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Effect of Combined Application of Urease and Nitrification Inhibitors on Yield and Quality of Wheat

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Abstract: This study was to determine the effect of the combined application of a urease inhibitor (thiophosphric triamide, NBPT) and a nitrification inhibitor (dicyandiamide, DCD) on yield and quality of wheat. By applying labeled urea into an aquic brown soil, a pot experiment with spring wheat as test crop was conducted in the Shenyang Experimental Station of Ecology (CAS). The results showed that ^{15}N-U+NBPT and ^{15}N-U+DCD, the combination of urea with NBPT and DCD (^{15}N-U+NBPT+DCD) could retard the hydrolysis of urea, increase ^{15}N uptake by wheat, reduce the loss of urea-N in soil-wheat system; the yield of wheat in ^{15}N-U+NBPT+DCD treatment increased 27.77% and the content of protein increased 10.19% in comparison with CK.

Key words: urea-^{15}N, urease inhibitor, nitrification inhibitor, yield of spring wheat, grain quality

INTRODUCTION

At present, about 50% of the N fertilizer used in China is in the form of urea, which occupies 40% of the globe. However, recovery of applied N by crops is often as low as 30-40%[1]. Negative environmental effects such as NH_3 volatilization and N_2O emission are also related to the use of urea[2].

The recovery of urea-N in soil-plant systems is largely controlled by its transformation (hydrolysis, nitrification, immobilization and re-mineralization) in the soil[3]. Recently, inhibitors together with N fertilizer have been used to improve its efficiency and to decrease environmental nitrogen pollution[3-6] and ^{15}N has often been used to quantify the portion of applied N in various subsystems of farmland ecosystem, i.e. in soil, plant and atmosphere[7]. Although urease inhibitor N-(n-butyl) thiophosphoric triamide (NBPT)[8,9] and nitrification inhibitor dicyandiamide (DCD)[2,10,11] have been applied with success to retard the hydrolysis of urea and the nitrification of ammonium, less attention has been paid to the synergistic effect of urease inhibitor and nitrification inhibitor on soil applied urea-N[12].

The objective of this research was to determine the synergistic effect of urease inhibitor NBPT and nitrification inhibitor DCD on yield and quality of wheat in a pot experiment.

MATERIALS AND METHODS

Experimental design: A pot experiment with spring wheat (*Triticum aestivum* L.) was conducted at the Shenyang Experimental Station of Ecology (41°31′ N, 123°22′ E), Chinese Academy of Sciences, belong to the Chinese Ecosystem Research Network (CERN). The soil is classified as an aquic brown soil (silty loam Hapli-Udic Cambosols in Chinese Soil Taxonomy)[6].

Four different treatments were compared: ^{15}N-labelled urea (^{15}N-U) (10.25 atom % excess, supplied by the Shanghai Chemical Institute, China), ^{15}N-labelled urea + N-(n-butyl) thiophosphoric triamide (^{15}N-U+NBPT), ^{15}N-labelled urea + dicyandiamide (^{15}N-U+DCD), ^{15}N-labelled urea+N-(n-butyl) thiophosphoric triamide + dicyandiamide (^{15}N-U+NBPT+DCD). Each treatment had 3 replicates. Air-dried soil (6.0 kg) was thoroughly mixed with the corresponding amount of fertilizers and inhibitors and placed into each experimental pot. Fertilizers of P and K as K_2HPO_4 (1.0 g pot^{-1}) were applied as basal dressing. ^{15}N-labelled urea was applied at 2.0 g pot^{-1} and NBPT, DCD were used at 1%, 2% (w/w) of urea, respectively.

Twenty seeds of wheat were sowed in each pot on 1 April 2003. The spring wheat in all pots relied on watering and after seedling, 12 plants were chosen and left in each pot until the end of this experiment. Soil and plant samples were collected at maturity stage (July 1st, 2003) of spring wheat.

Measurements: Total N in plants and soil was determined by Kjeldahl digestion, followed by NaOH distillation and measured by titration with 25 mM H_2SO_4 in boric acid indicator[13]. Soil NH_4^+-N and $(NO_2^-+NO_3)^-$N concentration were extracted by shaking in 2 mol L^{-1} KCl for 1 hr, steam distillation and titration[14]. ^{15}N was determined by MAT-251 ultra-precision isotope mass spectrometers and

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urea-^{15}N concentration in soil and plant was calculated as: M = (α*ω)/100.

Where, M = concentration of all forms of ^{15}N (mg N pot^{-1}); α= ^{15}N atom % excess of each form of ^{15}N (%); ω = concentration of all forms of N (mg N pot^{-1}).

Statistical analysis: All statistical analyses were performed by SPSS software package and all the data obtained in the study were subjected to statistical analysis of variance (ANOVA). Difference at $p<0.05$ level was considered as statistically significant.

RESULTS AND DISCUSSION

Effect of urease and nitrification inhibitors on ^{15}N recovery and loss: At this experiment, 80.92% ^{15}N by plant and soil had been conserved in the treatment ^{15}N-U+NBPT+DCD, which was significantly higher than that recovered in the other three treatments ($p<0.05$) (Table 1). A combined application of NBPT and DCD facilitated the retention of applied urea-^{15}N in a soil-wheat system more efficiently, but applying NBPT or DCD alone had little effect on it. Compared to ^{15}N-U, other three treatments with inhibitors can increase ^{15}N recovery by soil and plant with 4.46-23.92% and ^{15}N-U+NBPT+DCD increased largest (23.92%). For all treatments, the ^{15}N loss in a soil-wheat system ranged 19.08-34.71% of applied urea-^{15}N (Table 1). A combined application of NBPT and DCD showed the lowest ^{15}N loss (19.08%) in this system ($p<0.05$), decreasing 45.03% as compared with ^{15}N-U. Xu *et al.*[7] found the synergistic effects of urease and nitrification inhibitors could retard the hydrolysis of soil applied urea, reduce N loss and provide more soil available N within a longer time, so as to promote the growth of wheat. Hence, compared with other two treatments with urea inhibitor NBPT or nitrification inhibitor DCD alone, a combined amendment of NBPT plus DCD had the most positive effects on improving ^{15}N recovery and decreasing ^{15}N loss of applied ^{15}N in a soil-wheat system.

Effect of urease and nitrification inhibitors on yield of spring wheat: In comparison with ^{15}N-U, ^{15}N-U+NBPT, ^{15}N-U+DCD and ^{15}N-U+NBPT+DCD had better biological characteristics of spring wheat, which significantly increase plant height ($p<0.05$) by 6.91-16.82%, increasing spike length, spike number and decreasing sterile number of spring wheat by 9.11-16.49, 11.75-28.61 and 5.26-10.53%, respectively (Table 2). Meanwhile, plant height, spike length and spike number were significantly higher ($p<0.05$) in ^{15}N-U+NBPT+DCD than in other three treatments. These observations showed that, during wheat growth period, the application of NBPT in combination with DCD may increase N uptake by spring wheat, benefiting a greater proportion of dry materials accumulated in vegetative growth period translating to the accumulation of materials in reproductive growth period, so as to decrease sterile number and to increase spike number of spring wheat.

Even so, the treatments with NBPT or DCD alone had a higher yield of spring wheat, but no significant difference was found compared to the control. The combined application of NBPT and DCD gave the highest yield ($p<0.05$), 27.78% over the control (Table 3). The probable reason was that this treatment could supply more mineral N for the growth of wheat at its later growth stages and improved 1000-kernel weight and the yield of spring wheat. The N fertilizer could promote the growth of roots, straws and leaves and tillers of wheat and increase green area, so as to promote the photosynthesis and the accumulation of nutrition of wheat. Otherwise, the yield and grain quality of wheat may be declined due to the lack of nitrogen nutrition. In this experiment, treatments with inhibitors, especially ^{15}N-U+NBPT+DCD, relatively satisfied the need of N of wheat at its later growth stages and improved the yield of wheat.

Effect of urease and nitrification inhibitors on grain quality: The grain contained a significantly higher

Table 1: ^{15}N recovery by soil and plant and ^{15}N loss in different treatments (% of applied ^{15}N)

Treatment	^{15}N recovery	Increase (%)	^{15}N loss	Decrease (%)
^{15}N-U	65.20±3.08 b[a)]		34.71±1.28 a	
^{15}N-U+NBPT	68.20±1.57 b	4.46	31.80±1.53 a	8.38
^{15}N-U+DCD	69.37±4.39 b	6.25	30.63±3.95 a	11.75
^{15}N-U+NBPT+DCD	80.92±5.46 a	23.94	19.08±3.74 b	45.03

a)Values showed in the table are means ± SD. Values followed by the same letter within a column are not significantly different at p=0.05, as determined by dunan's multiple range test. The same below

Table 2: Biological characteristics of spring wheat at its maturity stage in different treatments

Treatment	Plant height (cm)	Spike length (cm)	Spike number	Sterile number
^{15}N-U	35.73±0.85 c	5.82±0.31 b	21.31±0.60 b	1.9±0.21 a
^{15}N-U+NBPT	38.20±0.50 b	6.35±0.34 b	23.67±0.58 b	1.8±0.19 a
^{15}N-U+DCD	39.78±0.95 b	6.25±0.15 b	23.97±1.09 b	1.8±0.15 a
^{15}N-U+NBPT+DCD	41.74±1.25 a	6.78±0.69 a	27.31±1.25 a	1.7±0.23 a

Table 3: Effect of urease and nitrification inhibitors on yield of spring wheat (g pot^{-1})

Treatment	Yield	Increase (%)
^{15}N-U	8.10±0.27 b	
^{15}N-U+NBPT	8.45±0.70 b	4.32
^{15}N-U+DCD	8.43±0.79 b	4.07
^{15}N-U+NBPT+DCD	10.35±0.90 a	27.78

Table 4: Effect of urease and nitrification inhibitors on crude protein in grain of spring wheat (%)

Treatment	Crude protein	Increase (%)
^{15}N-U	16.32±0.48 b	
^{15}N-U+NBPT	16.78±0.73 b	7.28
^{15}N-U+DCD	16.34±0.46 b	0.12
^{15}N-U+NBPT+DCD	18.00±0.38 a	10.29

($p<0.05$) percentage of crude protein in ^{15}N-U+NBPT+DCD than in the other three treatments (Table 4). It was shown that this treatment did not only improve the urea N recovery but also the grain quality of wheat. The reason was probably that ^{15}N-U+NBPT+DCD provided more mineral N in soil for a longer time and it was well known from tracer studies that this allowed for a greater proportion of absorbed N to be translocated directly into grain[15,16]. As a result, this combination was beneficial to the improvement of urea-N efficiency and of wheat quality.

CONCLUSION

In comparison with ^{15}N-U, ^{15}N-U+NBPT and ^{15}N-U+DCD, ^{15}N-U+NBPT+DCD gave the best urea- ^{1}N recovery, yield and grain quality of spring wheat, as well as reducing urea-^{15}N losses, which will have some important agronomic and environmental meanings. In this experiment presented, all fertilizer and inhibitors were mixed thoroughly with soil, which may not be typical for field-grown wheat, unless applied before sowing. Therefore, further studies under field conditions are required to assess the *in situ* effects of these inhibitors.

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