



## Nitrification Inhibition Studies of Neem Coating on Urea Prills†

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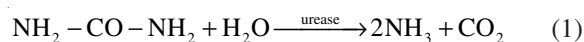
The Government of India introduced neem coated urea prills as an alternative to traditional urea prills with a view to improve the nitrogen usage efficiency by inhibiting nitrification. Neem was chosen because it contains *Azadirachtin*, which acts as a nitrification inhibitor in addition to its insecticidal properties. A laboratory experiment was set up for a study period of 18 days to compare the efficiency of neem coated urea as an effective nitrification inhibitor in comparison with normal urea. Results revealed that the neem coated urea was able to retard nitrate formation by 60 % at its maximum concentration and 52.5% on an average when compared with normal urea.

**Keywords:** Coating, Soil, Neem, Nitrification, Urea.

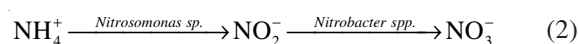
### INTRODUCTION

Crops require primary macronutrients such as nitrogen, phosphorous and potassium for its optimum growth. Roots cannot take the nitrogen in its direct elemental nature, hence it has to be converted to an inorganic form ( $\text{NH}_4^+$  or  $\text{NO}_3^-$ ) to be of any use [1,2].

Nitrogen is widely supplied to the soil in the form of urea. Urea contains about 46 % nitrogen which can be used up by the crops after conversion. It is hydrolyzed rapidly by ureases (produced by microorganisms) to form ammonium ( $\text{NH}_4^+$ ) as per eqn. 1 [3,4].



Ammonium is then oxidized to nitrites ( $\text{NO}_2^-$ ) and nitrates ( $\text{NO}_3^-$ ) in presence of nitrifying bacteria (*Nitrosomonas* spp., *nitrobacter* spp.) by the action of oxidoreductases and oxygenases [5,6] under appropriate conditions as per eqn. 2:



Nitrogen losses are in the form of ammonia volatilization, leaching, denitrification and surface run off [7]. Nitrates being negatively charged molecules, do not bind to the soil tightly and are prone to leaching losses. The excess nitrates leached into the soil can contaminate ground water below. Also, when nitrates undergo denitrification they give rise to nitrous oxide, a gas which has 300 times more greenhouse effect than  $\text{CO}_2$  [8,9].

Hence, it is essential to suppress nitrification.

One of the proposed ways to reduce and improve the usage is the use of nitrification inhibitors. These inhibitors slow down urea hydrolysis and the oxidation of ammonium to nitrates in the soil [10,11]. Even though these inhibitors are advantageous, globally, only two chemical nitrifications inhibitors are studied keenly; they are dicyanamide in Europe and nitrapyrin in United States of America. However, they are costlier in comparison with cheaply available fertilizers. Also, being chemical in nature they have potential to cause environmental hazards. Therefore, it is necessary to find an eco-friendly alternative nitrification inhibitor.

Neem (*Azadirachta indica*) is an ideal nitrification inhibitor and has an additional insecticidal property [12,13]. In order to increase yield and to reduce nitrate losses, the Government of India permitted fertilizer industries to coat neem oil on the entire quantity of urea production. It is claimed that the neem coated urea improves crop yield by 15-20 % [14]. In addition to improve the yield, neem coating of urea curbs the usage and illegal diversion of urea to other industries such as paints, dye and ply wood industries [15].

However, published literature on the effectiveness of neem coating on urea as a nitrification inhibitor is currently unavailable even though, it is established that neem helps in nitrification inhibition.

### EXPERIMENTAL

**Soil collection and characterization:** The soil required for experimental study was collected at a depth of one foot

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approximately from the agricultural field in Udupi district (South India). The location was at a latitude of 13°11'29.3"N. The collected soils were then mixed so as to maintain uniformity and were also made free from pebbles, plant materials and other unwanted materials by using a 4 mm sieve. This cleansed soil was then kept at 4 °C for 3 days before transferring to 22 °C [16]. Of the total quantity, a portion of it was separated and sieved through a 2 mm mesh for characterizing the collected soil. The various characterisation tests included cation exchange capacity, organic content in the soil, soil pH, specific gravity analysis and soil texture determination (Table-1). All the above tests were conducted according to the Indian Standards 2720.

TABLE-1  
CHARACTERIZATION OF EXPERIMENTAL SOIL

Parameter	Value	Ref.
Cation exchange capacity (meq/100 g)	8.6 (± 0.33)	[17]
Organic content soil (wt/wt %)	1.06 (± 0.17)	[18]
Soil pH	5.8 (± 0.11)	[19]
Specific gravity	2.67 (± 0.04)	[20]
Saturation water content (g moisture g <sup>-1</sup> of dry soil)	39 %	[21]
Soil texture	Loam, Sand 32 %,	[22]
	Clay 22 %,	
	Silt 46 %	

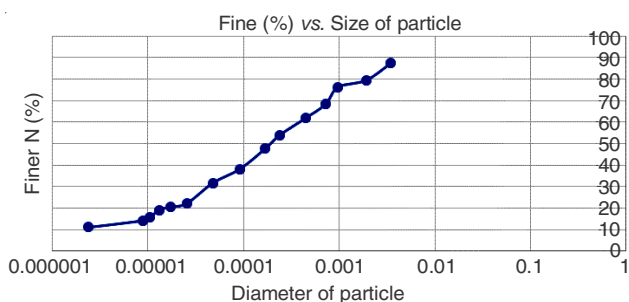


Fig. 1.

**Laboratory set up:** Two different but identical set ups were used, one as control containing uncoated urea in wet loam soil and another containing neem coated urea in wet loam soil. In order to generate the microbial activity, the saturated wet loam soil was incubated for a period of 10 days at 22 °C before treatment in both the identical setups. After 10 days of incubation, soil was removed from each of the two set ups. Urea N was added to both soil samples at approximately at 142 mg kg<sup>-1</sup> of soil assuming urea loading of 240 kg N hectare<sup>-1</sup> and a dispersion depth of 6 inches into the soil. Both the control and the test samples containing urea were thoroughly mixed for uniformity. The setups were immersed in a tray containing water and kept in an incubator at 27 °C. This was done to maintain air saturation and moist conditions inside the incubator and to prevent drying of the soil due to evaporation.

**Soil extraction and analysis:** Samples of about 3-5 g from the two set ups were analyzed at frequent intervals during the study period of 18 days and tested for nitrate and ammonium.

For ammonium analysis, random samples from the two set ups were taken and ammonium from the soil was leached out using 2 M KCl at a ratio of 1 g soil : 10 mL. The samples

were then subjected to shaking for 0.5 h and filtered using Whatman filter paper. Concentration of ammonium was determined by measuring the absorbance at 650 nm in a spectrophotometer as per the method prescribed by Baethgen and Alley [23]. Nitrate analysis was performed by spectrophotometric method using 0.01 M CuSO<sub>4</sub> and phenol disulfonic acid method by measuring absorbance at 415 nm [24]. All experiments were duplicated for checking the variations.

## RESULTS AND DISCUSSION

**Study of NH<sub>4</sub><sup>+</sup>-N:** At the start of the experiment, the concentration of NH<sub>4</sub><sup>+</sup>-N was minimal (3-6 mg kg<sup>-1</sup> of soil) in both the control and test. After urea hydrolysis was initiated, ammonium started accumulating. Hydrolysis is due to the extra cellular urease activity coupled with soil colloids and clayey particles that causes the urea hydrolysis to occur [25]. Several researchers have reported that urea hydrolysis follows first order kinetics [26,27]. This was clearly observed in Fig. 1, wherein a steep increase was noticed in first six days, after which there was a steady decrease in the concentration of NH<sub>4</sub><sup>+</sup>-N in both the samples probably due to nitrification. Thus, the concentration of NH<sub>4</sub><sup>+</sup>-N was dependent on the amount of urea present in the sample. As the urea-N depletes after the 6th day, the trend begins to descend. The maximum accumulation was observed on the 7th day for both uncoated urea (NU) and neem coated urea (NCU). The maximum concentration of NH<sub>4</sub><sup>+</sup>-N was found to be 136 mg kg<sup>-1</sup> (Fig. 2) and 109 mg kg<sup>-1</sup> for uncoated urea and neem coated urea, respectively. Neem oil probably enhanced hydrolytic activity of urease and of the 142 mg urea-N added, about 95% of the urea-N was converted to NH<sub>4</sub><sup>+</sup>-N.

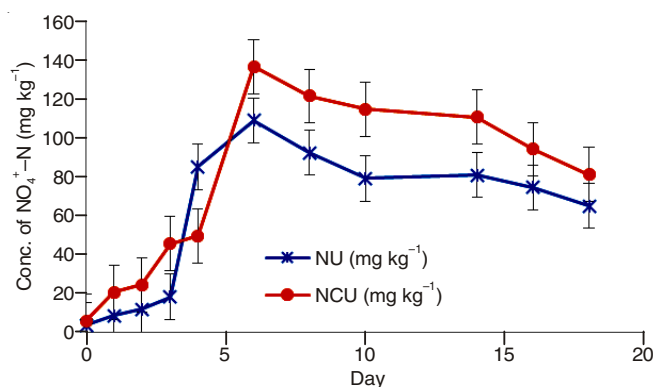


Fig. 2. Overall changes in concentration of NH<sub>4</sub><sup>+</sup>-N (mg kg<sup>-1</sup>) when uncoated urea prills and neem coated urea prills are used

**Study of NO<sub>3</sub><sup>-</sup>-N:** Similar results were obtained when nitrates were studied in both uncoated urea and neem coated urea variants as seen in Fig. 3. It was observed that the concentration of NO<sub>3</sub><sup>-</sup>-N rose from 1.345 mg kg<sup>-1</sup> for uncoated urea and 0.945 mg kg<sup>-1</sup> for neem coated urea. The maximum concentration for uncoated urea was 20 mg kg<sup>-1</sup> whereas it was 50.9 mg kg<sup>-1</sup> for neem coated urea. Thus, clearly at maximum concentrations, neem coated urea produced lesser nitrates. As can be observed from Fig. 3, nitrate production was always lower for neem coated urea. Thus, on an average over the study period of 18 days, neem coated urea was able to retard about 52.5% overall and about 60% at its maximum concentration when compared with uncoated urea.

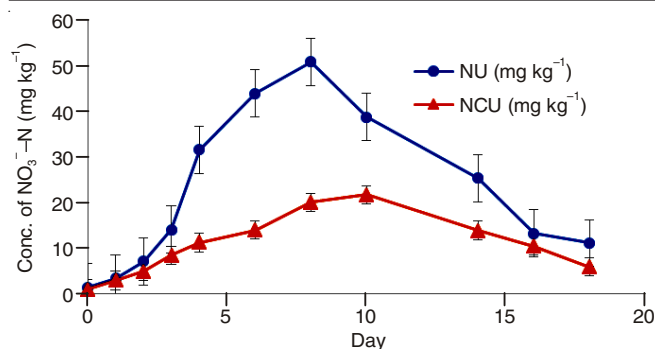


Fig. 3. Overall changes in concentration of NO<sub>3</sub><sup>-</sup>-N (mg kg<sup>-1</sup>) when uncoated urea prills and neem coated urea prills are used

**Time lag between nitrate and ammonia:** In Figs. 4 and 5, it is observed that the formation of nitrates is lagging in comparison with ammonium. In uncoated urea, the concentration of ammonium was found to be maximum on the 6th day (108.9 mg kg<sup>-1</sup>) and for nitrate on the 8th day (50.8 mg kg<sup>-1</sup>).

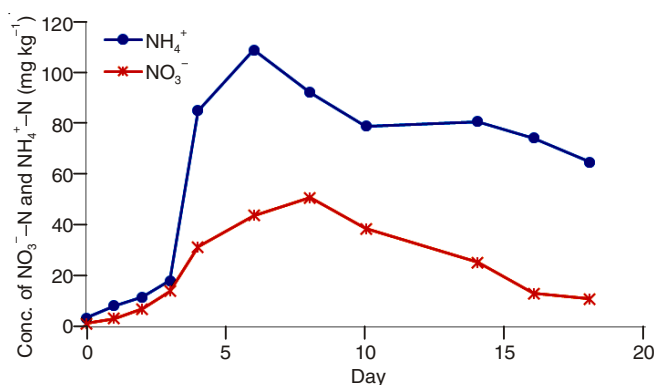


Fig. 4. Comparison of conc. of NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N (mg kg<sup>-1</sup>) when uncoated urea is used

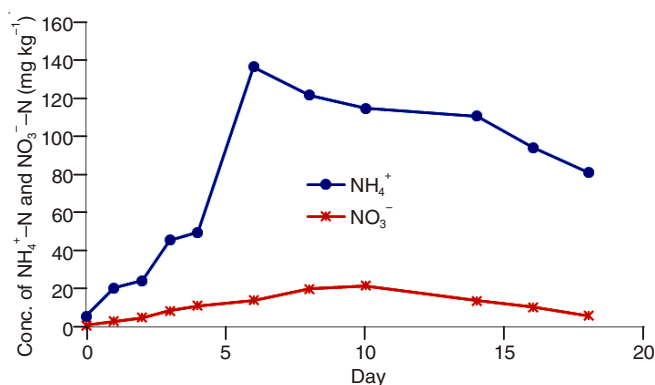


Fig. 5. Comparison of conc. of NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N (mg kg<sup>-1</sup>) when neem coated urea is used

Similar results were also observed in case of neem coated urea where ammonium maxima was found on the 6th day (136.47 mg kg<sup>-1</sup>), while nitrate maxima was observed on the 10th day (21.64 mg kg<sup>-1</sup>).

Hence, these observations are in accordance with the literature which states that nitrification occurs only under arable and favourable conditions and not immediately [28]. Also, as

evident from eqn. 2, nitrate production lags behind ammonium production

## Conclusion

The following conclusions can be drawn from the results obtained:

- Urease activity seems appears to enhance when neem is used as observed in Fig. 2 and in tandem with reported results [16,29].

- Nitrate concentrations obtained from neem coated urea was always less, hence neem coating retarded nitrification as already reported [12,13].

- Maximum nitrate concentration in neem coated urea was reduced by about 60 % in comparison with the maximum concentration of uncoated urea.

- Ammonium and nitrates were not accumulated simultaneously. The accumulation of nitrates was lagging behind ammonium.

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