Effect of Spentwash Application on Nitrogen Dynamics in Soil

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***Abstract*— Distillery spentwash contains all nutrients and organic matter and used in agriculture as a source of plant nutrients and irrigation water. Besides all the nutrients, spentwash contains appreciable amount of nitrogen also. The effect of different levels and methods of spentwash application on soil nitrogen dynamics was examined through a field experiment. The field experiment was conducted using Groundnut (*Arachis hypogea* L.) as a test crop. At all stages of groundnut growth, the amounts of NH_4-N and NO_3-N were greater in soil that received 120 m^3 of spentwash with the recommended dose of NP fertilizers. Results shown that the spentwash not only adds mineral N (NH_4-N and NO_3-N) to soil, but also promotes the mineralization of soil organic N, thus resulting in large amounts of NH_4-N and NO_3-N in soil. The present study was, therefore, undertaken to study the nitrogen dynamics in soil under spentwash application and its impact on soil, crop and environmental quality.**

***Index Terms*—Levels of spentwash, Mineralization, Nitrogen, Nitrogen dynamics.**

I. Introduction

Nitrogen plays an important role in increasing the agricultural production and as a constituent of protein, it increases the food value. It also influences the quality of environment. Available N includes NH_4^+, NO_3^- and NO_2^- forms. Since, NO_2 is highly unstable in soil, it is either immediately oxidized to NO_3^- form or reduced to NH_4^+ form. The dynamic of these two forms is influenced by the aeration status of the soil. The predominant form present is NO_3^-. The NO_3^- accumulated in the soil is either taken up by the crop or leaches down to the lower soil horizon as it is readily soluble in water. Some amount of NO_3-N is also immobilized by soil microbes during the process of mineralization of organic matter.

The spentwash, being loaded with organic and inorganic compounds could bring remarkable changes on the physical, chemical and biological properties of soils and thus influences the fertility of soil significantly. However, indiscriminate disposal results adverse impact on soil and environmental health [1]. Most studies investigating effluent irrigation and leaching losses have focused on NO_3-N [2], but in certain situations NH_4-N or organic N may be the dominant form of N to be leached. Information are scarce on nitrogen dynamics in soil under spentwash application and its environmental significance. Such information are needed for developing guidelines or strategies for effective utilization of distillery spentwash without any environmental hazards.

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II. Materials and Methods

A. Collection and characterization of spentwash

The biomethanated distillery spentwash sample was collected from the Salem Co-operative Sugar Mills Ltd, Mohanur, Namakkal District, Tamil Nadu, India. and characterized for its nutritive value and pollution potential by following standard methods [3], and used in the experiment (Table 1).

B. Experimental details

A field experiment was conducted at Research and Development Cane Farm, The Salem Co-operative Sugar Mills Ltd., Mohanur, Namakkal, District Tamil Nadu, India using Groundnut (*Arachis hypogea* L.) variety TMV 7 @ 125 kg/ha during the rabi season (August 31, 2007 to December 11, 2007). The experimental soil was taxonomically belongs to the family *Typic Rhodustalfs*. The soil was medium in mineral-N, high in $NaHCO_3$-P and high in NH_4OAc-K status (201.6, 18.1 and 278.0 kg /ha, respectively). A representative soil sample, at 0-15 cm depth, was collected from the experimental plot to determine the initial properties of the soil.

Design of the experiment: Split plot design; Replication: 3; Plot size: 13.5 m^2 size; Spacing: 30 cm x 10 cm

Main plots: M_1 - One time application; M_2 - Continuous split doses of application

Sub plots: T_1 – Control; T_2 - RD of NP; T_3 - Spentwash @ 40 m^3/ha; T_4 - Spentwash @ 40 m^3/ha+ RD of NP; T_5 - Spentwash @ 80 m^3/ha; T_6 - Spentwash @ 80 m^3/ha+ RD of NP; T_7 - Spentwash @ 120 m^3/ha; T_8 - Spentwash @ 120 m^3/ha+ RD of NP. (RD – Recommended Dose)

The different levels of spentwash was applied to the field uniformly by spraying manually to each plot 15 days before sowing for first main plot treatment (M_1). In the second main plot treatment (M_2), the spentwash was applied in three equal splits along with irrigation water. The first split dose of spentwash was applied 15 days after sowing. The crop was supplied with N and P fertilizers, as per the treatments at the recommended dose of 17 and 34 kg/ha, respectively. The K was entirely supplied through the spentwash. All other routine cultural operations until the harvest of the crop were followed as per the recommendations of crop production guide of Tamil Nadu Agricultural University.

C. Chemical analysis

$KMnO_4$ –N

The $KMnO_4$– N content of the soil was determined by the alkaline permanganate method [4].

Nitrogen fractions

The fractionation of soil nitrogen was carried out by the procedure given by [5] and [6]. The individual fractions were estimated according to the procedure given by [7].

- *Preparation of Equilibrium extract:* A quantity of 10 g of soil (0.2 mm) treated with 100 ml of 2 M KCl solution was shaken for one hour and filtered through Whatman No.1 filterpaper.

- *Ammonical nitrogen (NH_4-N):* An aliquot of 20 ml of the above filterate was distilled with freshly ignited MgO in distillation apparatus and the distillate collected in 2 % boric acid containing mixed indicator was titrated with standard H_2SO_4.

- *Nitrate nitrogen (NO_3-N):* On removal of NH_4 from the sample, added a pinch of devarda's alloy and continued the steam distillation. The amount of NO_3-N was determined as described for NH_4-N.

Statistical analysis

The data on various characters studied during the investigation were statistically analysed by the method given by [8]. The critical difference was worked out at 5 per cent (0.05) probability levels.

III. Results

Application of different levels of spentwash had remarkable impact on both NH_4-N and NO_3-N in soil (Tables 2 and 3). Before sowing NH_4-N content was only 72 mg/kg in the control; whereas, it ranged from 138 to 282 mg/kg under one time application of spentwash (M_1). Increase in the levels of spentwash significantly increased the NH_4-N and such increase was more due to NP fertilizers application. Soil that received 120 m^3 of spentwash plus NP fertilizers had the highest concentration of NH_4-N (282 mg/kg).

During the crop growth, the concentration of the NH_4-N was decreased both at pod formation and at harvest. At pod formation stage, the NH_4-N content ranged between 39 and 120 mg/kg due to one time application of spentwash; whereas, it was between 46 and 136 mg/kg under continuous application of split doses of spentwash. Significant difference was observed between treatments and also between the two methods of spentwash application. Further reduction in the concentration was observed in soils collected after the harvest of crop.

Initially, 15 days after one time application of spentwash (before sowing) the concentration of NO_3-N varied from 276 to 564 mg/kg, as against only 96 mg/kg in the control and 106 mg/ kg in soil that received only NP fertilizers. The highest concentration was observed in T_8 (spentwash at a rate of 120 m^3/ha plus NP fertilizers). During the crop growth, the NO_3-N was found decreased drastically which ranged from 56 to 226 mg/kg at pod formation, and 28 to 86 mg/kg at harvest. Continuous application (split application) of spentwash also significantly increased the NO_3-N content. The concentration was higher at pod formation stage, ranged between 124 and 240 mg/kg as against 40 mg/kg in the control. Marked reduction was observed after harvest. Increase in the rate of spentwash application had increased the NO_3-N significantly; however, such difference was narrow down in the post harvest soil. In general, continuous application of split doses of spentwash resulted in relatively higher amount of NO_3-N in soil.

Table 1. Characteristics of biomethanated distillery spentwash

S. No	Characters	Biomethanated spentwash*
1.	Colour	Dark brown
2.	Odour	Unpleasant burnt sugar
3.	pH	7.1
4.	EC (dS/m)	38
5.	Total dissolved solids	50000
6.	Total suspended solids	3300
7.	Total solids	53300
8.	Biological oxygen demand	12800
9.	Chemical oxygen demand	35000
10.	Carbon (g/L)	24
11.	Nitrogen	420
12.	Phosphorus	40
13.	Potassium	9097
14.	Sodium	357
15.	Calcium	4600
16.	Magnesium	1752
17.	Chloride	13471
18.	Bicarbonates	195
19.	Sulphate	947
20.	Oil and grease	19.6
21.	Total sugars (%)	3.49
22.	Reducing sugars (%)	1.77
23.	Total phenols	84
24.	Zinc	7.20
25.	Iron	78
26.	Manganese	5.3
27.	Copper	5.5
29.	Bacteria (x 10^6 CFU/ml of effluent)	12
30.	Fungi (x 10^4 CFU/ml of effluent)	19
31.	Actinomycetes (x 10^3 CFU/ml of effluent)	Nil

** Mean of triplicate samples; (Values are in mg/ L unless otherwise stated)*

IV. Discussion

A. Dynamics of nitrogen

The results of field experiment have shown that the transformation and the plant availability of N in soil was greatly influenced by the spentwash application. The two methods of spentwash application differed significantly in affecting the N dynamics. Before sowing (15 days after spentwash application), both NH_4-N and NO_3-N were significantly higher in soil with one time application, whereas, at later stages (pod formation and harvest stages), continuous application recorded higher values (Table 2 and 3). One time application (M_1) of single dose of spentwash added large amount of N and increased the ammonification and nitrification, resulting in higher concentration of NH_4-N and NO_3-N in soil. Similar results were also observed in the incubation experiment. At later stages crop removal and losses due to NH_3 volatilization and NO_3 leaching might have reduced the NH_4 and NO_3 contents in soil. In continuous application (M_2), nutrients were added to soil in split doses continuously which gradually increased the concentration of NH_4-N and NO_3-N. One time application or continuous application of different levels of spentwash had remarkably increased the NH_4-N and NO_3-N in soil. Increase in the N availability in soil due to spentwash application was reported by [9]-[11].

As expected the addition of inorganic N and P fertilizers had significantly improved the NH_4-N and NO_3-N concentration. Irrespective of methods of application, at all stages of crop growth, the amounts of both NH_4-N and NO_3-N were greater in soil that received spentwash at a rate of 120 m^3/ha plus recommended dose of NP fertilizers. The lowest concentration were registered in control followed by NP alone treatment (T_2). Incorporation of N through spentwash and inorganic N fertilizer (urea) added large amounts of N which have increased the NH_4-N and NO_3-N in soil. As the spentwash contains plant based proteinaceous substances the ammonification of organic N followed by nitrification of NH_4 might have occurred and increased the mineral N content in soil. The data on NH_4-N and NO_3-N have indicated that the spentwash not only adds N to the soil, but also enhances the mineralization of N from organic N pool in soil.

The spentwash used in the study contained about 420 mg N /L. At a rate of 40, 80 and 120 m^3 of spentwash, approximately 16.8, 33.6 and 50.4 kg of total N/ha, respectively, might have added to the soil. This corresponds to 25.8, 51.6 and 77.4 mg/kg, respectively. However, the results have shown that within 15 days of spentwash application, several fold increase in both NH_4-N and NO_3-N in soil. Such increase in the concentration of mineral N incorporated through spentwash application. It is likely that the spentwash application facilitated the mineralization of native (soil) organic N, which also increased the NH_4-N in soil. Whereas, microbial oxidation of NH_4-N, derived from spentwash as well as soil, had increased the NO_3-N in soil. The results from the incubation experiment also provide evidence for these findings.

The biological oxidation of NH_4 to NO_2 and NO_3 is believed to be carried out almost entirely by specialised bacteria, although oxidation by unspecialised bacteria has been observed in some environment. This may be one of the reasons for the reduction in NH_4-N at later stages. Besides, crop removal, part of NH_4-N is also adsorbed on clay particles, and/or lost through NH_3 volatilization. The high pH (> 8.2) and hot environment may have favored these processes in this soil under field condition. In control soil that has not received spentwash, the entire amount of NH_4-N was derived mostly from the decomposition of organic N which includes the soil humus N and recently added plant residues. Soil organic N decomposes relatively slowly. Under condition where organic N is accelerating or decreasing very slowly only a small portions of the total supply is converted into plant available N.

The net mineralization of N (N_m) for the spentwash application at the rate of 40 m^3 (T_3), 80 m^3 (T_5) and 120 m^3 (T_7) was calculated as in (1) [12].

$$N_m = \Delta Ni_{swt} - \Delta Ni_c \qquad (1)$$

$$\Delta Ni = Ni_{tx} - Ni_{to}$$

Where,

N_m = Net mineralization of N (mg N/kg soil)

Ni = inorganic N or mineral N

Ni_{swt} = inorganic N in spentwash treatment

Ni c = inorganic N in control treatment

t = time

The data are presented below

Net mineralization of N (mg N/kg soil) at different levels of spentwash application in soil

S. No.	Treatments	Before sowing		Pod formation stage		Post harvest stage	
		M_1	M_2	M_1	M_2	M_1	M_2
1.	Spentwash @ 40 m^3/ha	246	10.0	86.6	118.6	1.0	32.0
2.	Spentwash @ 80 m^3/ha	510	-2.0	176.0	212.0	53.6	77.0
3.	Spentwash @ 120 m^3 /ha	594	0.0	222.0	243.0	66.7	93.0

The net mineralization of N due to spentwash application was exorbitantly high and it was increased substantially with increasing the rate of spentwash application. The N_m during the crop growth for the three rate of spentwash application ranged from 1 to 594 mg of N/kg soil with one time application, and from -2 to 243 mg N/kg soil with continuous application of spentwash. With one time application, the net mineralization was very rapid initially and the rate of N_m was found decreased substantially during pod formation and harvest stages. Uptake of mineral N by groundnut, adsorption of NH_4 on soil exchange sites, volatilization of NH_3 and leaching of NO_3 could be attributed for the reduction in the amount of mineral N ($NH_4 + NO_3$ - N). Substantial amount of mineral N might have also been immobilized microbially in soil.

TABLE 2. EFFECT OF DIFFERENT LEVELS OF SPENTWASH APPLICATION ON NH_4-N (MG/KG) CONTENT OF SOIL

Treatments	Stage I			Stage II			Stage III		
	M_1	M_2	Mean	M_1	M_2	Mean	M_1	M_2	Mean
T_1- Control	72	76	74	39	46	43	33	32	33
T_2 - NP alone	82	80	81	55	62	59	48	41	45
T_3 - Spentwash @ 40 m^3 ha^{-1}	138	84	111	72	98	85	65	86	76
T_4- Spentwash @ 40 m^3 ha^{-1}+ NP	186	78	132	90	102	96	81	94	88
T_5- Spentwash @ 80 m^3 ha^{-1}	210	72	141	108	114	111	92	105	99
T_6- Spentwash @ 80 m^3 ha^{-1} + NP	228	76	152	114	129	122	108	115	112
T_7- Spentwash @ 120 m^3 ha^{-1}	252	78	165	112	120	116	98	112	105
T_8- Spentwash @ 120 m^3 ha^{-1}+ NP	282	72	177	120	136	128	114	118	116
Mean	181	77	129	89	101	95	80	88	84
	SEd		**CD (0.05)**	**SEd**		**CD (0.05)**	**SEd**		**CD (0.05)**
T	4.19		8.99	1.89		3.88	2.46		5.28
M	2.24		4.75	0.60		1.22	1.38		2.93
T x M	6.13		13.07	2.24		4.59	3.70		7.88
M x T	6.33		13.42	1.70		3.46	3.91		8.28

M_1 - One time application; M_2 - Continuous application Stage I – Before sowing; Stage II - Pod formation; Stage III - Post harvest

With continuous application of split doses of spentwash, the net mineralization was very slow initially probably due to relatively low input of N. The negative value indicates that immobilization (microbial and/or chemical) exceeded the mineralization, resulting in less net mineralization of N. However, the continuous application resulted in the build-up of N pool and facilitated higher rate of mineralization at pod formation stage. Crop removal and N losses (volatilization, leaching) further had decreased the mineral N concentration.

It could be attributed to microbial immobilization of NO_3 and NH_4, adsorption of NH_4^+ and leaching of NO_3. Both mineralization and immobilization of N are carried out by large numbers and many types of microorganisms. Hence the rate of mineral N production or depletion in soils is a net value of these two oppositive processes. Numerous experiments have shown that mineralization and immobilization occurs simultaneously in most soil. Furthermore, since the soil pH was > 8.2, it might also have favoured the volatilization loss of NH_3. According to [13], the microbial consumption of NO_3-N can be significant under high C availability and is strongly influenced by microbial substrate use efficiency. As the spentwash incorporates large amounts of easily oxidizable organic carbon, microbial immobilization of NO_3-N in the soil also could be responsible for the decrease in NO_3-N at pod formation and harvest stages.

It is also likely that some of the N appearing as NO_3-N was directly added as ammonium and nitrified (rather than mineralized from organic N) when conditions for microbial activity were appropriate for this transformation. In control plots lower soil NO_3-N concentration may be attributed to low availability of a substrate (organic matter) for N mineralization. A lag time of mineralization and nitrification rates is expected after application of spentwash [14].

The average annual rainfall at the field experimental site was 815.6 mm. The pattern of the monthly rainfall and predicted amount of net drainage for the experimental period are shown in Fig. 1. A total rainfall of 332 mm and a cumulative drainage of 460.4 mm were calculated from the sowing of groundnut (31.8.2007) to the harvesting (11.12.2007) which may have had a significant effect on leaching and the movement of nutrients in the soil profile. With crops grown during rainy season there is a possibility of leaching of large amount of NO_3 which could contaminate the groundwater. Thus, a high rate of net drainage predicted during the crop period may have resulted in significant leaching of mineral N. The rapid accumulation of NO_3 in plots receiving spentwash application implied the need for proper management of N to avoid environmental impact associated with N losses and leaching.

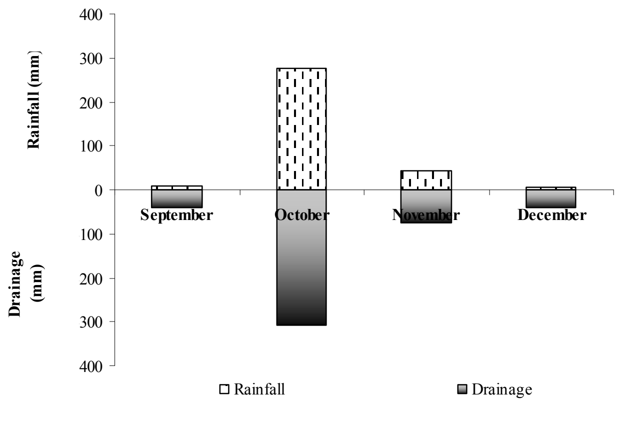

Fig.1. Monthly rainfall and predicted net drainage for the experimental period (Sep to Dec 2007)

TABLE 3. EFFECT OF DIFFERENT LEVELS OF SPENTWASH APPLICATION ON NO_3-N (MG/KG) CONTENT OF SOIL

Treatmets	Stage I	Stage II	Stage III

	M_1	M_2	Mean	M_1	M_2	Mean	M_1	M_2	Mean
T_1- Control	96	92	94	56	40	48	28	26	27
T_2 - NP alone	106	102	104	66	78	72	35	40	38
T_3 - Spentwash @ 40 m^3 ha^{-1}	276	94	185	118	124	121	38	49	44
T_4- Spentwash @ 40 m^3 ha^{-1}+ NP	330	90	210	134	166	150	49	63	56
T_5- Spentwash @ 80 m^3 ha^{-1}	468	94	281	172	202	187	65	75	70
T_6- Spentwash @ 80 m^3 ha^{-1} + NP	492	103	298	208	236	222	79	91	85
T_7- Spentwash @ 120 m^3 ha^{-1}	510	90	300	214	227	221	72	84	78
T_8- Spentwash @ 120 m^3 ha^{-1}+ NP	564	101	333	226	240	233	86	91	89
Mean	355	96	226	149	164	157	57	65	61
	SEd		**CD (0.05)**	**SEd**		**CD (0.05)**	**SEd**		**CD (0.05)**
T	8.21		17.60	4.54		9.73	1.77		3.80
M	4.28		9.08	2.65		5.62	0.99		2.11
T x M	11.86		25.29	6.98		14.87	2.66		5.67
M x T	12.12		25.68	7.50		15.91	2.81		5.96

M_1 - One time application ; M_2 - Continuous application Stage I - Before sowing ; Stage II- Pod formation Stage; III - Post harvest

It is also possible that an over estimation of NO_3 may also have occurred as a result of methodological biases associated with soil sampling in the field. Of particular concern is the large and variable NO_3 pools in these spentwash amended soil, and probable uneven distribution of microbial activities. It is likely that the 'hot-spots' of mineralization-nitrification may be formed due to uneven distribution of spentwash. These hot-spots would have had higher NO_3 concentration and more rapid nitrification than the rest of the soil.

It has been reported that organic manure with C/N ratio below 15:1 are more likely to give positive N mineralization after application in soil. In general a C/N ratio of 20:1 or narrower is assumed to have significant N to supply the decomposing microbes and release some N as well [15]. As has already been mentioned, the spentwash not only added nutrients and C, but also enhanced the microbial activity in soil which is evident from organic C content and enzyme activity. A strong correlation (R^2 = 0.78) observed between organic C and net mineralization of N (Fig. 2) demonstrates the role of C in controlling the N dynamics in soil [16].

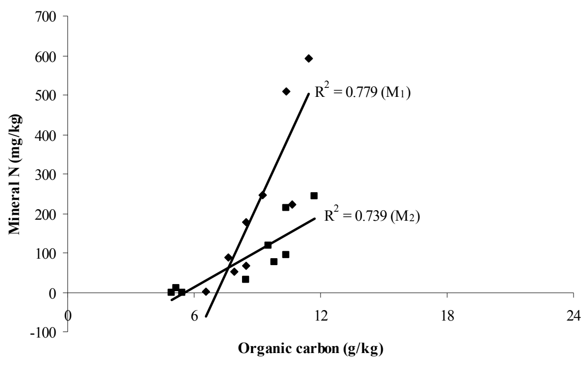

Fig. 2. Relationship between organic C and net mineralization of N

V. Conclusion

The results of the experiments have shown that the transformation and the plant availability of N in soil was greatly influenced by the spentwash application. The levels and methods of spentwash application differed significantly in affecting the N dynamics. Continuous application of split doses of spentwash at a rate of 120 m^3/ha with or without the recommended dose of NP fertilizers was found better in improving the nutrients status of soil. The spentwash not only adds nitrogen to soil, but also promotes the mineralization and/or solubilization of nitrogen in soil.

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