag dependent upon temperature, available water, rate of oxygen replenishment, pH, amount and nature of plant residues, and level of other nutrients. Addition of manures and fertilizers had great influence on mineralization of nutrients than control for the entire incubation period. Prasad and Singhania (1989), Trehan (2000) and Kar *et al.* (2007) also reported that application of manures or fertilizers increased NH_4^+–N and NO_3^-–N in the soil. The higher mineral N from the application of poultry manure, FYM and their separate combination with neem cake, further confirm the suitability of fresh organic materials as a more suitable soil amendment. Moreover, these materials also exhibited a high degree of ammonification concurrently with mineralization; which signified the occurrence of N immobilization and rapid remineralization over time (Khalil *et al.* 2005).

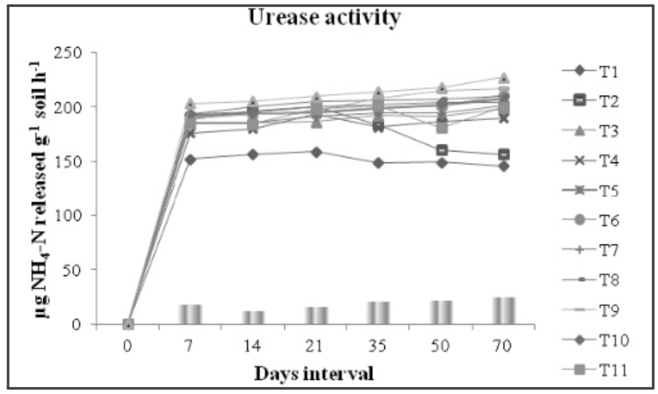

Fig. 5. Influence of manures and fertilizer on urease activity from incubated soil. Bars represent LSD (0.05).

Urease enzymes activity

Addition of nutrients through manures and fertilizers showed significant increase in enzyme activities (Fig. 5). Among the treatments, T12 showed higher urease activity followed by T8 and T2 upto 21 DAI. During the later period, the influence of T9 was more. In general, neem cake, poultry manure and FYM combinations released high urease enzyme during incubation. This may be due to the organic matter being more readily available to microorganisms through manures than in the fertilizer. Addition of organic sources acts as good source of carbon and energy to heterotrophs by which their population increased with an increase in enzymic activities. Similar relationships between organic carbon and enzymic activities were reported by Bohme and Bohme (2006).

In fertilizer treatment, there was sharp increased in urease activity up to 35 DAI and thereafter it declined rapidly. On the other hand, application of organic manures gradually increased urease activity up to 70 DAI except T10 and T11. Such variations could largely be due to changes in the organic carbon content of soil (Beri and Goswami 1978). Increased microbial population was responsible for an increase in urease activity of soil enzymes. The applied organic sources were able to get mineralized rapidly in early days of incubation and hence there was more mineralization than immobilization which consequently provided sufficient nutrition for the proliferation of microbes and their activities in terms of soil enzymes. Since, the mineralized nutrients were highly available up to 70 DAI, therefore rapid increase in

Table 2. Correlation matrix between the different soil properties

Parameters	CO_2	NH_4^+	NO_3^-	Urease activity
CO_2	1			
NH_4^+	0.782**	1		
NO_3^-	0.691**	0.566**	1	
Urease activity	0.786**	0.493**	0.757**	1

microbial activity was observed during this period of incubation. The same has also been confirmed by the findings of Nannipieri *et al.* (1983).

Relationship between variable

The relationship between CO_2 flux from the added organic materials and fertilizer was showed in Table 2. A highly significant positive relationship was shown by CO_2 flux with NH_4^+–N (r= 0.782), NO_3^-–N (r = 0.691) and urease activity (r = 0.786). Similarly, Urease activity was significantly correlated with NH_4^+–N (r = 0.493), NO_3^-–N (r = 0.757). The application of organic materials and fertilizer had significant relationship with the chemical properties of the soil. Similar relationship was also reported by Valarini *et al.* 2004; Khalil et *al.* 2005.

Conclusions

Soil available N and microbial activities were increased by application of organic amendments and fertilizer. Combinations of neem cake with poultry manures and FYM had better result of N availability. N availability varied based on decomposition and mineralization rate of manures and fertilizer.

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