

Fate of broadcast urea in a flooded soil when treated with N-(n-butyl) thiophosphoric triamide, a urease inhibitor

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Abstract. The compound N-(n-butyl) thiophosphoric triamide (NBPT) was found to be a more effective ureas inhibitor than phenyl phosphorodiamidate (PPDA) in flooded soils when compared at ureas of from 0.5 to 5% of the weight of urea. It allowed essentially no ammoniacal-N to accumulate in the floodwater when added at 0.5% of the weight of urea. The fate of urea was also determined in a flooded, unplanted soil with NBPT used as an inhibitor at a rate of 2% by weight of urea. At 41 days, fertilizer-N loss without the inhibitor was 73.4%, whereas with NBPT, 34.7% of the fertilizer was lost, presumably all by denitrification. With NBPT, urea hydrolysis was not inhibited below a 1 cm depth in the soil and most of the N (35.0%) accumulated as exchangeable NH_4^+ -N. Except for 15.0% of the fertilized accumulated as organic-N on the soil surface layer, immobilized N accounted for only an additional 7.0% in the soil at 22 days. Although the N saved from NH_3 volatilization loss obviously is eligible for denitrification losses, denitrification apparently was not enhanced to an appreciable extent by use of the inhibitor in that total losses were 15.7% at 22 days.

Introduction

Urea is a very important fertilizer in Asia [14], and its popularity has grown in recent years because of the low price compared to other N sources. It has been shown that, although ammonia volatilization losses from urea broadcast onto flooded rice are variable, they can account for up to 50% of the added fertilizer N [11]. These high losses occur when the fertilizer is top-dressed on a young crop or if it is poorly incorporated [11], practices which

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are very common in flooded rice culture. The high NH_3 volatilization losses are a result of rapid hydrolysis of the urea, which causes high aqueous NH_3 concentrations in the floodwater. The high pH conditions, due to urea hydrolysis and algal growth, sustain NH_3 volatilization [2]. A method proposed to reduce this loss in flooded soils is the use of urease inhibitors, which slow the conversion of urea to NH_3 and carbonate species and thus maintain the N as urea [21].

The urease enzyme, which is ubiquitous in surface soils, is extremely efficient at hydrolyzing urea. In a flooded soil, the biological activity generally is very high because sunlight, water, and nutrients are all present. The active biology and warm conditions of tropical rice paddies cause urea hydrolysis to be complete in 2-4 days [21] although it has taken up to 10 days in some studies for hydrolysis to be completed [17].

Urease inhibitors had not been extensively tested in plant studies until the discovery that phenyl phosphorodiamidate (PPDA) is a powerful urease inhibitor in soils [13, 15]. Although delayed hydrolysis was demonstrated for PPDA in flooded systems and nitrogen uptake generally increased by about 15%, few yield advantages were found [5, 8, 21]. The only exceptions occurred when the application of urea with PPDA was delayed and when wind speed was low at the time that inhibition ended and ammoniacal-N appeared in the floodwater [10, 17]. The degradation or basic hydrolysis of PPDA in the high-pH floodwater appears to explain why urease inhibition by PPDA is suddenly lost [1, 6].

Direct measurement of ammonia volatilization in conjunction with the ^{15}N balance technique has led some researchers to conclude that denitrification and NH_3 volatilization may be complementary loss mechanisms in rice paddies [11]. Obviously, N maintained from NH_3 volatilization and located near the oxidized zone of the soil is eligible for nitrification and subsequent denitrification loss; however, direct measurements of how important denitrification becomes when NH_3 volatilization losses are curtailed have not been possible because there have been no methods to preserve urea-N from NH_3 volatilization when at the soil surface.

The compound N-(δ -n-butyl) thiophosphoric triamide (NBPT) has recently been patented as a urease inhibitor [12]. It has been shown to be a powerful inhibitor in upland soil conditions [3], increasing N availability when conditions favored NH_3 volatilization losses from uninhibited urea [16].

These experiments were conducted to compare the urease inhibition by NBPT with that by PPDA in a flooded soil, to determine the effects of rates of inhibitor addition, and to determine the fate of the urea in an unplanted flooded soil when NBPT was used. In tracing the fate of the urea-N, the

purpose of the experiment was to determine the extent to which denitrification becomes a more important loss mechanism if ammonia volatilization is decreased while the N remains near the soil surface.

Materials and methods

Experiment 1

Three hundred grams (oven-dry basis) of Guthrie silty clay loam soil, a Typic Fragiaquult with a pH of 5.9 (soil:water 1:2) and 0.9% organic carbon, was placed into plastic containers of 10.8 cm diameter and 6 cm depth. The soils were flooded and puddled to provide a soil depth of about 3 cm and 2.5 cm of floodwater. The soils were then incubated in a greenhouse for 3 weeks. The urease inhibitors PPDA and NBPT¹ were pipetted as solutions at the rates of 0, 0.375, 0.75, 1.50, and 3.75 mg of the inhibitor to the floodwater of each pot. Then 75 mg of urea was pipetted as a solution to make the floodwaters approximately 15 mg L⁻¹ urea-N. Thus, inhibitor rates were 0, 0.5, 1, 2, and 5% of the weight of urea added. For reference, the 1% rate of the inhibitor is equivalent to 833 g/ha on the container area basis.

The floodwater pH was measured daily between 11:00 and 12:00; 3 mL aliquots of floodwater were taken daily, and the concentrations of ammoniacal-N and urea-N were determined by AutoAnalyzer [19, 20] for 2 weeks following fertilizer addition.

Experiment 2

Air-dried Guthrie soil was ground in a hammer mill to less than 1 mm size, and 200 g of air-dry soil was placed in the same type of plastic containers described above. The soil surface was levelled, a circular piece of fine nylon mesh cloth was placed over the soil, 50 g more soil was added, and another cloth was placed on the soil surface; then another 50 g of soil was added on top of the second cloth and levelled. The soil was slowly saturated with water to allow air to escape without disturbing the cloths and soil; then the soil was flooded. This system was used to simulate a flooded, puddled soil, with 2.2 cm of floodwater. The nylon cloth separated the soil into layers; the two

¹The PPDA was purchased from Parish Chemical Company, Orem, Utah; the NBPT was furnished by EniChem Corporation, Rome, Italy.

top layers were each 0.5 cm thick and the lowest layer was from 1 cm to 3 cm in depth. The cloth facilitated accurate sampling of the soil to these depths.

Higher rates of urea and NBPT were used in this experiment than in the first. A solution containing 3.6 mg NBPT was pipetted to each pot; then ^{15}N -labelled urea (4.7762 atom % excess) was pipetted into each pot at a rate of 0.179 g urea per pot. On an area basis, 93 kg N/ha was applied, which made the floodwater approximately 400 mg L^{-1} urea-N at the beginning of the experiment. The NBPT amount is equivalent to 2% of the weight of urea.

The soils were incubated in the greenhouse and then destructively sampled at 1.5 and 3 h and 1, 2, 4, 6, 13, 14, 22, and 41 days after fertilization. Pots not treated with NBPT were also incubated for 8 and 41 days. After each incubation period, the floodwater was siphoned from the soil surface, its volume was measured, and then the floodwater with algae was homogenized in a blender. Aliquots were removed for urea- and ammoniacal-N analyses and for a total-N digestion and ^{15}N analysis. The soil layers were removed at the cloth separations with a flat-ended spatula, the wet weight was determined, and samples were taken for exchangeable NH_4^+ -N, total-N, and dry-weight determination (dried at 105°C). Exchangeable NH_4^+ -N was extracted with 2 N KCl; the soil to KCl solution ratio was approximately 1:3. Aliquots were digested, distilled, titrated, and the ^{15}N ratio determined. The N analysis methods were as described by Buresh et al. [4] except that the total-N Kjeldahl digestions of the soil and floodwater were done by the block method of Stumpe et al. [18]. Extra pots, both with and without NBPT, were used to determine the ammoniacal- and urea-N concentrations in the floodwaters by AutoAnalyzer, but an ^{15}N balance was not determined on these pots.

In both studies, the greenhouse temperature was from $20\text{--}22^\circ\text{C}$ at night and $25\text{--}30^\circ\text{C}$ during the day. An oscillating fan was placed about 1.5 m from the pots to move the air gently around the pots to facilitate NH_3 volatilization losses. Air movement over the containers was approximately $0.4\text{--}0.8 \text{ m/s}$, depending on the distance from the fan. In both experiments, all treatments were in triplicate.

Results and discussion

Urease activity was very high in the biologically active flooded soil, and the urea was essentially gone from the floodwater by the third day after urea addition when an inhibitor was not used (Fig. 1). The average urea-N concentration immediately following addition was 205 mg L^{-1} ($\pm 5 \text{ mg L}^{-1}$);

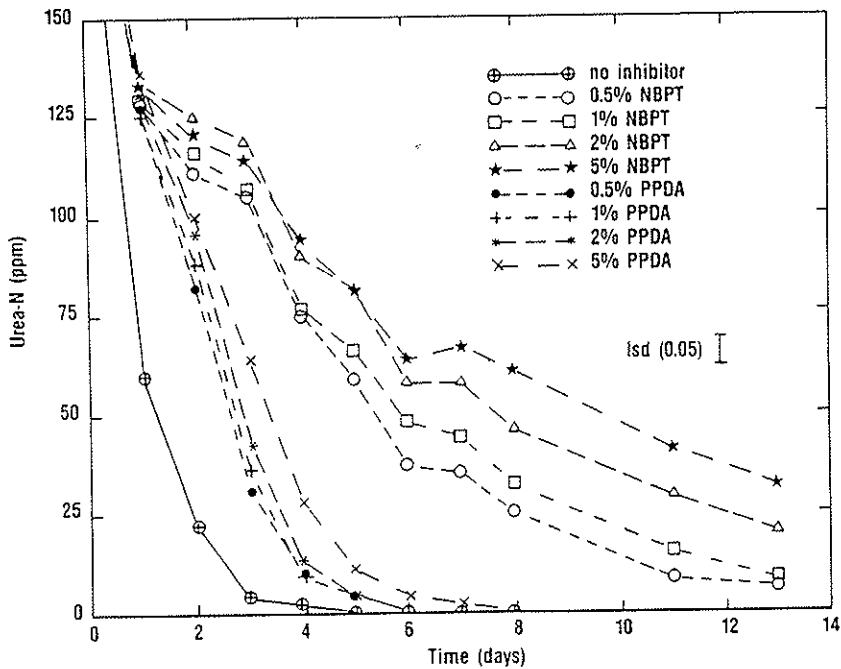


Fig. 1. Effect of inhibitor concentration on urea disappearance from the floodwater.

however, after 1 day only 61 mg L^{-1} urea-N remained in the uninhibited treatment, whereas all of the inhibited treatments had 125 to 130 mg L^{-1} . This rapid decrease of urea concentration in the inhibited treatments was due to adsorption, immobilization, and movement of urea into the soil as well as urea hydrolysis in that there was no ammoniacal-N in the floodwater for any of the inhibited treatments the first day. Without any inhibitor, peak ammoniacal-N concentration was $17 \text{ mg L}^{-1} \text{ NH}_4^+\text{-N}$ on the first day after urea addition. Ammoniacal-N was present in the PPDA treatments on the third day after addition at $5\text{--}10 \text{ mg L}^{-1} \text{ NH}_4^+\text{-N}$. Urea-N concentrations decreased rapidly after 1 day of inhibited urea hydrolysis with all of the PPDA treatments; there were only very slight differences between the 5%

DA and the lower PPDA concentrations. This lack of a prolonged inhibition by PPDA would be expected because of its rapid degradation at floodwater pH of 9.0–9.2 found during the daytime the first 2 days [6]. The hydrolysis of PPDA follows pseudo-first-order rate kinetics; therefore, there was not a large effect from the amount of PPDA applied.

The compound NBPT performed much better as a urease inhibitor than did PPDA at all rates of addition (Fig. 1), and differences in the plots of urea

disappearance with the different concentrations are apparent. Even though there were differences in the rate of urea-N disappearance from the floodwaters, essentially no NH_4^+ -N concentrations (less than 1 mg L^{-1}) developed in any of the NBPT treatments because of the slowed urea hydrolysis. Immobilization and adsorption processes were fast enough to prevent development of NH_4^+ -N concentrations in the floodwaters. The pattern of urea hydrolysis with NBPT was considerably different from that with PPDA. The PPDA completely lost its inhibitory effect, and urea hydrolysis then occurred at a rate that might be expected from an uninhibited system; NBPT, however, continued to inhibit at a particular level, depending on its addition rate, for up to 2 weeks.

The action of the 2% NBPT addition in the second experiment produced the same pattern of urea disappearance (Fig. 2) and no development of NH_4^+ -N concentrations, similar to that in Experiment 1. Because considerably more urea was added, initial concentrations were 400 mg L^{-1} urea-N, and it took about 5 days to hydrolyze practically all of the urea without inhibitor use. It took 41 days for the urea to completely disappear from the floodwater of the NBPT-inhibited treatments. With this high rate of urea

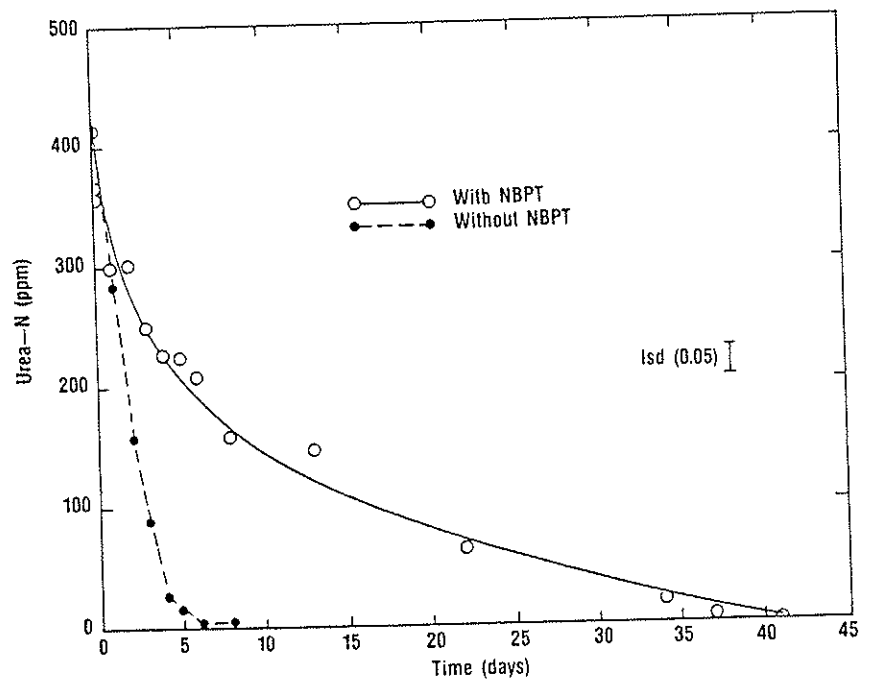


Fig. 2. Urea concentrations in the floodwater with and without NBPT.

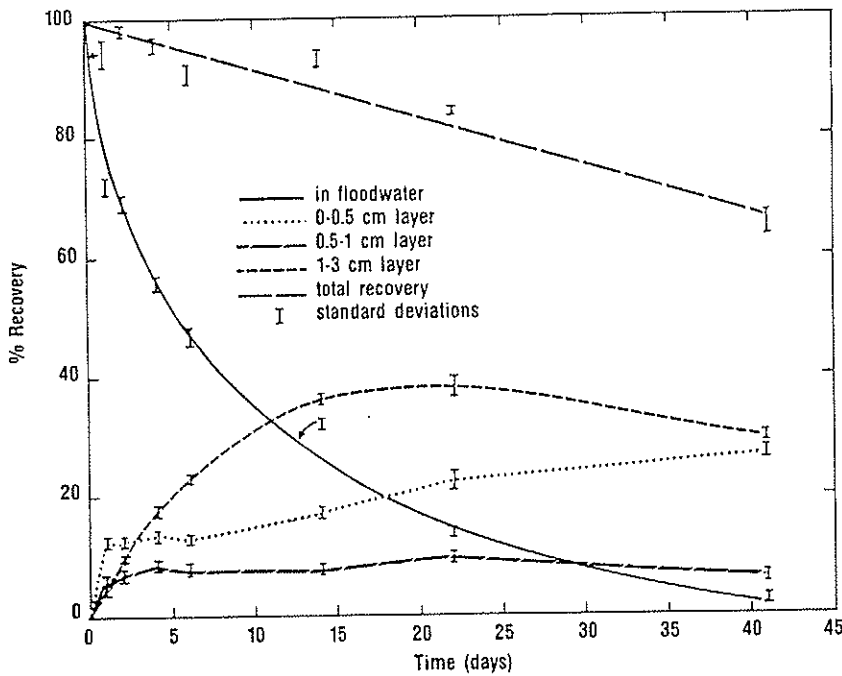


Fig. 3. Total ¹⁵N recoveries in the soil layers and floodwater, treatment with NBPT.

addition, ammoniacal-N concentrations peaked at $53 \text{ mg L}^{-1} \text{ NH}_4^+ \text{-N}$ on the third day without the inhibitor, whereas concentrations again remained very low (less than $2 \text{ mg L}^{-1} \text{ N}$) in the inhibited treatments. The sampling at 8 days with the uninhibited treatment was chosen because that was the day at which all the urea and most of the ammoniacal-N were gone from the floodwater. Without a plant to cause mass movement of water and applied urea into the soil, the urea in the floodwater of the NBPT treatment remained for 41 days, much longer than the 10 days found in pots planted with rice which received urea and NBPT at a 2% (w/w) basis [6].

was very little retention of ¹⁵N in the algae of the floodwater. As urea moved from the floodwater, it first resided in the 0–0.5 cm layer, and a relatively small and nearly constant amount accumulated in the 0.5–1 cm (Fig. 3). The major accumulation of the added N occurred in the soil layer below 1 cm. Total recovery in the uninhibited system at 8 days was 42.7% of the added urea-N because the large amount of ammoniacal-N production undoubtedly caused rapid and large NH_3 volatilization losses. At 41 days after the fertilizer addition, 65.3% of the fertilizer remained in the inhibited treatment, whereas only 27.0% remained in the uninhibited system.

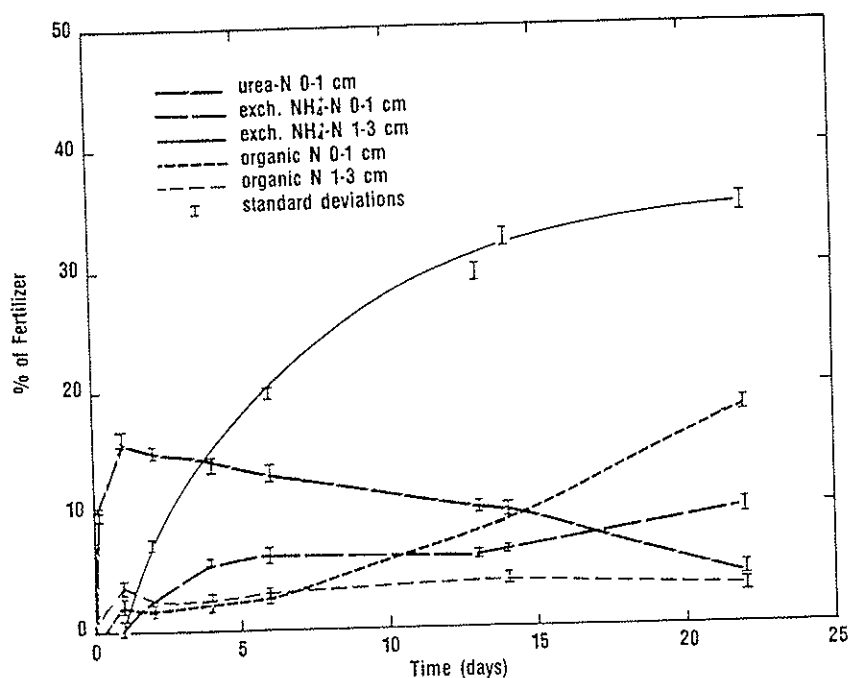


Fig. 4. Form of N recovered in the soil layers with time, treatment with NBPT.

The amount of fertilizer immobilized into the organic fraction was calculated by subtraction of the exchangeable NH_4^+ -N (^{15}N) and urea-N from the total ^{15}N recovery data for each soil layer (Fig. 4). Exchangeable NH_4^+ -N was not measured at 41 days, so that figure includes data only up to 22 days after fertilizer addition. The data for the top two 0.5 cm layers were combined to simplify presentation of the results. Urea entered the top 1 cm of soil very quickly, and its concentration then decreased with time. There was no urea found in the 1-3 cm layer at any time. Fertilizer-N in the lower soil layer was essentially exchangeable NH_4^+ -N except for 3 to 4% which was found as organic-N. The accumulation in the top 1 cm layer as exchangeable NH_4^+ -N was about 9.7% of the fertilizer. The accumulation at 22 days as exchangeable NH_4^+ -N in the lower soil layer was 35.0% of the added fertilizer.

The organic-N accumulation reached 15.0% in the 0-0.5 cm layer and only 3.2% in the 0.5-1 cm layer. This was apparently urea-N immobilized in algae growing or settled on the soil surface. To simplify presentation, the data for the two 0.5 cm layers are combined in Fig. 4. Urea in the floodwater is not shown in Fig. 4 because the recovery as urea was essentially the same as the total-N recovered in the floodwater (shown in Fig. 3).

Summary and conclusions

Even at the lower concentrations of the urease inhibitor NBPT, essentially no ammoniacal-N concentrations developed in the floodwater, which would indicate that ammonia volatilization losses were completely stopped. Its action produced a different pattern to that of PPDA, and inhibition was sustained for a long period at a particular level. Addition of higher amounts of PPDA is not a promising approach in the use of this inhibitor because the rapid degradation in the high-pH floodwater causes loss of inhibition. Other studies also showed little difference in the performance of PPDA used at different amounts although the stability of the inhibitor can be increased by lowering the floodwater pH [5, 6, 7].

Although denitrification has a greater opportunity to cause loss of N when N is preserved from NH_3 volatilization, apparent denitrification based on the ^{15}N balance caused much lower losses than those attributable to NH_3 volatilization in the uninhibited treatment. Most of the N preserved was not denitrified but was maintained in the soil. Thus, this study did not indicate that the losses by denitrification will negate gains in conservation from NH_3 volatilization as others have speculated [10]. It is quite possible that denitrification is constrained because of the lack of NH_4^+ -N as a result of deceleration in urea hydrolysis, thereby reducing denitrification.

The NBPT apparently did not inhibit urease activity below 1 cm in the soil in that the ^{15}N found in the 1-cm layer was NH_4^+ -N. The mobility of urea may be greater than the NBPT, or the soil contains too much urease for the NBPT to be effective at blocking hydrolysis. Thus, there was no indication that an effective urease inhibitor would decrease N uptake by rice plants by keeping the N in the form of urea. Unexpectedly, the N accumulated as exchangeable NH_4^+ -N in the lower soil layer rather than as organic-N; organic-N accumulated in an appreciable amount only at the soil surface. Conservation of the fertilizer-N as exchangeable NH_4^+ -N in the lowest layer indicates that urease inhibitors can aid in maintaining the N in a form readily utilized by rice.

The experiments demonstrate that the flooded soil system conserves N and that N cycles relatively slowly in the subsurface layers of soil. Plants would serve as a sink for N, and additional water movement would move N species into the soil and below the oxidized surface layer, and these factors would be expected to greatly reduce the amount of apparent denitrification losses found in this study [7, 9]. The rate and timing of plant uptake may have profound effects on the amount of N eligible for nitrification-denitrification. Further studies with NBPT and other effective urease inhibitors are needed to assess the benefits of urease inhibitors in planted rice paddies.

relative to other strategies to reduce ammonia volatilization, such as deep placement.

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