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Improving the Effectivity of Urea Fertilizer in Shallot by Using Urease and Nitrification Inhibitors

(Perbaikan Efektivitas Pupuk Urea Melalui Penghambat Aktivitas Urease dan Nitrifikasi pada Tanaman Bawang Merah)

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ABSTRACT

Nitrification inhibitors are used to decrease the rate of nitrification process so it can decreases the nitrate losses. The objective of this study was to investigate the improvement of urea effectiveness by using urease and nitrification inhibitors on shallot. The study was conducted at Blubuk Village, Tanjung, Brebes District, Central Java, Indonesia from December 2017 to April 2018. The experiment was arranged in a randomized block design with 8 treatments and 3 replications. The treatments were untreated group (P0), 100% dose of Urea without inhibitor (control) (P1), (3) 100% dose of Urea + Urease Inhibitor (P2), 100% dose of Urea + Nitrification Inhibitor (P3), 100% dose of Urea + Urease Inhibitor + Nitrification Inhibitor (P4), 80% dose of Urea + Urease Inhibitor (P5), 80% dose of Urea + Nitrification Inhibitor (P6), and 80% dose of Urea + Urease Inhibitor + Nitrification Inhibitor (P7). The results showed that 100% dose of Urea + Urease inhibitor, 80% dose of Urea + Urease inhibitor, 80% dose of Urea + Nitrification inhibitor, and 80% dose of Urea + Urease inhibitor + Nitrification inhibitor treatments significantly produced higher plants heights, number of leaves, and more number of tillers compared to control treatment (100% Urea without inhibitors), without affecting the yield and yield components.

Keywords: alluvial, fertilizer, Nitrobacter, nitrogen, Nitrosomonas

ABSTRAK

Zat penghambat aktivitas nitrifikasi digunakan untuk menurunkan proses nitrifikasi sehingga dapat menurunkan kehilangan pupuk nitrogen. Tujuan penelitian ini adalah untuk mempelajari perbaikan efektivitas pupuk urea melalui zat penghambat aktivitas urease dan nitrifikasi pada tanaman bawang merah. Penelitian dilaksanakan di Desa Blubuk, Kecamatan Tanjung, Kabupaten Brebes, Jawa Tengah, Indonesia dari bulan Desember 2017–April 2018. Rancangan penelitian menggunakan metode Rancangan Acak kelompok dengan 3 ulangan. Penelitian terdiri atas 8 perlakuan, yaitu tanpa perlakuan (P0), 100% dosis urea tanpa zat penghambat (kontrol) (P1), 100% dosis urea + zat penghambat urease (PU) (P2), 100% dosis urea + zat penghambat nitrifikasi (PN) (P3), 100% dosis urea + PU + PN (P4), 80% dosis urea +PU (P5), 80% dosis urea +PN (P6), dan 80% dosis urea + PU + PN (P7). Hasil penelitian menunjukkan bahwa perlakuan 100% dosis urea + PU, 80% dosis urea + PU, 80% dosis urea+PN, 80% dosis urea + PU+ PN nyata menghasilkan tinggi tanaman, jumlah daun, dan jumlah anakan lebih banyak dibandingkan kontrol, namun tidak berbeda nyata pada peubah komponen hasil dan hasil bawang merah.

Kata kunci: aluvial, Nitrobacter, nitrogen, Nitrosomonas, pupuk

INTRODUCTION

Nitrogen can be absorbed by the plant by converting ammonium to nitrite by *Nitrosomonas* spp. and then the conversion of nitrite to nitrate by *Nitrobacter* spp. In moderate temperature and high soil water content, nitrification occurs on most soils within a few days or weeks after application of ammonium sources (urine, manures, compost, or urea fertilizer) (IPNI 2018).

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Nitrate is the usual form of N in soil that can be absorbed by plant. Nitrates are taken up by the roots and transferred to either water or the atmosphere. It can leach below the root zone. In addition, nitrate can be denitrified to form nitrous oxides and dinitrogen by the other soil bacteria, in waterlogged conditions. Nitrification is rapid in warm soils (>25°C), and it mostly ceases below 5°C. This process mostly happen in the soils that well aerated and near field capacity. Nitrification inhibitors are used to decrease the rate of nitrification process, so that decreases the nitrate losses (Nelson & Huber 2001).

Urease inhibitors are some compounds added to urea that can reduce the rate of the first "hydrolysis" step, and ammonia production. Urea can be formulated as dry granules, pills, or as a fluid alone or mixed with ammonium nitrate (UAN) (Mohanty *et al.* 2008). The disadvantage of all forms of urea is the increasing loss of ammonia gas if it is not incorporated into the soil soon after application (IPNI 2018).

Urea is converted to ammonium bicarbonate by the urease enzyme which occurs naturally during a few days after application. When urea is hydrolyzed by the urease, most of the ammonium is stored at the site of soil cation exchange, the pH rises briefly, and ammonia gas is produced. Urea inhibitors that are generally used to reduce the transient enzyme activity and slow down the rate at which urea is hydrolyzed (Du *et al.* 2012) are hydroximate (Mishra *et al.* 2002), fluoride (Prakash & Bhusha 1998), thiols (Todd & Hausinger 1989), hydroxyureas (Uesato *et al.* 2004), biscoumarin (Khan *et al.* 2004), and natural extracts (Juszkiewicz *et al.* 2004). The most widely used urease inhibitor is N- (n-Butyl) thiophosphoric triamide (NBTPT or NBPT), which is converted into active N-(n-Butyl) phosphate triamide (NBPTO or BNPO), phenylphosphorodiamidate (PPD/PPDA) and hydroquinone. Nitrification inhibitors slow down the process by inhibiting the activity of Nitrosomonas bacteria and increasing ammonium concentration. Urease and nitrification inhibitors are useful for reducing ammonia losses in urea fertilizer. They can control urea hydrolysis for 7 to 14 days, after rain, irrigation, or soil mixing. Urease and nitrification inhibitors are useful to keep the nitrogen fertilizer in the root zone, so the environmental condition is good. The good environmental conditions can increase plant growth and yield (Tustiyani *et al.* 2014; Kuswantoro 2016). The purpose of the research was to study the effect of urease and nitrification inhibitor on the effectiveness of nitrogen fertilization, growth, and yield of shallot.

MATERIALS AND METHODS

Study Area and Materials

This experiment was conducted at the Blubuk Village, Tanjung, Brebes District, Central Java, Indonesia with altitude 3 m above sea level, type of rainfall E, alluvial type soil, with pH 4.8–5.23 in December 2017 to April 2018. Soil analysis was carried out in the testing laboratory of the Agronomy and Horticulture Department, IPB University. The material used in this research was local variety of shallot seed (“Bima Kuning Brebes”), urease and nitrification inhibitors, Urea, Super Phosphate/SP-36 (36% P_2O_5), and Muriate of Potash/MOP (60% K_2O). The tools used to process data were computer and statistical analysis program (SAS).

Procedures

- **Plant preparation**

Soil tillage was done perfectly until the land was ready to be planted. After soil tillage, then the beds are made with a width of 1 m. The distance between the beds was about 50 cm. One unit of testing consisted of 5 beds each with a long of 5 m. Plant spacing used was 20 x 20 cm with 1 seedling/hole. Urease and nitrification inhibitors were applied at 10 DAP (Days After Planting), urea fertilizer was splitted in three times: 1/3 dose of urea fertilizer was applied at 10 DAP, 1/3 dose at 20 DAP, and the remainder at 30 DAP. SP-36 was applied 1 week before planting (300 kg/ha). MOP fertilizer at a dose of 200 kg/ha applied twice, 50% dose was applied at 10 DAP and the remainder at 30 DAP. Pest and disease controls were conducted according to the level of attacks with pesticides on a limited basis.

- **Observation**

Parameters observed in this study were (i) Plant height, the number of leaves, the number of tillers, were observed from 3–7 week after planting (WAP), (ii) soil analysis before and after treatment (N-total (Kjeldahl)), (iii) N-Content and N-Uptake (multiplication results from N-content and dry weight/plant) in biomass after harvesting, (iv) the number of bulb per plant, (v) the weight of 10 bulbs, (vi) the yield per plant (wet and dry), and (vii) the yield per plot and yield per hectare.

Data Analysis

The study was designed as a completely randomized block design. The treatments were untreated (P0), 100% dose of Urea without inhibitor (control) (P1), 100% dose of Urea + Urease Inhibitor (P2), 100% dose of Urea + Nitrification Inhibitor (P3), 100% dose of Urea + Urease Inhibitor + Nitrification Inhibitor (P4), 80% dose of Urea + Urease Inhibitor (P5), 80% dose of Urea + Nitrification Inhibitor (P6), 80% dose of Urea + Urease Inhibitor + Nitrification Inhibitor (P7). Each treatment was repeated 3 times. One experimental unit was 25 m^2. The data were analyzed statistically using F-test and Duncan's Multiple Range Test (DMRT) at 5% significant level.

RESULTS AND DISCUSSION

Shallot Plant Growth

In general, the use of urease and nitrification inhibitors did not increase the effectiveness of Urea at the beginning of the growth period (3 and 4 WAP) of shallot (Table 1). At 5 WAP, 100% dose of Urea + Urease Inhibitor, 80% Urea + Urease Inhibitor, 80% Urea + Nitrification inhibitor, and 80% Urea + Urease inhibitor + Nitrification inhibitors treatments actually produced higher plants than the dose of 100% Urea without inhibitors (control). Reduction of urea dose up to 20% in treatment of 80% urea + Urease Inhibitor, 80% Urea + Nitrification Inhibitor, and 80% Urea + Urease Inhibitor+ Nitrification Inhibitor could produce significantly higher plant heights than the dose of 100% Urea without inhibitors treatment at 5 WAP. It showed that the use of Urease and Nitrification inhibitors could increase the effectiveness of urea in 5–6 WAP. The inhibitory effect is usually between 25 to 55 days, and the effectiveness is low when it is leaching (IPNI 2018).

Table 1 The growth of shallot plant treated with urease and nitrification inhibitors

Treatment	3 WAP	4 WAP	5 WAP	6 WAP
		Plant height (cm)		
Untreated	18.1^{d}	22.7^{b}	26.7^{c}	31.3d
100% dose of Urea without inhibitor (control)	21.3^{ab}	26.9^{a}	**31.5^{b}**	36.5ab
100% dose of Urea + Urease Inhibitor	20.7^{abc}	26.2^{a}	**38.5^{a}**	35.2bc
100% dose of Urea + Nitrification Inhibitor	21.9^{a}	26.6^{a}	**31.3^{b}**	36.8ab
100% dose of Urea + Urease Inhibitor + Nitrification Inhibitor	21.3^{ab}	26.3^{a}	32.1^{b}	36.2ab
80% dose of Urea + Urease Inhibitor	20.3^{abc}	25.8^{a}	**38.7^{a}**	33.7c
80% dose of Urea + Nitrification Inhibitor	19.7^{c}	26.6^{a}	**39.2^{a}**	35.7bc
80% dose of Urea + Urease Inhibitor + Nitrification Inhibitor	20.1^{bc}	27.0^{a}	**39.0^{a}**	37.9a
		Number of leaves		
Untreated	8.5^{b}	13.3^{c}	18.0^{b}	17.9c
100% dose of Urea without inhibitor (control)	8.9^{ab}	13.9^{c}	18.1^{b}	18.3c
100% dose of Urea + Urease Inhibitor	8.9^{ab}	**17.2^{ab}**	**26.5^{a}**	**25.2ab**
100% dose of Urea + Nitrification Inhibitor	10.0^{ab}	14.5^{c}	18.5^{b}	19.2bc
100% dose of Urea + Urease Inhibitor + Nitrification Inhibitor	9.2^{ab}	13.5^{c}	17.7b	18.2c
80% dose of Urea + Urease Inhibitor	9.6^{ab}	**17.5^{ab}**	**28.3a**	**27.7a**
80% dose of Urea + Nitrification Inhibitor	10.5^{a}	**16.3^{b}**	**26.3a**	**25.3ab**
80% dose of Urea + Urease Inhibitor + Nitrification Inhibitor	10.3^{a}	**18.5^{a}**	**27.1a**	**24.7ab**
		Number of tillers		
Untreated	4.5^{a}	4.5^{bcd}	4.5b	4.6b
100% dose of Urea without inhibitor (control)	4.3^{a}	4.4^{cd}	4.5b	4.5b
100% dose of urea + Urease inhibitor	3.9^{a}	**5.2^{ab}**	**6.1a**	**6.1a**
100% dose of urea + Nitrification inhibitor	4.5^{a}	4.6^{bcd}	4.7b	4.7b
100% dose of urea + Urease inhibitor + Nitrification inhibitor	4.2^{a}	4.2^{d}	4.3b	4.3b
80% dose of urea + Urease inhibitor	4.1^{a}	**5.5^{a}**	**6.8a**	**6.8a**
80% dose of urea + Nitrification inhibitor	4.4^{a}	4.9^{abc}	**6.3a**	**6.3a**
80% dose of urea + Urease inhibitor + Nitrification inhibitor	4.3^{a}	**5.5^{a}**	**6.4a**	**6.5a**

Description: The numbers followed by the same letter in the same column are not significantly different according to DMRT on 5% level.

The effects of urease and nitrification inhibitors on the number of leaves and number of tillers on shallots were observed on 4 WAP (Table 1). 100% dose of urea + urease inhibitor, 80% dose of urea + urease inhibitor, 80% dose of urease + nitrification inhibitors, and 80% dose of Urea + Urease inhibitor + Nitrification inhibitor treatments consistently produced significantly higher number of leaves than the control treatment (100% dose of Urea without inhibitors) in 4-6 WAP. The effects of inhibitors of both urease inhibitor and nitrification inhibitor were observeds in the variable of number of tillers. Shallot treated with urea fertilizers added with urease inhibitor and nitrification inhibitor produced higher number of tillers than those treated with urea without inhibitors. Shallot treated with 100% dose of Urea + Urease inhibitor, 80% dose of Urea + Urease inhibitor, and 80% dose of Urea + Urease inhibitor + Nitrification inhibitor also produced significantly higher number of tillers than control treatment (without inhibitors) in 4‒6 WAP.

Shallot Yield and Yield Component

The yield components observed were the number and weight of shallot bulb (Table 2). The treatments of urease inhibitors and nitrification inhibitors did not affect the number and weight of shallot bulb. The highest number of shallot bulb (9 bulbs) produced by shallot treated with 100% dose of urea + Nitrification inhibitor. Shallot treated with 80% dose of urea + Urease inhibitor produced the least amount of shallot bulb, but produced the highest weight of shallot bulb compared to the other treatments.

Both treatments of urease and nitrification inhibitors did not effect on the yield of bulb/plants either wet bulb or dried bulb. The treatments with Urease and Nitrification inhibitors increased the yield/plant of shallot but the increase was not significantly different from the control. The yield per hectare showed that in general the application of urea fertilizer with urease and nitrification inhibitors did not affect the increase in shallot yield.

Soil Analysis

From the results of soil analysis, it was found that the treatment of urea with Urease and Nitrification inhibitors had no effect on N-Total. Treatment of urea fertilizers with urease and nitrification inhibitors was found to increase soil N-Total. In the untreated, group (P0), the soil N-Total after the trial was slightly decreased, while the soil N-Total in the treatment of fertilizer with or without inhibitors was found to increase. The percentage of total soil N after a trial in the treatment by the inhibitor was higher than the treatment without inhibitor (control). This result showed

Table 2 Yield components and yields of shallot treated with urease and nitrification inhibitors

Treatment	Number of Bulb/Plant	Weight of 10 Bulb (g)
Untreated	6.1a	40.3ab
100% dose of Urea without inhibitor (control)	8.5a	33.9b
100% dose of urea + Urease inhibitor	8.3a	36.0b
100% dose of urea + Nitrification inhibitor	8.8a	43.2ab
100% dose of urea + Urease inhibitor + Nitrification inhibitor	8.1a	39.2ab
80% dose of urea + Urease inhibitor	5.3a	**53.4a**
80% dose of urea + Nitrification inhibitor	5.9a	43.9ab
80% dose of urea + Urease inhibitor + Nitrification inhibitor	7.7a	42.2ab
	Yield/plant (g)	
	Fresh	Dry
Untreated	39.4a	20.7a
100% dose of Urea without inhibitor (control)	43.0a	23.2a
100% dose of urea + Urease inhibitor	44.2a	29.0a
100% dose of urea + Nitrification inhibitor	45.9a	27.3a
100% dose of urea + Urease inhibitor + Nitrification inhibitor	42.8a	20.6a
80% dose of urea + Urease inhibitor	39.3a	21.7a
80% dose of urea + Nitrification inhibitor	37.3a	17.7a
80% dose of urea + Urease inhibitor + Nitrification inhibitor	46.4a	24.7a
	Yield/25 m^2 (kg)	Yield/ha (kg/ha)
Untreated	17.0b	6800b
100% dose of Urea without inhibitor (control)	21.0a	8400a
100% dose of urea + Urease inhibitor	19.3ab	7733ab
100% dose of urea + Nitrification inhibitor	20.3a	8133a
100% dose of urea + Urease inhibitor + Nitrification inhibitor	19.7a	7867a
80% dose of urea + Urease inhibitor	19.0ab	7600ab
80% dose of urea + Nitrification inhibitor	19.7a	7867a
80% dose of urea + Urease inhibitor + Nitrification inhibitor	19.7a	7867a

Description: The numbers followed by the same letter in the same column are not significantly different according to DMRT on 5% level.

that N in the treatment given the inhibitor was still released after the shallot plant was harvested, so that N levels were still stored in plant tissue (Figure 1). Shallot is a short-lived plant and requires N during its lifetime, so the use of inhibitors is not effective. In the treatment without inhibitors (control), the total N content of the soil after the experiment was lower than the treatment with inhibitors. This shows that the total N in the treatment without inhibitors is lost from the soil because it is absorbed by plants, volatilization or dissolved in the flow of water (Figure 1). The efficiency of urea fertilizer is reduced by the presence of ammonia evaporation (Sangoi *et al*. 2016). To overcome these problems, plant residues are added to the soil in the no tillage system (Rojas *et al*. 2012).

Based on the results of the observation, the level of N in the treatment added with the inhibitor was higher than without the inhibitor (Table 3). Nitrogen in the treatment given the inhibitor was still not available to plants because it is still inhibited by urease and nitrification inhibitors. Urease and nitrification inhibitors cause the release of N available to plants to be slow. In addition, the age of shallot harvesting in this study was shorter (45 days) than the normal harvest age (60 days) due to the explosion of caterpillar pests, so that N added to the treatment given urease and nitrification inhibitors was not fully released (still in the form of urea). N-Uptake is the result of multiplication between N-content and dry yield/plants. N-Uptake for treatment with urease and /or nitrification inhibitors did not differ

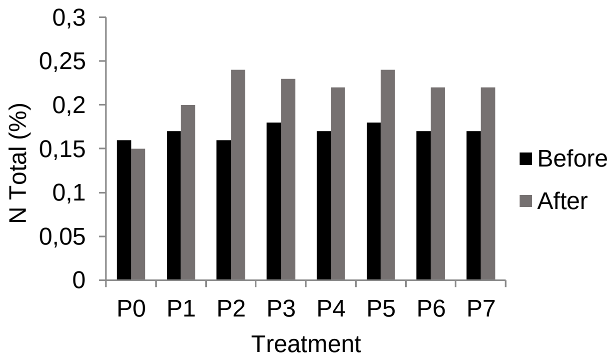

Figure 1 N-total analysis results before and after treatment on soil; P0 (no treatment); P1 (100% dose urea without treatment); P2 (100% dose urea + urease inhibitor); P3 (100% dose urea + nitrification inhibitor); P4 (100% dose urea + urease inhibitor + nitrification inhibitor); P5 (80% dose urea + urease inhibitor); P6 (80% dose urea + nitrification inhibitor); and P7 (80% dose urea + urease inhibitor + nitrification inhibitor).

Table 3 N contents and N uptakes in bulb after harvesting

Treatment	N-Content (%)	N Uptake (mg/plant)
Untreated	3.13a	659.94a
100% dose of Urea without inhibitor	3.14a	712.44a
100% dose of urea + Urease inhibitor	3.02a	862.08a
100% dose of urea + Nitrification inhibitor	2.95a	790.97a
100% dose of urea + Urease inhibitor + Nitrification inhibitor	2.83a	587.71a
80% dose of urea + Urease inhibitor	2.83a	613.09a
80% dose of urea + Nitrification inhibitor	2.91a	501.30a
80% dose of urea + Urease inhibitor + Nitrification inhibitor	2.72a	669.61a

significantly compared to the control treatment (100% dose of urea without inhibitors) (Table 3).

Discussion

In trial on the application of urease inhibitors and nitrification inhibitors on shallot plants it could be seen that there were effects of inhibitor of both urease inhibitors and nitrification inhibitor in the variables of plant height, number of leaves and tillers, but had not significant effect on yield, yield component, and nitrogen content of plants. Treatments with 100% dose of Urea + Urease inhibitor, 80% dose of Urea + Urease inhibitor, 80% dose of Urea + Nitrification inhibitor, and 80% dose of Urea + Urease inhibitor + Nitrification inhibitors significantly produced higher plant height, number of leaves, and higher number of tillers compared to control group (100% Urea rate without inhibitors). Some of the causes of the absence of inhibitor effects are due to the climate and soil types. According to Nelson & Huber (2001), nitrification inhibitors are not very effective on sandy soil. Experiments were carried out in Brebes that had alluvial soil (sandy clay). In addition, the effectiveness of inhibitors will decrease if rainfall is high due to the leaching effect. The experiment was carried out in December–April which took place during the rainy season, so the effectiveness of inhibitors was low.

Both urease inhibitor and nitrification inhibitor should inhibit the release of nitrogen or nitrogen-containing fertilizers faster so they can be taken by plants during their lifetime and reduce nitrogen loss after becoming nitrate. Mohanty *et al.* (2008) state that urease inhibitor can reduce the rate of the first “hydrolysis” step, the rate of ammonia production and ammonia loss to the atmosphere.

There were some alternatives to minimize nitrogen losses i.e., slow-release nitrogen fertilizers, nitrification inhibitor, urease inhibitor, and etc. Slow release nitrogen fertilizers are fertilizers that are covered by hydrophobic chemicals so that it can provide a physical barrier against water. Nitrification inhibitors can delay NH_4+ oxidation by nitrifying bacteria, preventing NO_3 formation and nitrogen leaching from the soil (Akiyama 2010). Urease inhibitors can delay urea hydrolysis, increasing the chances of urea incorporation in soil (Artola 2011). The urease inhibitor such as N-(n-butyl) thiophosphoric triamide (NBPT) added to urea was effective, easy method to reduce ammonium losses and increase the efficiency of N fertilization (Cantarella *et al*. 2008; Soares *et al*. 2012; Viero *et al*. 2015).

Plant nitrogen levels in plants treated with inhibitors were statistically not significant and the tended to be lower than treatments without inhibitors. It showed that the effect of inhibitors to inhibit nitrification activities so that nitrogen becomes available to plants. Unfortunately, the availability of nitrogen for plants was quite short and it did not affect the increase in shallot yields, because the 60 DAP should be harvested into 45 DAP. This young harvesting activity was due to the shallot caterpillar pest attack.

CONCLUSION

Based on research results showed that 100% dose of Urea + Urease inhibitor, 80% dose of Urea + Urease inhibitor, 80% dose of Urea + Nitrification inhibitor, and 80% dose of Urea + Urease inhibitor + Nitrification inhibitor treatments significantly produced more plant height, number of leaves, and more number of tillers compared to control treatment (100% Urea rate without inhibitors), but it did not significantly affect the yield and yield component.

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