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PJBS

ISSN 1028-8880

**Pakistan
Journal of Biological Sciences**

ANSI*net*

Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Response of Wheat (*Triticum aestivum* L.) to Application of Nitrogenous Fertilizer and Sewage Sludge

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Abstract: Interactive effect of inorganic fertilizer and sewage sludge on nitrogen nutrition and growth of wheat was studied. Nitrogen was applied as ^{15}N -labelled $(\text{NH}_4)_2\text{SO}_4$ at 0, 50, 150, and 300 mg pot^{-1} in all possible combinations with 0, 16, 24, 32, and 64 g pot^{-1} sewage sludge (SS). Fertilizer N had no significant effect on the dry weight of roots. The above-ground plant components responded positively to the application of both fertilizer N and SS. The positive effect increased with the rate of application. In absence of SS, grain yield increased from 4.8 g pot^{-1} in the control to 10.6 g pot^{-1} at the highest level of fertilizer N. Likewise, the increase in grain yield due to different treatments ranged between 97 and 233% as compared to 23.3 and 82.5% recorded for straw component. The trends in N content of different plant components were fairly similar to those observed for dry matter yield and a significant correlation was observed between two parameters. Combination of both treatments at highest rate resulted in 127% increase in the total N yield of the plants. The contribution of N fertilizer to the total N content of the whole plant and its components remained fairly low and ranged between 11 and 45% in different treatments. The percent fertilizer N uptake (%FNU) varied from 22.6% at the highest level of application in the absence of sludge to 79.4% at the lowest level of application and in the presence of highest amount of SS. Fertilizer N uptake increased with the amount of SS; the extent of increase being more at the lower level of fertilizer N. Application of SS significantly improved the amount of unlabeled N determined in plants, with maximum effect being observed at the highest level of application. A part of this increase was due to N uptake from SS itself, while a substantial amount could be derived from the soil organic matter.

Key words: Nitrogen fertilizer, fertilizer efficiency, ^{15}N , sewage sludge, wheat

Introduction

The increased production of sewage sludge (SS) on world-wide basis has led to the consideration of its application to deserts, forests and agricultural lands as a means of disposal, nutrient cycling and enhancing the ecosystem productivity (Benton and Wester, 1998; Ferrier *et al.*, 1996; Hani *et al.*, 1996; Harrison *et al.*, 1996; Lerch *et al.*, 1992). The information already available suggests a highly positive effect of SS on ecosystem functioning (Benckiser and Simarmata, 1994; McGrath *et al.*, 1994). However, some reservations prevail on the possible negative effects due to the contamination of SS with pathogens, heavy metals and organic pollutants (Benckiser and Simarmata, 1994; McGrath *et al.*, 1994; Unken, 1987).

The problem of pathogens in SS could fairly be overcome by aerobic/anaerobic digestion. Radiation of the material before use may be more assuring (Anonymous, 1994) but it requires the facilities for mass scale radiation. Nevertheless, this approach is worth consideration. Aside from pathogens, heavy metal content of most sewage sludges, particularly in situations where industrial wastes get mixed with domestic wastes, may pose a serious problem following accumulation after repeated applications and their entry into plant and animal systems (McGrath *et al.*, 1994; Ibeke *et al.*, 1998; Weissenhorn *et al.*, 1995). However, once the metals are in soil there is limited removal by plants or movement down the soil especially in heavy soils high in organic matter (Koskela, 1985). Persistence of elevated concentrations of heavy metals in the plough layer following repeated applications of sludge has been reported (Chang *et al.*, 1984). It would appear that soils in Pakistan which are generally very low in organic matter may retain relatively higher content of mobile heavy metals following sludge application. However, the positive effects of SS through supplemental nutrient supply and improvement in physico-chemical and biological properties of soil may more than balance the negative effects of heavy metals.

Soils in Pakistan are fairly low in organic matter content (generally less than 1%) because of the prevailing climatic conditions (high temperatures, wetting and drying) that are quite conducive for a rapid loss of C. Therefore, SS may prove as a good organic amendment for such soils. Traditionally, application of SS to farm lands is not very common in Pakistan and there is no systematic collection of SS for use in agriculture. However, with increasing environmental concerns of unorganized dumping of SS and the realization that it could help increase agricultural productivity, the situation may change in near future.

The objectives of the experiment reported here were to study the effect of γ -irradiated sewage sludge on i) growth of wheat, ii) uptake of N from ^{15}N -labelled fertilizer and soil/sewage sludge.

Materials and Methods

Soil: The soil was collected from the experimental fields at the Nuclear Institute for Agriculture and Biology, Faisalabad, Pakistan. Air-dried and sieved (<2 mm) soil sampled to a depth of 20 cm, had the following characteristics: organic C, 0.56%; total N, 0.08%; pH (saturation paste), 7.7; water holding capacity, 28%; sand, 52%, silt, 28%; clay, 20%.

Sewage sludge: The sewage sludge obtained from a treatment plant in Islamabad had the following characteristics: pH, 7.05; organic C, 23.2%; total N, 1.5%; C/N, 15.5; P, 0.55%; K, 0.2%; Zn, 63.5 ppm; Fe, 5596 ppm; Mn, 288 ppm; Co, 18.7 ppm; Cu, 113 ppm. A portion of the sludge contained in cuboid packets was radiated at Pakistan Radiation Services (PARAS), Lahore, at a dose of 5 kGy. Density of coliform bacteria in irradiated and non-irradiated sludge samples was 12 and 900 (MPN index per 100 ml), respectively, suggesting an almost complete kill of the pathogenic bacteria.

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Experimental setup: Five-kg portions of air-dried and sieved soil were filled in plastic pots and amended with nitrogen and SS. Nitrogen was applied as ^{15}N -labelled $(\text{NH}_4)_2\text{SO}_4$ at 0, 50 (10 atom % ^{15}N), 150 (2 atom % ^{15}N), and 300 (2 atom % ^{15}N) mg pot^{-1} in all possible combinations with 0, 16, 24, 32, and 64 g pot^{-1} SS. Triplicate pots were used for each treatment. The treated soil was adjusted to desirable moisture level (15% of the soil) and sown to wheat (*Triticum aestivum* L. var. Punjab-96). Five seeds were sown pot^{-1} and the stand was thinned to 3 seedlings following establishment of seedlings. Irrigation was given as required using tap water. The plants were harvested at physiological maturity and data on total dry matter yield and its distribution in grain and straw portions was recorded.

Analyses: Aliquots of oven-dried (70 °C) and finely powdered root, straw and grain portions were analyzed in triplicate for Kjeldahl N (Bremner and Mulvaney, 1982). The distillates thus obtained were processed (Buresh *et al.*, 1982, Mulvaney, 1986) for ^{15}N isotope ratio analysis on an IRGA mass spectrometer with a precision of ca 0.001 atom % ^{15}N . The data was subjected to analysis of variance followed by least significant test.

Results and Discussion

Fertilizer N had no significant effect on dry weight of roots that varied between 2.8 and 3.1 g pot^{-1} (Table 1). Application of sewage sludge (SS) alone or along with fertilizer N had an inconsistent effect on root dry matter, with both a negative and positive effect being observed. This inconsistency may be attributed to the difficulty in complete recovery of roots from the soil. However, both negative and positive effects of fertilizers and organic amendments on root biomass have often been reported (Azam, 1990; Azam *et al.*, 1991). The above-ground plant components responded positively to the application of both fertilizer N and SS. In the absence of SS, dry matter of straw increased from 15.2 g pot^{-1} in the control to 17.2 g pot^{-1} at the highest level of fertilizer N. Application of SS at the highest level led to an increase of 40.7%. A maximum of 82.5% increase was observed when both SS and fertilizer N were applied. In general, the positive effect of both the amendments increased with the rate of application; the two showing a high degree of synergism. Trends in response of the grain component to different treatments were essentially similar to those noted for straw, although the effect was more pronounced. For example, in the absence of SS, grain yield increased from 4.8 g pot^{-1} in the control to 10.6 g pot^{-1} at the highest level of fertilizer N. Likewise, the increase in grain yield due to different treatments ranged between 97% and 233% as compared to 23.3% and 82.5% recorded for the straw component. There appeared to be a significant improvement in harvest index (partitioning of dry matter to grains) due to the application of N fertilizer and SS (data not presented). It increased from 0.24 in the control to 0.38 at the highest level of N fertilizer when applied alone; the change due to sludge was from 0.24 to 0.37. Combination of the two treatments did not cause consistent changes in harvest index. Benefit of fertilizer N in terms of increased crop biomass is universally accepted and does not need further literature support. A major portion of the nitrogenous fertilizers is applied to cereal crops (Raum and Johnson, 1999). In comparison to fertilizer N, the effect of organic amendments is variable depending upon their chemical composition,

especially N concentration. Benefits of organic amendments are derived mainly from a net release of N from decomposing organic matter with high N concentration and narrow C/N

Table 1: Dry matter yield and its distribution in different plant components

| Treatments ¹ | SS0 (g pot^{-1}) | SS1 | SS2 | SS3 | SS4 | % increase or decrease over SS0 | | | | |
|-----------------------------|--------------------------------|-------|-------|-------|-------|---------------------------------|--|--|--|--|
| | | | | | | | | | | |
| Roots | | | | | | | | | | |
| N0 | 3.1 | 7.1 | 20.0 | -14.5 | 19.7 | | | | | |
| N1 | 2.8 | 0.3 | 17.1 | -7.4 | 4.2 | | | | | |
| N2 | 3.0 | 3.6 | -15.8 | 17.1 | -4.2 | | | | | |
| N3 | 3.0 | 4.5 | -9.4 | 10.3 | 10.0 | | | | | |
| LSD (P=0.05), 0.3 | | | | | | | | | | |
| Straw | | | | | | | | | | |
| N0 | 15.2 | 23.3 | 34.6 | 30.9 | 40.7 | | | | | |
| N1 | 16.1 | 29.7 | 44.1 | 51.0 | 82.5 | | | | | |
| N2 | 17.2 | 45.7 | 38.1 | 37.6 | 68.6 | | | | | |
| N3 | 17.2 | 46.9 | 47.2 | 49.3 | 55.3 | | | | | |
| LSD (P=0.05), 1.5 | | | | | | | | | | |
| Grain | | | | | | | | | | |
| N0 | 4.8 | 111.7 | 113.3 | 122.9 | 162.7 | | | | | |
| N1 | 6.1 | 116.5 | 129.2 | 115.4 | 158.1 | | | | | |
| N2 | 8.6 | 97.1 | 127.9 | 143.5 | 160.0 | | | | | |
| N3 | 10.6 | 157.7 | 137.9 | 185.4 | 233.3 | | | | | |
| LSD (P=0.05), 1.1 | | | | | | | | | | |
| Root + Straw + Grain | | | | | | | | | | |
| N0 | 23.1 | 44.5 | 53.5 | 53.0 | 70.0 | | | | | |
| N1 | 25.0 | 50.5 | 64.5 | 66.5 | 100.7 | | | | | |
| N2 | 28.8 | 58.0 | 59.0 | 63.0 | 90.5 | | | | | |
| N3 | 30.8 | 73.5 | 69.0 | 82.0 | 98.0 | | | | | |
| LSD (P=0.05), 2.8 | | | | | | | | | | |

¹N0, N1, N2, and N3 are the rates of N addition i.e., 0, 50, 150, and 300 mg pot^{-1} , respectively; SS0, SS1, SS2, SS3, and SS4 are the rates of sewage sludge addition i.e., 0, 16, 24, 32, and 64 g pot^{-1} , respectively.

Table 2: Nitrogen yield and its distribution in different plant components

| Treatments ¹ | SS0 (mg pot^{-1}) | SS1 | SS2 | SS3 | SS4 | % increase or decrease over SS0 | | | | |
|-----------------------------|---------------------------------|-------|-------|-------|-------|---------------------------------|--|--|--|--|
| | | | | | | | | | | |
| Roots | | | | | | | | | | |
| N0 | 20.7 | 25.9 | 36.4 | 1.7 | 37.1 | | | | | |
| N1 | 19.7 | 15.3 | 17.0 | 9.2 | 15.2 | | | | | |
| N2 | 22.7 | 32.8 | -7.8 | 41.9 | 0.2 | | | | | |
| N3 | 23.1 | 14.2 | 3.4 | 29.4 | 24.8 | | | | | |
| LSD (P=0.05), 2.1 | | | | | | | | | | |
| Straw | | | | | | | | | | |
| N0 | 48.8 | 34.6 | 56.9 | 52.0 | 54.5 | | | | | |
| N1 | 56.6 | 45.9 | 45.9 | 68.4 | 109.2 | | | | | |
| N2 | 54.5 | 81.7 | 65.0 | 47.2 | 102.8 | | | | | |
| N3 | 61.9 | 58.4 | 78.5 | 60.1 | 68.7 | | | | | |
| LSD (P=0.05), 7.7 | | | | | | | | | | |
| Grain | | | | | | | | | | |
| N0 | 53.8 | 79.5 | 111.4 | 89.1 | 165.1 | | | | | |
| N1 | 74.4 | 133.9 | 141.4 | 121.2 | 178.9 | | | | | |
| N2 | 102.2 | 91.8 | 115.0 | 169.6 | 204.1 | | | | | |
| N3 | 102.7 | 166.9 | 150.7 | 203.3 | 218.5 | | | | | |
| LSD (P=0.05), 12.2 | | | | | | | | | | |
| Root + Straw + Grain | | | | | | | | | | |
| N0 | 123.3 | 52.7 | 77.2 | 59.7 | 99.8 | | | | | |
| N1 | 150.7 | 60.8 | 82.7 | 81.5 | 123.8 | | | | | |
| N2 | 179.4 | 96.2 | 94.5 | 99.7 | 129.7 | | | | | |
| N3 | 187.7 | 98.3 | 97.4 | 117.3 | 126.6 | | | | | |
| LSD (P=0.05), 20.9 | | | | | | | | | | |

¹see table 1 for details

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Table 3: Percent N of different plant components and whole plant derived from fertilizer

| Treatments | SS0 | SS1 | SS2 | SS3 | SS4 |
|-----------------------------|------|------|------|------|------|
| N1 | 13.8 | 13.2 | 12.5 | 13.1 | 10.6 |
| N2 | 17.7 | 14.7 | 16.2 | 16.8 | 14.4 |
| N3 | 21.5 | 21.6 | 20.8 | 18.8 | 18.8 |
| LSD (P=0.05), 2.3 | | | | | |
| Straw | | | | | |
| N1 | 16.0 | 19.1 | 16.0 | 17.0 | 12.4 |
| N2 | 24.1 | 23.0 | 25.2 | 21.0 | 23.1 |
| N3 | 27.0 | 26.2 | 28.9 | 28.1 | 24.0 |
| LSD (P=0.05), 3.8 | | | | | |
| Grain | | | | | |
| N1 | 23.0 | 18.1 | 17.3 | 16.1 | 16.3 |
| N2 | 38.0 | 37.0 | 39.1 | 36.1 | 31.0 |
| N3 | 45.0 | 41.0 | 42.1 | 42.1 | 43.1 |
| LSD (P=0.05), 5.3 | | | | | |
| Root + Straw + Grain | | | | | |
| N1 | 19.2 | 17.9 | 16.4 | 16.2 | 14.4 |
| N2 | 31.2 | 29.3 | 31.9 | 35.7 | 27.0 |
| N3 | 36.2 | 34.1 | 35.5 | 29.4 | 35.2 |
| LSD (P=0.05), 3.3 | | | | | |

see Table 1 for details

Table 4: Effect of soil treatments on % fertilizer N uptake by plants (%FNU), fertilizer N derived from fertilizer (Ndff) and plant N derived from soil or/and sewage sludge (Ndfs)

| Treatments | SS0 | SS1 | SS2 | SS3 | SS4 |
|----------------------------------|-------|-------|-------|-------|-------|
| %FNU | | | | | |
| N0 | 0 | 0 | 0 | 0 | 0 |
| N1 | 57.8 | 70.5 | 73.9 | 72.3 | 79.4 |
| N2 | 37.3 | 47.3 | 45.5 | 48.2 | 51.0 |
| N3 | 22.6 | 27.8 | 28.8 | 31.8 | 32.8 |
| LSD (P=0.05), 4.21 | | | | | |
| Ndff, mg pot⁻¹ | | | | | |
| N0 | 0 | 0 | 0 | 0 | 0 |
| N1 | 28.9 | 35.3 | 37.0 | 36.1 | 39.7 |
| N2 | 55.9 | 71.0 | 68.3 | 72.2 | 76.5 |
| N3 | 67.9 | 83.3 | 86.3 | 95.4 | 98.3 |
| LSD (P=0.05), 7.32 | | | | | |
| Ndfs, mg pot⁻¹ | | | | | |
| N0 | 123.3 | 188.3 | 218.5 | 196.9 | 246.3 |
| N1 | 121.8 | 163.0 | 188.3 | 187.6 | 236.3 |
| N2 | 123.5 | 171.0 | 146.9 | 174.0 | 206.8 |
| N3 | 119.8 | 161.2 | 157.0 | 172.6 | 181.2 |
| LSD (P=0.05), 15.67 | | | | | |

see Table 1 for details

ratio. Thus green manures are reported to have a positive effect on crop yields (Azam, 1990; Ventura and Watanabe, 1993). However, the benefits are derived more from overall positive effects rather than from N supply alone (Ladd *et al.*, 1983; Azam, 1990). Studies carried out with ¹⁵N-labelled leguminous materials revealed that only a small proportion of the applied organic N is available to plants (Azam, 1990; Azam *et al.*, 1985; Ladd *et al.*, 1983; Woods *et al.*, 1987). Sewage sludge used in the present study had a relatively wider C/N ratio (15.5) and low N concentration (1.5%). Therefore, the positive effect observed may not be solely due to the additional N supply by SS, but could have resulted from additional benefits in terms of increased microbial activity. Organic amendments, including application of SS, have been reported to increase the crop yields as a result of improvement in physico-chemical and biological characteristics of the soil and availability of plant nutrients especially N (Hani *et al.*, 1996; Lerch *et al.*, 1992; McGrath *et al.*, 1994). Hence, any increase in crop yield will be attributable to improvement in physico-chemical and biological characteristics of the soil.

Such effects of organic amendments have frequently been reported (Morris *et al.*, 1986). In some other studies (Azam *et al.*, 1999 and unpublished data), a consistent ¹⁵N enrichment instead of dilution of plant N in different treatments suggested that SS did not make a net contribution to plant available N.

Table 2 gives the data on % increase or decrease in N content of the whole plant and its components. The trends in N content of different plant components were fairly similar to those observed for dry matter yield and a significant correlation was observed between the two parameters (coefficient of correlation for root, straw, grain and total dry matter being 0.93, 0.98, 0.97 and 0.93, respectively). The close correlation clearly demonstrated the dependence of dry matter yield on N content of the plants and thus the availability of the later. Thus, SS also appeared to exert a positive effect on dry matter yield through its contribution to N availability either directly (N being released from SS) or indirectly through enhanced release of N from soil organic matter. Interaction of applied N with the native soil N resulting in greater mineralization and plant uptake of the later has been reported (Jenkinson *et al.*, 1985; Hart *et al.*, 1986; Woods *et al.*, 1987; Azam, 1990; Azam *et al.*, 1991). The two kinds of treatments (fertilizer N and SS) had a fairly synergistic effect on N yield of the three plant components. When applied alone, fertilizer N caused 52% increase (calculation based on last set of data in column 2, Table 2) in the N content of the whole plant at the highest rate of application. However, when the same amount was applied in the presence of the lowest level of SS the benefit increased to 98%. Combination of both the treatments at highest rate of application resulted in ca 127% increase in the total N yield of the plants. Synergistic effects of inorganic and organic N sources have been reported (Azam, 1990). In a previous study, sewage sludge was found to serve as an additional source of N as well as to conserve fertilizer N (Azam *et al.*, 1999).

The contribution of N fertilizer to the total N content of the whole plant and its components remained fairly low and ranged between 11 and 45% in different treatments (Table 3). Thus 55-89% of the plant N was derived from SS and soil. The grain portion showed the maximum concentration of fertilizer N (16-45%), the straw portion derived 12 to 30% of its N from fertilizer, while roots contained 11-22% of their N from fertilizer. In general, there was a significant increase in the proportion of fertilizer N in all three components with the amount applied. In all cases, application of SS lead to a decrease in the contribution of applied N to the total N of whole plants or components. For example, at the lowest level of application, contribution of fertilizer N to the whole plant N decreased from 19.2% in absence of sludge to 14.4% at the highest level of sludge. A decrease in contribution of fertilizer N to total plant N was understandable because of the N availability from SS and soil organic matter that would be higher at increasing rates of application. Contribution of fertilizer N to the plant N is reported to decrease in the presence of other N sources (Mian and Stewart, 1985; Azam, 1990).

The percent fertilizer N uptake (%FNU) varied from 22.6 at the highest level of N application in the absence of sludge to 79.4% at the lowest level of application and in the presence of the highest amount of SS (Table 4). Fertilizer N uptake increased with the amount of SS; the extent of increase being more at the lower level of fertilizer N. Plant recovery of fertilizer N decreased with the amount applied at all levels of SS. However, the net amount of fertilizer N (mg N derived from fertilizer pot⁻¹) increased significantly with the rate of application at all levels of sludge. The improvement in fertilizer

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N availability could be attributed to its net immobilization in the presence of SS. Organic residues low in N concentration and having wider C/N ratio are reported to cause a net immobilization of fertilizer N (Azam *et al.*, 1985). By causing immobilization of fertilizer N, SS ensured its prolonged and continued availability to plants thereby leading to higher fertilizer use efficiency. Thus SS had a stabilizing and conserving effect on fertilizer N. In present study, although the net amount of fertilizer N taken up by plants increased at higher rate of application, % FNU (% fertilizer N uptake) decreased, which is a commonly observed phenomenon (Maman *et al.*, 1999). In present study, SS enhanced the availability of added ^{15}N labelled N. Not only the plants treated with SS derived a higher percentage of their N from applied ^{15}N -labelled fertilizer but the absolute amounts of fertilizer N taken up by plants was also substantially higher compared to non-sludge treatment. This may mean that sludge conserved fertilizer N (by providing additional C for microbial immobilization) and the plants were able to make use of it over an extended period of time. In view of being a labile source of carbon but not that of N (Lerch *et al.*, 1992), SS could be expected to cause an immobilization of fertilizer N, but with restricted release of N over short term.

Application of SS significantly improved the amount of unlabeled N determined in the plants, with a maximum effect being observed at the highest level of application. A part of this increase was due to N uptake from SS itself, while a substantial amount could be derived from the soil organic matter. Organic amendments are known to enhance the plant availability of soil N through added nitrogen interaction (Azam, 1990; Jenkinson *et al.*, 1985; Hart *et al.*, 1986; Woods *et al.*, 1987). While, increasing levels of fertilizer N had an almost negligible effect on unlabeled N in the plants in absence of SS, a substantial decrease was observed in sludge-amended soils. Plant N derived from soil was not affected by the amount of fertilizer N when applied alone.

Acknowledgments

This work was partially financed by FAO/IAEA, Vienna, Austria through a CRP, and Alexander von Humboldt Foundation of Germany. Mass spectrometric analysis of soil and plant samples were carried out at RIAD, PINSTECH, Islamabad. Technical assistance of Mr. Ansar Mahmood is thankfully acknowledged.

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