



Assessment of nitrogen utilization from urea applied in rubber seedling nursery using ^{15}N tracer technique

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Abstract

Nitrogen uptake from urea applied to rubber seedlings in the nursery was evaluated using ^{15}N tracer methodology, in micro-plots established in the field. Effect of varying rates of N on growth, and the relative contribution of soil N and fertilizer N towards total N uptake were studied. Influence of rate of N application on growth of seedlings was observed up to three months, and after six months, growth of plants was comparable for all the four N levels. The efficiency of N absorption by the plant, expressed as percentage N utilization ranged from 13.62 per cent at 125 kg N ha⁻¹ to 5.84 per cent at 500 kg N ha⁻¹. The ^{15}N balance sheet averaged over the four levels of N shows that about 9.27 per cent of applied N was taken up by rubber seedlings, about 8.78 per cent remained in 0-60 cm depth of soil and about 81.95 per cent was unaccounted N.

Keywords: Nitrogen uptake, ^{15}N labeled urea, N utilization, rubber seedlings

Introduction

Nitrogen application is essential in rubber seedling nurseries to generate healthy and vigorous seedlings. Current recommendation of N for rubber seedling nursery is application of 500 kg N ha⁻¹ in two equal split doses, and urea is the widely used source. Information on N use efficiency of applied urea by rubber seedlings is rather scarce.

Nitrogen fertilizer recommendations often diverge from the optimum because of the lack of information on the fate of applied N. There is growing concern about increasing nitrogen use in agricultural systems, due to reduced N use efficiency and increased risk of environmental pollution (Fang *et al.*, 2006). Efficiency of N use by most crops ranges from 20-60 per cent (Prasad Babu and Sarkar, 2002 ; Kundu *et al.*, 2000). Nitrogen recovery in crops rarely exceeds 50 per cent (Roberts, 2008). Nitrogen applied but not taken up by the plant is susceptible to loss through leaching, erosion, denitrification and volatilization. For improving N

fertilizer efficiency and for reducing N loss, it is imperative to obtain more information on the fate of N fertilizer applied to crops. Using the ^{15}N tracer methodology, it is possible to obtain precise information on the uptake of applied N by the plant, residual N in soil and the relative contribution of soil and fertilizer N towards total N uptake (Bremner, 1965; Buresh *et al.*, 1982). The use of ^{15}N permits the construction of a ^{15}N balance sheet, in order to determine the quantity of fertilizer N lost from the soil-plant system (Kundu *et al.*, 2000; Sarkar *et al.*, 1994).

The objective of the present study was to evaluate the uptake of N by rubber seedlings following the application of ^{15}N labeled urea at different levels under rainfed/irrigated field micro-plot condition, and to work out the N use efficiency. Effect of varying rates of N on growth of seedlings and the relative contribution of soil N and fertilizer N towards total N uptake were also studied. Attempts were also made to study the distribution of fertilizer N in different plant parts, fertilizer N remained in

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the soil after uprooting the plant and N loss from the soil-plant system. A ^{15}N balance sheet comprising plant N uptake, residual N in soil and N loss was attempted.

Materials and methods

An experiment was conducted at the Central Experiment Station of the Rubber Research Institute of India, during 2007-08. Initial soil samples at two depths, *viz.*, 0-30 and 30-60 cm were collected and analyzed for organic carbon, pH and available nutrients as per the procedure outlined by Jackson (1958). Germinated seeds were planted on 8'x4' beds, at a distance of 30x30 cm, in RBD with five replications. The treatments in the main plots included four levels of nitrogen, *viz.*, 125, 250, 375 and 500 kg ha⁻¹, and a no-N control. Micro-plots (60 cm x 60 cm x 60 cm) were established within the beds, by inserting GI sheets into the soil enclosing four plants, for the purpose of labeled urea application. ^{15}N labeled urea (2.9 atom% excess) at four levels, *viz.*, 125, 250, 375 and 500 kg N ha⁻¹ was applied inside the micro-plots, while rest of the bed was supplied with ordinary urea. Half dose of N was applied at six weeks after planting along with recommended doses of P, K and Mg. Remaining half dose of N was applied six weeks after the first application. The plants were irrigated alternate daily and maintained in the field for six months. Diameter and height of plants were recorded at three and five months after planting. Plants were uprooted and dry matter accumulation in different plant parts were estimated, separately for plants within the micro-plot and outside the micro-plot.

Leaf, stem and root samples of plants grown in the micro-plot were analyzed for ^{15}N atom% excess by mass spectrometry (Buresh *et al.*, 1982). Total N in plant samples were estimated by micro-Kjeldahl method (Bremner, 1965). Soil samples at two depths, *viz.*, 0-30 and 30-60 cm, collected from the micro-plot were also analyzed for residual ^{15}N in soil. Total N in soil samples were estimated by standard method outlined by Jackson (1958).

Per cent N derived from fertilizer (%Ndff) in leaf, stem, and root, and residual ^{15}N in soil were quantified, and total fertilizer ^{15}N uptake by the plant was calculated. Contribution of fertilizer N and soil

N towards total N uptake, and percentage N utilization were estimated. A ^{15}N balance sheet comprising fertilizer N uptake, residual ^{15}N in soil and N loss (^{15}N unaccounted) was prepared (Kundu *et al.*, 2000; Sarkar *et al.*, 1994).

Following equations were used for computing various parameters.

1. Atom% ^{15}N excess = Atom% ^{15}N – 0.3663
2. Percentage of N derived from fertilizer (%Ndff) = [Atom% ^{15}N excess (sample)]/[Atom% ^{15}N excess (fertilizer)] x 100
3. Percentage of N derived from soil (%Ndffs) = 100- %Ndff
4. ^{15}N uptake by plant = [%Ndff / 100] x total N uptake
5. Per cent N utilization = [Fertilizer N uptake] / [Rate of fertilizer application] x 100

Results and discussion

Initial soil properties of the experimental area is shown in Table 1. Soil was acidic and organic carbon content was in the medium range (0.75-1.5%). Available P and Mg were high and available K was in the low range.

Table 1. Initial soil properties of the experimental area

Soil depth (cm)	OC (%)	pH	Available nutrients (mg/kg)				Mechanical analysis (%)		
			Av. P	Av. K	Av. Ca	Av. Mg	Sand	Silt	Clay
0-30	1.50	5.27	39.0	44.0	85.9	39.0	50.9	9.7	36.8
30-60	1.24	5.14	19.7	21.0	72.3	19.9	47.9	10.1	39.3

Response to fertilizer N

Diameter, height and number of whorls of plants recorded at three and six months after planting are shown in Table 2. Diameter and height of plants were significantly higher for the higher rates of N application, *viz.*, 375 and 500 kg N ha⁻¹, while that at 125 and 250 kg N ha⁻¹ were comparable with control. Compared to control, number of whorls was significantly higher for all the N applied treatments. However, after six months, all the N applied treatments showed significantly higher diameter and number of whorls compared to control, and no significant difference was observed among the different N levels. Height of plants after six months did not vary significantly among treatments.

Table 2. Diameter, height and number of whorls of plants at three and six months growth

N rate (kg ha ⁻¹)	3 months			6 months		
	Diameter at 5 cm (mm)	Height (cm)	No. of whorls	Diameter at 10 cm (mm)	Height (cm)	No. of whorls
Control	5.18	55.22	2.96	5.57	81.89	3.93
125	5.15	56.38	3.23	6.29	83.16	4.11
250	5.18	53.70	3.22	6.39	82.43	4.14
375	5.44	60.28	3.23	6.68	90.20	4.44
500	5.60	60.08	3.40	6.65	87.23	4.49
SE	0.084	1.40	0.07	0.15	2.11	0.08
CD	0.25	4.21	0.25	0.46	NS	0.24

Dry matter accumulation after six months was significantly higher for the N applied treatments compared to control, but no significant increase was observed with increasing levels of N (Table 3). Total dry matter accumulation varied from 1529.3 kg ha⁻¹ for control to 2155.4 kg ha⁻¹ for 375 kg N ha⁻¹. Dry matter accumulation was highest in leaf, followed by stem and lowest in root. Buddability (%) also was significantly higher in the N applied plots compared to control, but did not increase significantly with increasing N rates (Table 3).

Table 3. Dry matter accumulation (kg ha⁻¹) and buddability percentage (Angular transformation)

N rate (kg ha ⁻¹)	Dry matter accumulation (kg ha ⁻¹)				Buddability (%)
	Leaf	Stem	Root	Total DM	
Control	657.8	601.9	357.5	1529.3	41.06 (64.6)*
125	862.6	780.7	418.6	2062.1	60.97 (74.6)
250	869.7	745.1	403.7	2020.2	61.14 (76.0)
375	974.4	774.2	407.6	2155.4	66.24 (82.6)
500	943.2	763.1	425.1	2131.5	65.51 (78.7)
SE	63.44	53.69	24.25	128.42	4.71
CD	190.1	NS	NS	384.87	14.12

(*Actual values are shown in paranthesis)

N content in leaves and roots were comparable for all the treatments. In the stem, it was significantly higher for the N applied treatments compared to control (Table 4). Total N uptake was significantly higher in N applied plots compared to control, and did not increase significantly with increasing rates of N application. About 70-75 per cent of the assimilated N was partitioned into the leaves followed by the stem (16-21 per cent) and then the roots (8-10 per cent).

Fertilizer N and soil N uptake

Total N uptake of plants grown inside the micro-plot did not show significant difference

Table 4. N content and N uptake in leaf, stem and root and total N uptake

N rate (kg ha ⁻¹)	N content (%)			N uptake (kg ha ⁻¹)			Total N uptake (kg ha ⁻¹)
	Leaf	Stem	Root	Leaf	Stem	Root	
Control	3.24	0.79	0.74	22.30	4.88	2.67	2.67
125	3.24	1.07	0.85	28.15	8.45	3.58	3.58
250	3.25	1.00	0.95	28.41	7.48	3.84	3.84
375	3.35	1.07	0.92	32.63	8.13	3.71	3.71
500	3.54	1.22	0.98	33.28	9.36	4.16	4.16
SE	0.11	0.07	0.06	2.6	0.78	0.33	0.33
CD	NS	0.21	NS	NS	2.28	0.98	0.98

Table 5. Nitrogen uptake in different plant parts and total N uptake (kg ha⁻¹) of plants grown in the micro-plot

N rate (kg ha ⁻¹)	Leaf	Stem	Root	Total N uptake
125	23.00	6.62	3.70	33.32
250	26.59	8.54	4.30	39.44
375	29.47	8.19	4.23	41.89
500	27.41	8.55	4.37	40.33
SE	2.57	0.92	0.41	3.37
CD	NS	NS	NS	NS

among treatments (Table 5). The results of the ¹⁵N analysis of plants grown inside the micro-plots showed that percent nitrogen derived from fertilizer (%N_{dff}) was significantly influenced by rate of N application (Table 6). %N_{dff} in leaf, stem and root samples increased significantly with increasing N rate up to 375 kg N ha⁻¹. %N_{dff} in leaf, stem and root at 500 kg N ha⁻¹ was comparable to that at 375 kg N ha⁻¹. %N_{dff} in leaf varied from 58.58 per cent at 125 kg N/ha to 81.66 per cent at 500 kg N ha⁻¹.

Fertilizer N uptake was lowest (17.02 kg ha⁻¹) at the lowest rate of N, viz., 125 kg N ha⁻¹, and this was comparable with 250 kg N ha⁻¹ level. Significantly higher fertilizer N uptake was observed at the higher rates of 375 and 500 kg N ha⁻¹ (29.96

Table 6. %N_{dff}, fertilizer ¹⁵N uptake, soil N uptake and %N utilization

N rate kg ha ⁻¹	%N _{dff}			Fertilizer N uptake (kg ha ⁻¹)				Soil N uptake (kg ha ⁻¹)	%N utilization
	Leaf	Stem	Root	Leaf	Stem	Root	Total		
125	58.58	48.34	44.24	13.44	3.92	1.61	17.02	16.31	13.62
250	71.42	55.23	54.13	18.69	4.90	2.38	24.08	15.36	9.63
375	80.82	70.49	63.67	23.80	5.67	2.73	29.96	11.92	7.99
500	81.66	69.71	64.30	22.47	6.02	2.80	29.05	11.28	5.84
SE	2.58	3.47	2.18	2.17	0.84	0.35	2.93	1.30	1.17
CD	7.96	10.69	6.70	6.58	2.66	0.98	8.97	3.90	3.44

Table 7. Residual fertilizer ¹⁵N in 0-30 and 30-60cm depth of soil

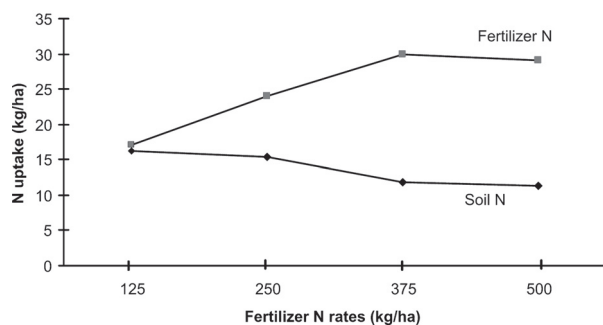
N rate (kg ha ⁻¹)	Soil depth			
	0-30 cm		30-60 cm	
	kg ha ⁻¹	%	kg ha ⁻¹	%
125	8.24	6.59	5.35	4.28
250	13.20	5.28	9.98	3.99
375	19.38	5.17	11.85	3.16
500	21.65	4.33	11.65	2.33
SE	2.42	0.757	1.55	0.681
CD	7.45	2.24	NS	NS

and 29.05 kg ha⁻¹, respectively), which was comparable with that at 250 kg N ha⁻¹ level (24.08 kg ha⁻¹). An earlier experiment to evaluate the efficacy of foliar urea spray in rubber seedling nurseries showed no response to N application above 250 kg N ha⁻¹, either as soil application or as foliar spray (Prasannakumari *et al.*, 2009).

Soil N uptake varied from 16.31 kg per ha⁻¹ at 125 kg N ha⁻¹ to 11.28 kg ha⁻¹ at 500 kg N ha⁻¹ level. Significantly higher soil N uptake was observed at the lowest N rate of 125 kg ha⁻¹ compared to the higher rates of 375 and 500 kg ha⁻¹, and this was comparable to that at 250 kg N ha⁻¹. Though increase in the rate of N application from 125 kg ha⁻¹ to 500 kg ha⁻¹ resulted in an increased fertilizer N uptake (from 17.02 kg ha⁻¹ to 29.05 kg ha⁻¹), the native soil N was found to contribute to as high as 48.9 per cent at 125 kg N ha⁻¹ and 27.97 per cent at 500 kg N ha⁻¹ towards the total N uptake of the rubber seedlings (Fig. 1).

Per cent N utilization

The efficiency of N absorption by the plant, expressed as per cent N utilization ranged from 13.62 per cent at 125 kg N ha⁻¹ to 5.84 per cent at 500 kg N ha⁻¹ (Table 6). Per cent N utilization at 125 kg

**Fig. 1. N uptake by rubber seedlings from fertilizer and soil at varying N rates**

N ha⁻¹ was significantly higher compared to all the higher N rates. Per cent N utilization at 500 kg N ha⁻¹ was significantly lower compared to 125 and 250 kg levels, and was on par with 375 kg level. With increase in N application from 125 to 500 kg ha⁻¹, N utilization decreased from 13.62 to 5.84 per cent of the applied N. Negative relationship between N utilization and N application rates was reported in burly tobacco by Sifola and Postiglione (2003). Santhi and Ponnuswami (1993) compared two levels of N fertilization (75 and 150 kg N ha⁻¹) in chewing tobacco and showed that N use efficiency decreased with increasing levels of applied N, and reached a maximum of 39 per cent for the 75 kg N ha⁻¹ level. Per cent N utilization by rubber seedlings observed in this study (13.62 to 5.84%) is quite low. Studies on N utilization of mustard from labeled urea showed substantially higher values of 45.7 per cent and 54.5 per cent, but at much lower N application rates of 40 and 80 kg N ha⁻¹ respectively (Prasad Babu and Sarkar, 2002). Nitrogen utilization of 30-50 per cent was reported for cereal crops like wheat, again at lower N application rates of 40-80 kg N ha⁻¹ (Kundu *et al.*, 2000).

Residual N in soil

The quantity of fertilizer ¹⁵N remained in 0-30 cm depth of soil ranged from 8.24 kg ha⁻¹ (6.59 % of applied N) at 125 kg N ha⁻¹ to 21.65 kg ha⁻¹ (4.33% of applied N) at 500 kg N ha⁻¹ (Table 7). Residual N in 0-30 cm soil was significantly higher at the higher N rates of 375 and 500 kg N ha⁻¹, compared to the lower rates of N application. Residual ¹⁵N in 30-60 cm depth of soil ranged from 5.35 kg ha⁻¹ (4.28%) at 125 kg ha⁻¹ level to 11.65 kg ha⁻¹ (2.33%) at 500 kg ha⁻¹ level, and did not vary significantly with varying N rates. The recovery of about 2-4 per cent of applied N from 30-60 cm depth of soil indicates that leaching loss was substantial under the present experimental condition of irrigation or high rain fall.

Table 8. Balance sheet of ¹⁵N at different rates of N application

N rate (kg ha ⁻¹)	Plant uptake (%)	Remaining in 0-60 cm soil (%)	Unaccounted N (%)
125	13.62	10.87	75.51
250	9.63	9.27	81.10
375	7.99	8.33	83.68
500	5.84	6.66	87.50
Mean	9.27	8.78	81.95

¹⁵N balance

The ¹⁵N balance sheet averaged over the four levels of N (Table 8) shows that about 9.27 per cent of applied N was taken up by rubber seedlings, about 8.78 per cent remained in 0-60 cm soil and about 81.95 per cent was unaccounted N. Considering the amount of unaccounted N as a measure of fertilizer N loss from the soil-plant system, it could be observed that fertilizer N loss increased from 75.51 per cent at 125 kg N ha⁻¹ to 87.5 per cent at 500 kg N ha⁻¹. Factors responsible for N loss that could explain the low N use efficiency include leaching, denitrification and ammonia volatilization (Kundu *et al.*, 2000; Sarkar *et al.*, 1994).

Conclusion

The study revealed that under field conditions, per cent N utilization from urea by rubber seedlings, averaged over the four levels of N, *viz.* 125, 250, 375 and 500 kg N ha⁻¹, was as low as 9.27 per cent. Influence of rate of N application on growth of seedlings was observed up to three months, and after six months, growth of plants was comparable for all the four N levels. Considering the amount of unaccounted N as a measure of fertilizer N loss, about 81.95 per cent of the applied N was lost from the soil-plant system. The study indicated the possibility of reducing the recommended dose of N for rubber seedling nurseries.

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