

Effect of Organic amendments on Nitrogenase activity (nmoles C₂H₄/20 g soil/hr) of Azospirillum lipoferum

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ABSTRACT

In the present study the effect of different organic amendments like compost, partially composted paddy straw was seen on nitrogenase activity of *A. lipoferum*. Both organic amendments were applied at 0.5% and 1%. The observations were taken at 7th and 20th day after incubation. The inoculation of soil with *A. lipoferum* enhanced the nitrogenase activity of the soil substantially. Amongst the two organic amendments, partially biodegraded paddy straw with the C/N ratio 32:1 showed greater activity than compost.

Paddy straw was biodegraded at two moisture levels. Paddy straw partially biodegraded at 80% moisture level applied at the rate of 1% showed the maximum nitrogenase activity but it was less than *Azospirillum* inoculated soil. The beneficial role of partially degraded paddy straw and improving the nitrogenase activity may be explained that it might have served as a better source of carbon and energy for this organism.

I. Introduction:

The effect of different factors was examined on nitrogen fixation by *Azospirillum* inoculation using acetylene-ethylene assay method. Rice and Paul 1972 found that nitrogenase activity in a waterlogged soil straw system was related to the number of nitrogen fixing clostridia and to some extent to the microbial counts. The nitrogenase activity indicated a change in the microbial activity at about the 8th day of incubation this was attributed to the sequential use of the different organic components with a short lag in activity between the periods of maximum hemicellulose and cellulose utilization

II. Materials and Methods.

Soil was collected from topsoil for use in the experiment. It was air dried and passed through a 2 mm screen. The soil analysis showed that it was neutral in reaction and was poor in organic matter (organic carbon 0.28%) and total nitrogen (0.035%), the water holding capacity of the soil was 36%.

Incubation Studies:

One hundred grams soil samples were weighed, appropriate amount of distilled water was added and mix to maintain the soil moisture at 1/3rd of its water holding capacity except in case of study with different levels of moisture. Soil was inoculated with *Alipoferum* (strain 12) according to the treatments. Keeping proper controls and were placed in 250 ml conical flasks. The flasks were incubated 4 different periods under aerobic conditions at 30 degree centigrade. Samples were drawn at periodic intervals and assay was done.

Procedure for nitrogenase activity:

20 grams soil samples were drawn, transferred to conical flasks and one person glucose was added for substantial amount of easily utilizable energy source. Soils after 24 hours were put in the 100 ml vacutainer brand vials, were stoppered and the gas phase was replaced with high pure Acetylene (10% by volume) through a gas tight hypodermic syringe. Vials were incubated at 30 degrees centigrade for one hour. One ml sample of the gas phase from each tube was analysed for ethylene production on a gas chromatography fitted with hydrogen flame ionization detector and PorPack Q of 80-100 mesh column temperature at 90 degree centigrade. N₂ nitrogen at flow rate 80 ml per minute nitrogenase activity is expressed as μ moles C₂H₄ per 20 gram soil per hour.

Preparation of organic materials:

Three different organic sources i.e. Finished compost and partially degraded Paddy straw at 80% moisture and 100% moisture for two months at room temperature (25-30°C) were used. Compost and partially degraded Paddy straw were air-dried and pass through 2 mm sieve. These were incorporated in soil at the rate of

0.5 and 1%. samples were drawn at 7 and 25 days of incubation and assays were done. The Treatments were as given below:

Soil alone

Azospirillumlipoferum

Compost (0.5%)

Compost (0.5%)+ A.lipoferum

Compost (1 %)

Compost (1 %)+ A.lipoferum

Paddy straw (80% moisture)(0.5%) P .S.I

P .S.I(0.5%)+ A.lipoferum

P .S.I(1.0 %)

P .S.I (1.0 %) + A.lipoferum

Paddy straw(100 % Moisture)(0.5 %) P.S.II

P.S.II (0.5%) + A.lipoferum

P.S.II (1.0%)

P.S.II (1.0%)+ A.lipoferum

Samples were drawn on 7th and 20th days and nitrogenaseactivity wasdetermined

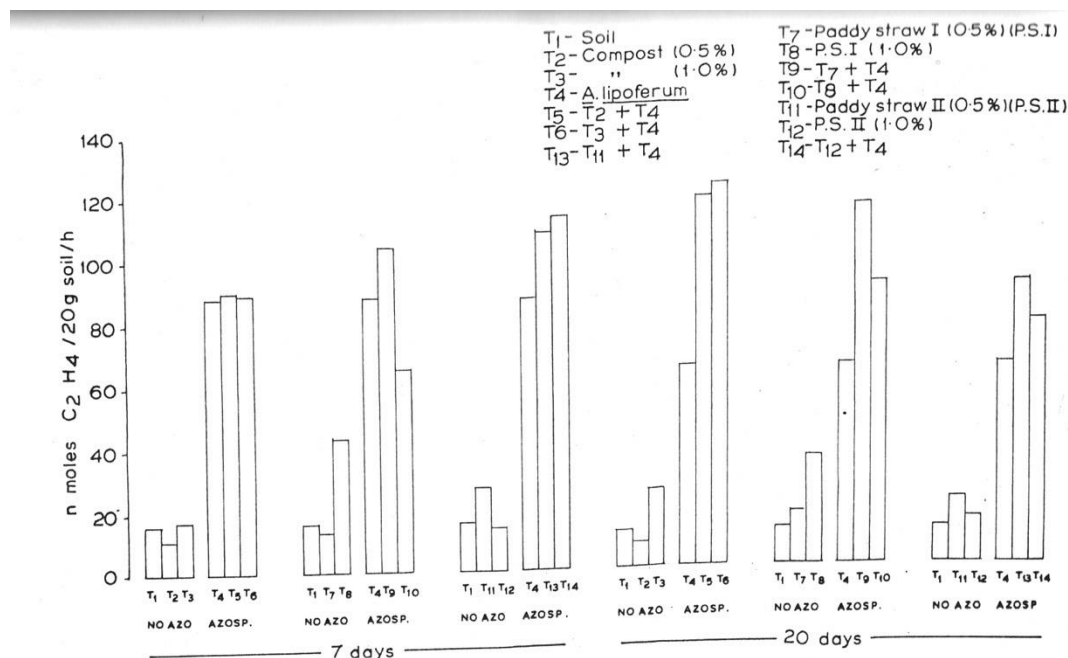
III. RESULTS :

The effect of different organic amendments on nitrogenase activity of A.lipoferum Is presented in table 1

Treatments	Incubation		Mean
	7	20	
Soil	16.03(256.96)	12.03(144.72)	14.03
Compost (0.5%)	11.52(132.71)	8.74(76.39)	10.13
Compost(1 %)	17.15(294.12)	25.31(640.60)	21.23
A.lipoferum	88.76(7878.34)	64.23(4125.49)	76.49
Compost (0.5%) + A.lipoferum	90.37(8166.74)	118.70(14089.69)	104.54
Compost(1%) + A.lipoferum	89.38(7988.78)	122.97(15121.62)	106.18
Partially composted paddy straw (P.S. I) 0.5%	12.99(168.74)	17.98(323.28)	15.49
P.S.I(1%)	43.43(1886.16)	35.03(1227.10)	39.23
P.S.(0.5%)+ A.lipoferum	104.51(10922.34)	115.29(13291.78)	109.9
P.S.I(1%)+ A.lipoferum	65.89(4341.49)	90.88(8259.17)	78.39
Partially composted paddy straw (P.S.II) (0.5%)	26.92(724.69)	21.11(445.63)	24.02
P.S.II (1%)	14.29(204.20)	15.01(225.30)	14.65
P.S.II(0.5%)+A.lipoferum	108.69(11813.52)	90.96(8273.72)	99.82
P.S.II(1%)+ A.lipoferum	113.46(12873.17)	78.77(6204.71)	96.12

	Stages.	Treatments.	Interaction
S.em+	0.44	1.16.	1.64
C.D. 5%	1.25.	3.30.	4.67

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Effect of different organic amendments on nitrogenase activity of *A. lipoferum*
Fig. 4

A significant increase in nitrogenase activity of *A. lipoferum* from inoculated soil (88.76 64.23 n moles C₂ H₄ as compared to control (16.03 and 12.03 n moles C₂ H₄) was found at 7th and 20th day of incubation respectively. On 7th day of incubation where partially composted Paddy straw(P.S.I) at 1% and partially composted Paddy straw(P.S.II) (0.5%) were applied to soil increased the nitrogenase activity significantly While the data with P.S. I at 0.5% compost and P.S.II at 1 % We're almost identical to control.

The nitrogenase activity of soil when inoculated with *Azospirillum* was stimulated with compost (90.37 and 89.38 n moles C₂H₄) at the rate of 0.5 and 1% respectively, P.S.I (104.5 n moles C₂H₄) at 0.5% and P .S. II (108.69 and 113.46 n moles C₂ H₄) at 0.5 and 1 % as compared to inoculated control (88.76 n moles C₂H₄) except P.S.I. (65.89 n moles (C₂H₄) at 1%. On 20th day of incubation except compost (8.74 n moles C₂ H₄) at 0.5% There was a significant increase in organic amended soil (25.31 ,17.98, 35.03 ,21.11 ,15.01 n moles C₂ H₄) compared to unamended soil (12.03 n moles C₂H₄). There was also a significant increase in nitrogenase activity of inoculated soil with different amendments in comparison to inoculated soil alone.

The mean value was highest when P.S. I was applied at(0.5%) with. *A. lipoferum* followed by compost(106.18 n moles C₂H₄) at 1% And compost (104.54 n moles C₂H₄)at 0.5%. The mean value also shows that each amendment either at 0.5% or at 1 % is stimulatory for nitrogenase activity except compost at 0.5%.

IV. Discussion:

The inoculation of soil with *A. lipoferum* enhanced the nitrogenase activity of the soil substantially (Figure). Hegazi etal. (1980) reported that nitrogenase activity of soil was positively correlated with counts of spirilla.

The amendment of soil with finished compost (C/N ratio 15:1) during the first week of incubation was not effective in increasing the nitrogenase activity of soil. However partially biodegraded paddy straw with a

C/N ratio 32:1 showed greater activity than compost. Paddy straw partially biodegraded at 80% moisture applied at the rate of 1% showed the maximum nitrogenase activity but it was less than *Azospirillum* inoculated soil. The results also showed that the nitrogenase activity of *A. lipoferum* was augmented in presence of partially biodegraded straw irrespective of moisture regime for straw biodegradation. Rai (1982) also reported that partially biodegraded straw was better source of energy than FYM for nitrogen fixation by *Azospirillum*.

The results of nitrogenase activity recorded after 20 days of incubation showed that paddy straw continued to be a better source for nitrogenase activity of the uninoculated soil. The performance of 1 percent compost at this stage was found favourable for enhanced nitrogenase activity which was comparable to paddy straw biodegraded at 80% moisture level. It was observed at this stage of incubation that compost irrespective of dosage proved slightly superior to paddy straw in inoculated soil. Paddy straw biodegraded at 80% moisture and applied at the rate of 0.5% was quite comparable to compost and continued to be best source from the very beginning.

The beneficial role of partially degraded paddy straw in improving the nitrogenase activity may be explained that might have served as a better source of carbon and energy for this organism. These findings are corroborated by Charyulu and Rao (1980) who has reported that the addition of rice straw stimulated the population of *Azospirillum* in soil and their nitrogen fixing activity.

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