

EFFECTS OF SULFUR AND UREASE COATED CONTROLLED RELEASE UREA ON DRY MATTER YIELD, N UPTAKE AND GRAIN QUALITY OF RICE

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ABSTRACT

The application of urea fertilizer has always been associated with heavy N losses therefore, improving its efficiency is critical to minimize economic and environmental losses associated with its application. One possible approach to improve the nitrogen losses from the surface applied urea is to coat it with sulfur, urease inhibitor (agrotain) and other biodegradable materials. A pot experiment was carried out using two different textured: silt loam and clay loam soil to assess the effect of sulfur and urease inhibitor (agrotain) on dry matter yield, N uptake and grain quality at the green house of the Zhejiang University, Hangzhou, China during 2011-12. The experiment consisted of treatments arranged in a completely randomized block design (CRD) with 3 replications. Urea coated with sulfur and urease inhibitor (agrotain) was applied at 60 kg N ha⁻¹ in 2 splits. The results showed that sulfur and urease inhibitor (agrotain) coated urea significantly increased the dry matter yield, N uptake and grain quality of rice cultivars over granular urea (GU) and control treatment applied split, particularly in silty loam soil than clay loam soil. The N release was more pronounced in silty loam (SL) soil as compared to clay loam (CL) soil where, the effect of the inhibitors was less evident. Japonica cultivar (xinshui 123) showed its superiority over indica cultivar (9311) in all the studied parameters. It is concluded from these results that controlled released urea fertilizer improved dry matter yield by 55-68% ha⁻¹, enhanced N uptake upto 39.4% and grain protein percentage from 5.8% -14.9% as compared to granular urea and control treatment. The improvement in rice dry matter yield and grain quality caused by sulfur and urease inhibitor (agrotain) coating may have been due to increased N use efficiency with split application in these two different textured soils.

Keywords: rice, sulfur, slow release urea, N uptake and grain quality.

INTRODUCTION

Poor nutrient utilization and nitrogen (N) losses from urea applications have been reported for many years (Khalil *et al.*, 2009). The N losses from applied urea were estimated at 30 to 60% in tropical soil (Khalil *et al.*, 2003; Zhang *et al.*, 2010). The main reason of these losses is the immediate increase in pH and NH₄⁺ concentration around the fertilizer micro site due to the activity of enzyme urease (Ahmed *et al.*, 2008; Mohsina *et al.*, 2004). When urea is applied to the soil, the soil pH and urea concentration increases immediately on the fertilizer microsite and urease enzymes start their activity to hydrolyse the applied urea (Krajewska, 2009). The rapid hydrolyses process of urea caused N losses and accumulation of NH₄⁺ which can be resulted in seedling damage and restricted germination of plant crops (Watson, 2000). Various approaches have been adopted to inhibit the urease activity and to delay the hydrolysis process of urea. The coating of urea with sulphur, urease inhibitors and other biodegradable materials are the possible remedies to reduce N loss and enhance urea efficiency (Shaviv, 2001). Urease inhibitors (agrotain) are compounds, which are used to inhibit the urease enzyme activity in soil and slow down the hydrolysis process of

urea in soil (Bolan *et al.*, 2004). Various studies have been done on urease inhibitors (agrotain) and polymer coatings to improve urea efficiency. A number of chemical compounds were tested as urease inhibitors but their toxic effects were poorly documented (Watson, 2000). The uses of these chemical compounds and synthetic coating materials have not practical applications due to the high cost, lack of availability and phototoxic effects (Purkayastha, 2009). The use of environmental friendly polymer coating and micronutrient as urease inhibitors can be beneficial as two in one. They can provisionally retard the urease activity in soil and have effect as an essential nutrient on plant and soil. These materials are easily available, biodegradable and low in cost. The use of these natural materials as the adhesive agent keeps together nitrogen and added micronutrient on the micro site. Therefore, the urease inhibitory effects and acidifying properties of these materials will control the increase in pH and urea concentration on the applied soil surface. These coatings have been tested previously in laboratory studies for reduction in ammonia volatilization loss. The outcomes indicated that use of sulfur and urease (agrotain) coated urea can reduce 50% of the ammonia volatilization losses from sandy loam acidic soils (Junejo, 2009a). Use of slow release nitrogenous fertilizers such as sulfur and agrotain coated urea is a potential solution

to this problem. Interest in slow release fertilizers is evident from several new products marketed in recent years. The potential benefits claimed are increased plant use efficiency by prolonged soil retention and less fertilizer applications, thus saving fertilizers and application costs (Leong, 2002). Numerous researchers have demonstrated the superiority of slow release N fertilizers in experiments with grain, forage and vegetables crops grown in greenhouses (Mohsina *et al.*, 2004). They concluded that single application of sulfur coated urea gave yield and seasonal distribution of growth similar to those obtained with multiple (3-5) application of urea. Supplying N in a slowly available form reduces volatilization, leaching or denitrification losses and the evidence suggests that sulfur and agrotain coated urea are effective for crops in irrigated coarse textured soil (Ahmad *et al.*, 2008). New slow release fertilizers, like sulfur and agrotain coated urea be fully evaluated under local conditions. Therefore, this greenhouse study was initiated to compare the response of rice cultivars by using sulfur and urease inhibitor (agrotain) coated urea on dry matter yield, N uptake and grain quality under different textured soils.

MATERIALS AND METHODS

The two soils (silt loam and clay loam) used in this greenhouse study were collected from Huajiang and Zijingang campuses of the Zhejiang University of China. The basic properties of the both soils were determined (Table 1). Soils were air dried and sieved through a 5 mm screen. Fertilizers were mixed with soil and then 6 kg of the mixture was added to the greenhouse pots. Pots were arranged in completely randomized block design (CRD) with three replications. The experiment were carried out under natural greenhouse environment. The sulfur coated urea used in this study was obtained from Shanghai Chemicals Product, P.R.China. Sulfur Coated Urea was produced by coating urea granules with sulfur, then coated with a microcrystalline wax sealant containing a microbicide and finally with a clay conditioner, whereas AgrotainU fertilizer refers to urea granules treated with only N-(n-butyl) thiophosphorictriamide (NBPT or Agrotain). Agrotain (N-(n-butyl)-thiophosphorictriamide (NBPT) is a urease inhibitor, and is used to retard hydrolysis of urea which is catalyzed by the urease enzyme. The net beneficial effect is to reduce the loss of ammonia by volatilization for up to 14 days when used in surface applications. The urease (agrotain) coated urea was obtained from the department of Agronomy and Plant Breeding and Genetics, Zhejiang University, Hangzhou, P.R. China. The sulfur and urease (agrotain) coated urea was compared with pre plant and split application of urea (Table 2). NPK at the rate of 60 kg ha⁻¹ were added to all pots 24 hours before according to the recommended dose for rice in China. Urea, Triple super

phosphate (TSP) and KCl were used as sources of N, P and K, respectively. The initial fertilizer application was uniformly mixed with the entire soil prior to seedlings transformation and the second half portion of uncoated urea, sulfur and urease (agrotain) coated urea was applied at panicle formation stage. Seven treatments *viz.*, (N₀ = Control) (N₁=urea alone) (N₂= urease (agrotain) coated urea) (N₃= Sulfur coated urea) (N₄= Urea, Split) (N₅= urease (agrotain) coated Urea, Split) (N₆= Sulfur coated Urea, Split) as shown in (Table 2). Two contrasting rice cultivars of Japonica (xinshui 123) and indica (9311) were chosen because of their contrasting agronomic characteristics. Japonica (xinshui 123) is short and late maturing cultivar whereas indica (9311) is tall and early maturing cultivar. Seed of both the cultivars were sown one month before for transplantation purpose. Four (4) healthy seedlings hill⁻¹ of 26 days old were transplanted into each pot. Tap water (1100 ppm total salts) was used for irrigating the pots and the soil moisture was maintained at about 60-70% of the soil water holding capacity by measuring the volume of water added. To induce leaching and simulate approximate field conditions, pots were occasionally irrigated with excess water. Metrological data for the rice growing period are given in (Table 3). The foliar portions of the two representative plants were harvested after 115 days and then washed thoroughly with dilute acid and de-ionized water. All plants parts were placed in paper bags, dried in a forced oven at 70°C and weighted after 72 hours. The dry plant material was ground and nitrogen was determined on a Kjeld-Foss automatic nitrogen analyzer, model 16210. Nitrogen use efficiency (NUE) was calculated by subtracting N removal in the check plot from N removal in the treatment and then dividing by applied N (60 kg ha⁻¹). The fertilizer N in the soil and plants, uptake efficiency were calculated by the following formula:

$$F_{\text{plant/soil}} = W \times a \times (b - c) / (b1 - c)$$

in which $F_{\text{plant/soil}}$ is N from the fertilizer in the plant or soil.

W: Weight of plant or soil sample (g)

a: N content in the plant or soil sample (mg/kg)

b: ¹⁵N atom (%)

c: ¹⁵N atom in the CK treatment (%)

b1: ¹⁵N atom in fertilizers

N-uptake efficiency (%) = $F_{\text{plant}} / \text{Fertilizer N applied} \times 100$

Determination of rice quality: Rice quality was determined by the Rice Product Quality Supervision and Inspection Center, Ministry of Agriculture, Hangzhou, China. The protein content in kernels was determined by the micro-Kjeldahl method by the following formula: Protein content = (6.25 × nitrogen content/dry matter weight) × 100%.

Statistical Analysis: Data were statistically analyzed according to completely randomized design. Least significant difference (LSD) at 5 % probability level was employed upon obtaining significant F-values between treatment means.

RESULTS AND DISCUSSION

Dry Matter Yield: Responses to single and split application of granular, sulfur and urease (agrotain) coated urea fertilizers are presented in Fig.1. Significantly higher dry matter yields took place with sulfur and urease (agrotain) coated urea in both cultivars grown in the two textured soils. Split application of sulfur and urease (agrotain) coated urea and granular urea (GU) gave maximum dry matter yield over single applications. However, the differences among split applications of the two coated urea at the two application time were significant. Control and pre-plant application of urea @60 kg N ha⁻¹ yielded the lowest dry matter production. Data in (Fig.1) suggested that consistent difference occurred among coated and granular (uncoated) urea. The crop response was measured in terms of dry matter yield and the plant N uptake which were basically dependent on level of N available to the plants keeping all other parameters similar. It is generally accepted that the fate of N fertilizers added to soil is the NO₃ form. Therefore, when water is added to the soil it dissolves the nitrate (NO₃) and moved it down to the bottom of the wetting front. The pots were over irrigated on every third irrigation and a good deal of the readily soluble (NO₃) from urea was expected to be leached down and lost probably before the plants were established. The measurements of the leachate or its N content were not done in this study. Superiority of the sulfur and urease (agrotain) coated urea over granular urea (GU) alone under our conditions are consistent with reports by other workers under different conditions (Villar and Guillaumes, 2010). Sulfur and urease (agrotain) coated urea gave the highest dry matter yield in both rice cultivars grown in both soils. Japonica cultivar (xinshui 123) performed better than Indica cultivar (9311) in terms of dry matter accumulation. This could be related to the releasing characteristics of the sulfur and urease coated urea fertilizers used. Release of N from sulfur and agrotain coated urea is controlled by the thickness of coating, microbicide and time of contact with the soil (Hayat and Khan, 2013). Since in the first 14 days period about 30-40% of the N would have been released from sulfur and agrotain coated urea respectively. It is speculated that newly established plants under these treatments probably did not make much use of the release N. Further more, induced leaching in the pots might have contributed to more losses from sulfur and agrotain coated urea because of its lower rate of N release in the first 7 days, sulfur coated urea apparently resulted in a

better N utilization and higher dry matter yield (Iqbal *et al.*, 2013).

Nitrogen Uptake: Nitrogen uptake by rice cultivars in silty loam and clay loam soils are presented in Table 4. It was observed that sulfur and urease (agrotain) coated urea, as slow release fertilizers significantly enhanced the N uptake by rice plants as compared to granular urea (GU) and control pots. The dissolution rate of urea alone was faster as compared to sulfur and urease (agrotain) coated urea and its effect on N uptake by rice cultivars in both soils. Maximum N uptake was observed in sulfur and urease (agrotain) coated urea in both rice cultivars grown in both soil. Split application of urea at the rate of 300 kg N ha⁻¹ resulted in a higher N uptake by Japonica rice cultivar (xinshui 123) grown on silt loam soil as compared to indica cultivar (9311) of rice grown in clay loam soil. This suggested that the commonly used practice of applying/adding urea at different growth stages appeared to be a sound and effective methods of fertilizer application. The poor N uptake associated with urea alone treatments could be due to the large volatilization and leaching alkaline calcareous soils (Nasima *et al.*, 2010). The comparison of N uptake by both rice cultivars growing in the two soils is of particular interest since all fertilizer treatments used resulted significantly higher N uptake in the silt loam soil over the clay loam soil (2 to 5 times). It showed that plants roots were more developed in this light textured soil and thus enhancing N utilization. This lighter soil also contained less CaCO₃ (14.1 %) than the other soil (16.1%), a property which is more conducive to losses of applied N through ammonia volatilization. A soil which is alkaline in pH becomes temporarily more alkaline after the addition of urea and others ammonium carrier through hydrolyses and reaction with CaCO₃ and that leads to more volatilization and losses of nitrogen (Junejo *et al.*, 2009b). High losses of ammonia through volatilization from soils of low exchange capacity, high lime content, alkaline pH and high temperature have been reported by many researchers (Reddy and Sharma, 2000). Leaching of nitrate in coarse textured soils is an established fact. As mentioned earlier, the induced leaching in this present study might have contributed to considerable losses of applied N from soluble N fertilizers. Sulfur and urease (agrotain) coated urea fertilizers have slower dissolution rate and thus slower nitrification making less susceptible to NO₃ leaching. These results are in line with the findings of Hayat and Khan, (2013) they have reported the effectiveness of super and urease inhibitor (agrotain) coated urea over granular urea in coarse textured soils under leaching conditions.

Grain Protein: Data regarding the impact of single and split application of granular, sulfur and urease inhibitor (agrotain) coated urea fertilizers on grain protein (%) are presented in Fig. 2. The grain contained a significantly

higher percentage of protein in sulfur and agrotain coated urea as compared to granular urea and control treatments. It was observed that these treatments did not only improve the urea N recovery but also the grain quality of rice. The reason was probably coating of urea with sulfur and urease inhibitor (agrotain) provided more mineral N in soil for a longer time. It is well known from tracer studies that this allowed for a greater proportion of absorbed N to be translocated into grain. As a result, urea coating with sulfur and urease inhibitor (agrotain) was beneficial for the improvement of urea N efficiency and rice quality. Split application of controlled release urea fertilizers gave maximum protein (%) as compared to non coated urea and control treatment over single or pre-plant application. The differences among split applications of the two coated urea at the two application time were significant. Japonica cultivar gave more protein (%) than indica cultivar in both textured soils and the amount of increase was considerably higher in japonica cultivar (xinshi 123) than indica cultivar (9311). Silt loamy soil performed better than clay loam soil with respect to protein content in both cultivars, indicating that more soil-derived N was absorbed by rice in this soil (Fig. 3). In both soils the presence of sulfur and urease inhibitor (agrotain) caused a significant decrease in fertilizer-derived N recovered, compared to granular urea alone (GU) and control treated pots. On the contrary, an

increase in the fertilizer-derived N taken up by plants in the presence of both urease and nitrification inhibitors has been found by Xu *et al.*, (2000). Split application of urea and coated fertilizers was more advantageous than single or pre-plant application. Delaying urea hydrolysis by urease inhibitor (agrotain) and other biodegradable material increases the chances of availability of the applied urea to move from surface soil into sub-surface soil layers by irrigation/rain, where volatilization cannot occur. Generally plants take up the majority of their N in nitrate (NO_3^-) form as enforced on them by the ubiquitous urease enzymes and nitrifying bacteria. But plants extra energy to convert (NO_3^-) to NH_4^+ form via nitrate reductase followed by amide and protein conversion, each of these steps occurs at the expense of energy. However urea, an un-charged particle can be taken up by plant roots as an intact molecule relatively faster as well as without releasing any charge (H^+ or OH^-) into the root rhizosphere and can thus, be converted into plant protein at less energy cost compared with NH_4^+ or NO_3^- . This means that plants have less energy expenditure when utilizing urea/ NH_4^+ as opposed to NO_3^- . Other researcher also found an improvement in crop productivity after coating urea with sulfur, agrotain and other biodegradable materials in different environmental conditions Zaman *et al.*, (2008).

Table 1. Selected properties of the two soils under study.

Soil	Sand %	Silt %	Clay %	Texture Class	pH (H ₂ O)	CEC cmol kg ⁻¹	CaCO ₃ ² %	Total N ³ %
1	15.6	60	22.6	Silt Loam	6.91	2.1	14.1	0.06
2	17.0	45	38.0	Clay Loam	7.90	2.7	16.1	0.14

1 The hydrometer method (Foth *et al.*, 1977)

2 As described in USDA Salinity Laboratory Handbook 60 (Richards, 1954)

3 Macro-Kjeldahl Method to include nitrate (Bremner, 1965)

Table 2. Rates and sources of N applied. Nitrogen rate: 100mg/pot = 60 kg ha⁻¹

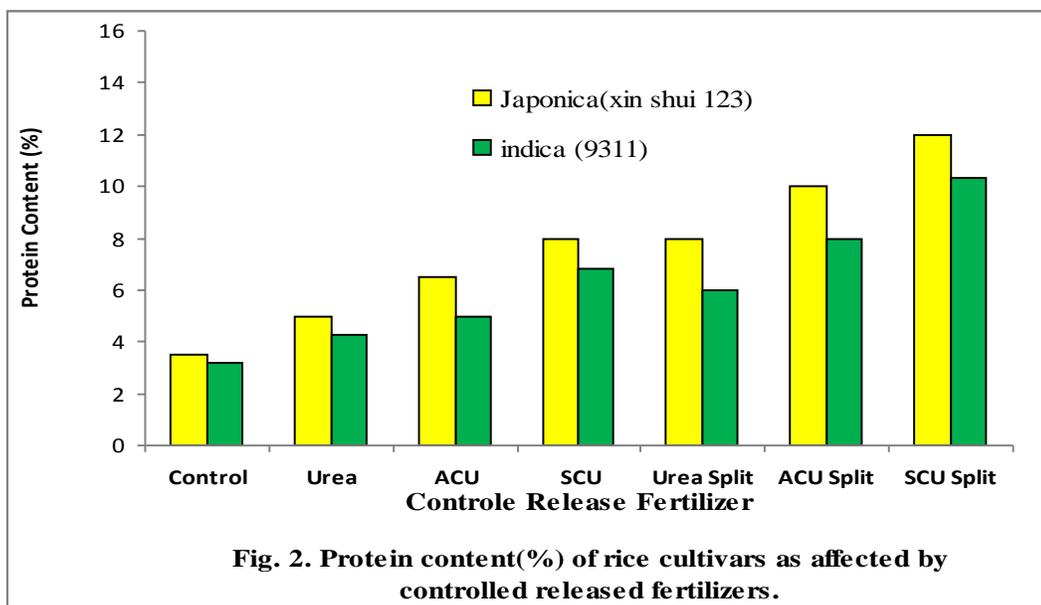
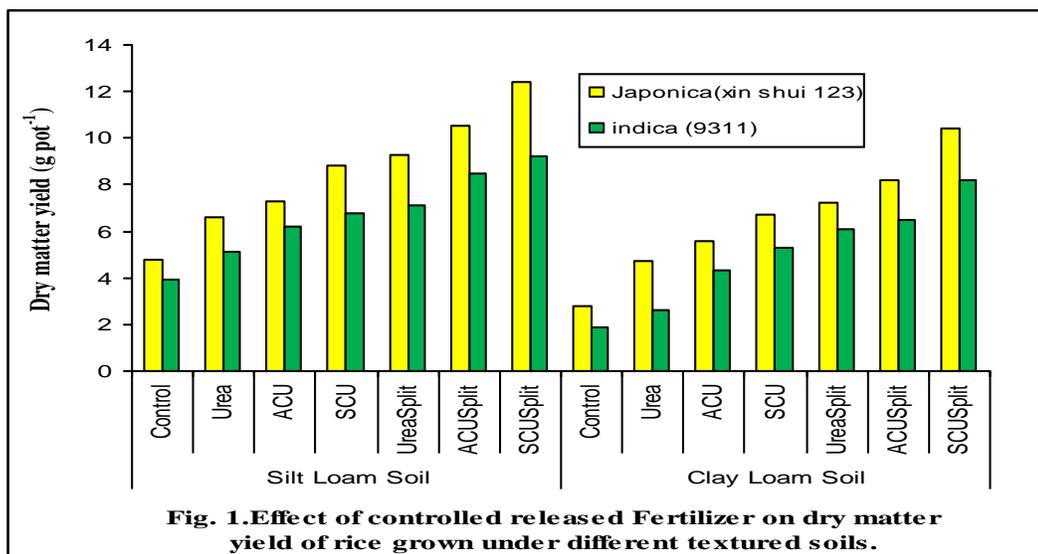
Treatment	Sources of Nitrogen	mg N/pot
N0	Control	0
N1	Urea (U)	600
N2	Agrotain Coated Urea (ACU)	600
N3	Sulfur Coated Urea (SCU)	600
N4	Urea, split (U)	300 + 300
N5	Agrotain Coated Urea, split (ACU)	300 + 300
N6	Sulfur Coated Urea, split (SCU)	300 + 300

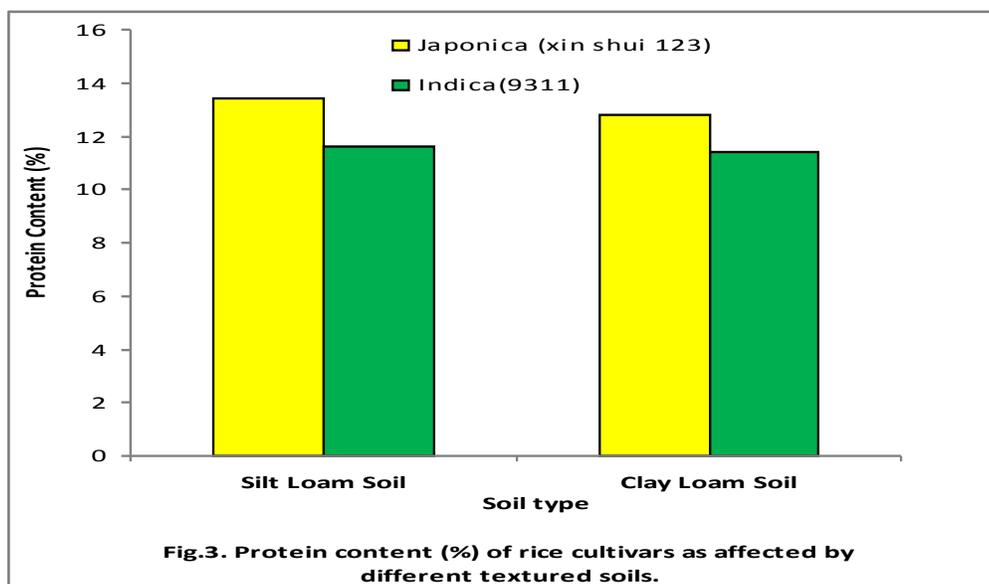
Table 3. Metrological data of rice growing period during 2011.

Weather Parameters	MONTHS						
	May	June	July	August	September	October	November
Mean Temperature °C	29	30	33	32	28	25	23
Max. Temperature °C	34	35	38	36	34	32	28
Min. Temperature °C	24	25	28	26	22	19	18
Precipitation (mm)	8	23	18	23	5	3	1
Mean Humidity (%)	55	60	73	85	63	60	55

Table 4. Effect of controlled released fertilizer on N uptake by rice cultivars grown in two different textured Soils.

Treatment kg ha ⁻¹	Application time	mg N/pot	Silt Loam		Clay Loam	
			Japonica (xinshui 123) mg N/pot	Indica (9311) mg N/pot	Japonica (xinshui 123) mg N/pot	Indica (9311) mg N/pot
N6	Sulfur coated urea, split (SCU)	300 + 300	172.4	144.2	147.4	138.2
N5	Agrotain coated urea, split (ACU)	300 + 300	162.5	134.5	111.2	96.5
N4	Urea, split (U)	300 + 300	45.8	34.5	31.4	26.8
N3	Sulfur coated urea (SCU)	600	68.8	57.2	56.3	52.2
N2	Agrotain coated Urea (ACU)	600	57.4	49.3	46.2	41.5
N1	Urea (U)	600	43.6	35.7	38.7	24.6
N0	Control	0	20.8	18.2	14.6	12.3
			L.S.D: 13.23		L.S.D: 11.76	





Conclusion: The split application of controlled release urea fertilizers are fruitful for improving yield, crop quality and soil fertility as compared to single or pre-plant application. Moreover, sulfur and urease inhibitor (agrotain) coated urea fertilizers are less susceptible to various N losses which occur in coarse textured soils. These fertilizers are eco-friendly and are environmentally safe. The superiority of these controlled release urea fertilizer can be affirmed by giving significantly higher dry matter yield, N uptake and grain quality (Protein %) by rice crop. However, the cost of these coated fertilizers has limited the use of these fertilizers.

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REFERENCES

- Ahmed, O.H., A. Hussain and H.M.H. Ahmad (2008). Ammonia volatilization and ammonium accumulation from urea mixed with zeolite and triple super phosphate. *Acta Agric. Scandinavia*, 58:182–186
- Bolan, N., S. Saggar and J. Singh (2004). The role of inhibitors in mitigating nitrogen losses in grazed pasture. *New Zealand Soil News*, 52: 52–58
- Hayat, K., and A.Z.Khan (2013). Influence of super and agrotain coated Urea on growth, yield and quality of Wheat. M.Sc (H) thesis Department of Agronomy, University of Agriculture, Peshawar, Pakistan
- Iqbal, M. F., Z. Iqbal, M. Farooq, L. Ali and M. Fiaz (2013). Impact of nitrogenous fertilizer on yield and quality of oat. *Pakistan J. Science.*, 65(1):1-4
- Junejo, N., Y.M. Khanif and M.M. Hanafi (2009a). Effect of Cu and palm stearin coating on ammonia volatilization of Urea. *Res. J. Agric. Biol. Sci.*, 5: 608–612
- Junejo, N., S. Sagar and M.Y. Khanif (2009b). Reducing NH₃ emission loss from urea fertilizer: urease inhibitors, polymer coatings and micronutrient. *New Zealand Soil News*, 57: 193–199
- Khalil, S. K., Zakria, A. Z. Khan, and M. Ramazan (2003). Effect of plant density, nitrogen and zinc on sweet corn. *Pakistan J. Sci.*, 55(3-4):62-66
- Khalil, I. M., R. Gutser and U. Schmidhalter (2009). Effects of urease and nitrification inhibitors added to urea on nitrous oxide emission from loess soil. *J. Plant Nutr. Soil Sci.*, 172: 651–660
- Krajewska, B., (2009) Urease I. Functional, catalytic and kinetic properties: A review. *J. Mol. Catal. B: Enzymatic*, 59: 9–21
- Leong, T.K., (2002). The development of cooper-coated urea for rice production. Master and Agric. Sci. Thesis, UPM, Serdang, Malaysia
- Mohsina, H., R. Khalil, A.S. Muneer and M.A. Shahzad (2004). Effect of substrate concentrations, temperature and cropping system on hydrolysis of urea in soils. *Int. J. Agric. Biol.*, 6: 964–966
- Nasima, J., M.Y. Khanif, W.Y. Wan and K.A. Dharejo (2010). Maize Response to Biodegradable Polymer and Urease Inhibitor Coated Urea. *Int. J. Agric. Biol.*, 12: 773–776
- Purkayastha, T.J. (2009). Evaluation of some modified urea fertilizers applied to rice. *Fert. News*, 42: 53–56

- Reddy, D., and K. Sharma (2000). Effect of amending urea fertilizer with chemical additives on ammonia volatilization loss and nitrogen-use efficiency. *Biol. Fert. Soil*, 32: 24–27
- Shaviv, A. (2001). Improvement of fertilizer efficiency Product processing, positioning and application methods. *Proc. Int. Fert. Soc.*, 469: 1–23
- Villar, J.M and E. Guillaumes (2010). Use of nitrification inhibitor DMPP to improve nitrogen recovery in irrigated wheat on a calcareous soil. *Spanish J. Agric. Res.* 8 (4):1218-1230
- Xu, X., L. Zhou, V. Cleemput, and O. Wang (2000). Fate of urea-¹⁵N in a soil-wheat system as influenced by urease inhibitor hydroquinone and nitrification inhibitor dicyandiamide. *Plant Soil* 220:261–270
- Watson, C.J. (2000). Urease activity and inhibitors Principle and practice. *Int. Fert. Soc., Proceed. No.* 454
- Zaman, M., M.L. Nguyen, J.D. Blennerhassett, and B.F. Quin (2008). Reducing NH₃, N₂O and NO₃-N losses from pasture soil with urease or nitrification inhibitors and elemental S-amended nitrogenous fertilizers. *Biol. Fert Soils* 44, 693–705
- Zhang, J.H., J.L. Liu, J.B. Zhang, F.T. Zhao, Y.N. Cheng, and W.P. Wang (2010). Effects of Nitrogen Application Rates on Translocation of Dry Matter and Nitrogen Utilization in Rice and Wheat. *Acta Agron Sin*, 2010, 36(10): 1736–1742.