EFFECTS OF APPLIED UREA IN THE SOIL ON DAILY CHANGES OF PH AND LENGTH OF PADDY (ORYZA SATIVA) GROWN IN THE SEEDLING

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Abstract: Seeds of oryza sativa were cultivated in the seedlings and the effects of urea on the changes of pH in the soil and growth of paddy were observed. Urea (80 kg/ha) was applied to one of the seedlings after 20 days of germination and the soil pH and length of paddy were determined from 25 days of urea application. Soil pH was gradually decreased with increasing paddy grown in the control seedling. However, no changes of pH were found in the urea treated seedling up to 20 days of germination and were similar to the control paddy. The pH of soil compared to the control falls gradually with increasing paddy growth. We also examined the effect of urea on paddy length and compared to the control paddy. Paddy length was gradually increasing in both the control and urea treated seedlings from the germination. However, higher length was found in the urea treated seedling. Up to 20 days of germination of paddy, no differences of paddy length in the two seedlings were observed. These findings suggest that urea is the potent compound inducing growth of paddy and pH measurements indicate the period where optimum uptake of urea is happened.

Key words: Soil pH, BRRI-27, Urea, Paddy length, Plant growth

Introduction

Plant growth and development is an important feature of plant. Various factors are involved in the development of plant growth. Soil fertility because of the higher activity of soil microorganisms is also an important factor for plant growth development. Most plants utilize urea and amino acids as N substrates and responses to these compounds vary among species. Some have evolved strategies that favor one specific substrate or a combination of substrates. Most crop species grow optimally with a mixture of ammonium and nitrate, the latter generally being the most abundant N form in freely drained aerobic soil environments (Crawford et al., 1998) becomes an increasingly important substrate on ammonia-fertilized soils or on poorly drained, acidic soils where nitrification by micro-organisms is limited (Rice et al., 1972). Urea has been recently used as a potent fertilizer inducing fertility and plant growth. Nitrification is the oxidation of ammonium to nitrate. In soils, the process is mediated by autotrophic and heterotrophic bacteria. The nitrification process is controlled primarily by ammonium and oxygen concentrations. The transport of ammonium by diffusion is influenced by the organic matter status and cation exchange capacity of the soil, the presence of reduced iron and manganese, the bulk density, and the rate of nitrification in the oxidized soil layer and rice rhizosphere. Biological denitrification is

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the dissimilatory reduction of nitrate and nitrite to produce NO, N₂O and N₂. Soil factors that strongly influence denitrification are oxygen (which is controlled primarily by soil water content), nitrate concentration, pH, temperature, and organic carbon (Peoples et al., 1995). The loss of soil nitrogen in various ways is compensated by the addition of urea. Recent investigation reveals that growth in response to urea is characterized by the enhanced cellular function, increased protein and carbohydrate synthesis (Nishi et al., 2001) as well as increased cell proliferation and differentiation. To clarify the urea induced plant growth, we measured soil pH before and after application of urea in the soil and also measured the length of paddy.

Materials and Methods
Plant Materials: In this study, rice seedlings were raised in the Rajshahi University Campus and grown with BRRI-27 seeds. The seeds were collected from the near by Bangladesh Rice Research Institute (BRRI), Sympur, Rajshahi, Bangladesh. For germination of seed, light, watering and other essential requirements were implied. The efficiency of germination of seeds was 99 % from the observation. Plants in the control seedling were not treated with urea while the other seedling was treated with urea (80 Kg/ha). The plants grown in the seedlings were taken care of weed control.

Determination of soil pH: 20 g soil was taken in a 250 ml beaker and 50 ml distilled water was added with the soil. The contents were thoroughly stirred with magnetic stirrer for half an hour. pH of the suspension was measured with a digital pH meter. Soil pH was determined before germination of seed, 20 days after application of urea. In the control seedling, pH was determined similarly before and after germination of seeds.

Determination of paddy length: Paddy length was determined after germination and before and after 20 days of urea application. Paddy length was determined by using measuring scale in different stages of growth both in the control and urea exposed seedlings.

Results
Determination of soil pH in the seedlings before germination of seeds
Soil pH determination is an indication of the acidic or basic phase of soil. The higher pH of soil is because of the lower utilization of amino nitrogen in the plant. The optimum pH for the growth of paddy is 5-9. To examine whether the soil is suitable for the growth of paddy, we determined the soil pH in the seedlings. The pH was 8.07 in the control and urea-treated seedlings which were within the normal range. The pH indicates that the amino nitrogen in the soil is suitable for growing of paddy.

Determination of soil pH after germination and before urea application
Soil pH was determined in the seedlings up to twenty days from germination of seeds before urea application. Soil pH was determined by the procedure mentioned before. The results are compared with the control seedling and illustrated in Table 1. There were no differences in soil pH between two groups. However, pH was gradually decreasing from the germination of paddy in the two seedlings because of their increasing growth. The results indicate that the higher utilization of amino nitrogen by paddy has been taken place.

Table 1. Showing soil pH in the control and urea-treated seedlings after germination of paddy. The experiment was carried out in absence of application of urea. Measurement was taken at five days interval. No differences of soil pH in the two seedlings were observed.

| Days after germination of paddy | Soil pH in the control seedling treated without urea | Soil pH in the urea-treated seedling |
|---------------------------------|-----------------------------------------------------|-------------------------------------|
| Five days                       | 7.8                                                 | 7.8                                 |
| Ten days                        | 7.5                                                 | 7.5                                 |
| Fifteen days                    | 7.33                                                | 7.33                                |
| Twenty days                     | 7.15                                                | 7.15                                |
| CV %                            | 3.71                                                | 3.71                                |
Determination of soil pH after first dose of urea application

To check whether urea changes the soil pH, we determined soil pH in the control and urea treated seedlings from 20 to 40 days after application of the first dose of urea. The results are illustrated in Table 2. The initial pH determined at 25 days was 7.1 and 7.25 for the control and urea-treated seedlings. Soil pH was gradually decreasing up to 40 days of the measurement for both groups. Application of urea enhances soil pH because of the increased production NH4+ from urea.

Table 2. Soil pH after application of urea in one of the seedlings. The other seedling was not treated with urea. Soil pH was determined after urea application. The results are compared with control seedling. Measurement was taken at five days interval.

| Days after germination of paddy | Soil pH in the control seedling treated without urea | Soil pH in the urea-treated seedling |
|--------------------------------|-------------------------------------------------|-------------------------------------|
| Twenty five days               | 7.10                                            | 7.25                                |
| Thirty days                    | 7.05                                            | 7.15                                |
| Thirty five days               | 6.85                                            | 7.10                                |
| Forty days                     | 6.75                                            | 7.05                                |
| CV %                           | 2.38                                            | 1.19                                |

Determination of paddy plant length after germination and before urea application

Paddy length gradually increases in urea treated and non-urea treated (control) seedlings. The length of paddy in both the seedlings were same. Table 3 indicates the length of paddy plant in urea treated and without urea treated (control) seedlings. Measurement was taken from 5 to 20 days after germination of seeds. 15 cm, 24 cm, 28 cm and 30 cm paddy length were observed in both the seedlings. Increased paddy length in both seedlings is caused by the utilization of soil nitrogen and other nutrients. The soil nitrogen might be from the degradation of soil urea.

Table 3. Showing paddy length in the control and urea-treated seedlings after germination of paddy. The experiment was carried out in absence of application of urea and from 5 to 20 days. Measurement was taken at five days interval. No differences of paddy length in the two seedlings were observed.

| Days after germination of paddy | Length of paddy in the control seedling | Length of paddy in the urea-treated seedling |
|--------------------------------|----------------------------------------|---------------------------------------------|
| Five days                      | 15 cm                                  | 15 cm                                       |
| Ten days                       | 24 cm                                  | 24 cm                                       |
| Fifteen days                   | 28 cm                                  | 28 cm                                       |
| Twenty days                    | 30 cm                                  | 30 cm                                       |
| CV %                           | 27.42                                  | 27.42                                       |

Determination of paddy plant length after first dose of urea application

Urea was applied after 20 days of germination of seeds. Paddy length was measured at 25, 30, 35 and 40 days. As shown in Table 4, paddy length gradually increases in urea treated and without urea treated (control) seedlings. However, paddy length in urea treated seedling is higher than without urea treated (control) seedling. The changes of increase in paddy length up to 40 days of observation were stimulated by urea application compared to the control paddy. The results show that urea is the most potent fertilizer utilized in enhancing plant cellular growth.

Table 4. Paddy length after application of urea in one of the seedlings. The other seedling was not treated with urea. Paddy length was determined after urea application. The results are compared with control seedling. Measurement was taken at five days interval.

| Days after germination of paddy | Length of paddy in the control seedling | Length of paddy in the urea-treated seedling |
|--------------------------------|----------------------------------------|---------------------------------------------|
| Twenty five days               | 30 cm                                  | 33 cm                                       |
| Thirty days                    | 30.5 cm                                | 42 cm                                       |
| Thirty five days               | 33 cm                                  | 50 cm                                       |
| Forty days                     | 35 cm                                  | 56 cm                                       |
| CV %                           | 7.22                                   | 22.03                                       |
Discussion

Urease which is involved in the degradation of urea in the soil results in the release of $\text{NH}_4^+$ and $\text{CO}_2$. The higher activity of urease enhances the higher production of $\text{NH}_4^+$ and stimulates the increased pH of soil. The changes of soil pH are an index of utilization of soil urea to the plant cells. In our study, after application of urea, the soil pH was abruptly increased and was gradually decreasing with the increasing paddy length because of the higher utilization of $\text{NH}_4^+$ by the cells. In the urea induced paddy seedling, growth of paddy is stimulated as observed by the higher paddy length. At 25th day after application of urea, soil pH 7.25 was observed while for the control seedling, the pH was changed to 7.1. The paddy length was co-related with the pH and were 30 cm and 33 cm respectively for the control and urea induced paddy.

Ammonium ions in the soil solution enter into an equilibrium reaction with $\text{NH}_3$ in the soil solution. The soil solution $\text{NH}_3$ is, in turn, subject to gaseous loss to the atmosphere. Soil pH and concentration of $\text{NH}_4^+$ in the soil solution are important factors affecting amount of $\text{NH}_3$ loss to the atmosphere. As soil pH increases, the fraction of soil-solution $\text{NH}_4^+$ plus soil-solution $\text{NH}_3$ in the $\text{NH}_3$ form also increases by an order of magnitude for every unit of pH above 6.0, thus increasing losses of soil-solution $\text{NH}_3$ to the atmosphere. Collectively, $\text{NH}_3$ volatilization: 1) is of most importance on calcareous soils, especially as soil pH exceeds 7; 2) losses increase with temperature and can be appreciable for neutral or alkaline soils as they dry out; 3) is greater in soils of low CEC, such as sands; 4) losses can be high when high-N organic wastes, such as manure, are permitted to decompose on the soil surface; 5) losses are high from urea applied to grass or pasture, as a result of hydrolysis of the urea to $\text{NH}_3$ by indigenous urease enzyme; 6) losses from soil and fertilizer N are decreased by growing plants (Stevenson, 1986). It is assumed that if higher $\text{NH}_3$ loss is prevented, the utilization of amino nitrogen in plant will be enhanced. However, atmospheric $\text{NH}_3$ and the utilization of soil $\text{N}_2$ from $\text{NH}_3$ should be balanced.

As ammonia formed from urea is oxidized to nitrite by nitrosomonas bacteria, the nitrite is again oxidized to nitrate by nitrification process and caused by nitrobacter. The nitrate which is the plant food and is utilized by the cells in the form of protein. Therefore, nitrification causes the enhancement of total growth of paddy. The utilization of $\text{N}$ in plant is an essential metabolic function and has been involved in the regulation of nutrition and nitrogen signaling as demonstrated by Parsons and Sunley (2001). It is reported that urea enhances the synthesis of proteins, enzymes (Claus-Peter et al., 2002; El-Shora, 2001) and other cellular functions (Nishi et al., 2001). Cell proliferation and differentiation in response to urea has been observed. Amylopectin, the major component of starch, is synthesized by concerted actions of ADP-Glc pyrophosphorylase (AGPase), soluble starch synthase (SS), starch-branching enzyme (BE) and starch-debranching enzyme. It has been demonstrated (Satoh et al., 2003) that the gelatinization properties of the starch in response to urea affects the structural differences of amylopectin by mutation of genes. Urea is utilized by the cells by interaction with nitrate-induced nitrate transporter. Recent investigation reveals that Nrt1 and Nrt2 have been cloned in the plant (Crawford et al., 1998). Therefore, uptake pathways of urea in the plant are not properly clarified. It is probable that urea is taken by the cells through urea transporter. Therefore, further studies are needed to clarify the mechanism of urea-induced growth of paddy.

Conclusion

Our study indicates that urea has stimulatory effect on growth of paddy which is reciprocally related to the soil pH caused by the applied urea. The rate of uptake of amino nitrogen in the plant is also related to the availability of urea in the soil as well as with the disappearance of soil urea. It is assumed that higher the disappearance of soil nitrogen, higher the utilization to cells thereby augmentation of plant growth.
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References

Crawford, N.M. and Glass, A.D.M. 1998. Molecular and physiological aspects of nitrate uptake in plants. Trends in Plant Science, 3: 389–395.

Claus-Peter, W., Tiller, S. A., Taylor, M.A. and Davies, H.V. 2002. Leaf Urea Metabolism in Potato. Urease Activity Profile and Patterns of Recovery and Distribution of 15N after Foliar Urea Application in Wild-Type and Urease-Antisense Transgenics. Plant Physiol, 128: 1129-1136.

El-Shora, H. M. 2001. Properties and immobilization of urease from leaves of Chenopodium album (C3). Bot. Bull. Acad. Sin., 42: 251-258.

Nishi, A., Nakamura, Y., Tanaka, N. and Satoh, H. 2001. Biochemical and Genetic Analysis of the Effects of Amylose-Extender Mutation in Rice Endosperm. Plant Physiol, 127: 459-472.

Parsons, R. and Sunley, R. J. 2001. Nitrogen nutrition and the role of root–shoot nitrogen signalling particularly in symbiotic systems. Journal of Experimental Botany, 52: 435-443.

Peoples, M. B., Freney, J. R., Mosier, A. R. 1995. Minimizing gaseous losses of nitrogen. In: Nitrogen Fertilization in the Environment, P.E. Bacon (ed.). Marcel Dekker, Inc., New York, pp. 565-602.

Rice, E. L., Pancholy, S. K. 1972. Inhibition of nitrification by climax ecosystems. American Journal of Botany, 59: 1033–1040.

Satoh, H., Nishi, A., Yamashita, K., Yoko, Takemoto, Y., Tanaka, Y., Hosaka, Y., Sakurai, A., Fujita, N. and Nakamura, Y. 2003. Starch-Branching Enzyme I-Deficient Mutation Specifically Affects the Structure and Properties of Starch in Rice Endosperm. Plant Physiology, 133: 1111-1121.

Stevenson, F. J. 1986. Cycles of Soil: Carbon, Nitrogen, Phosphorus, Sulfur, Micronutrients. John Wiley & Sons, NY. 380 pp.
Haque, M.S.; Hasan, M.K; Islam, M.A. and Salequzzaman, M. 2008. Effects of Applied Urea in the Soil on Daily Changes of Pn and Length of Paddy (*Oryza Sativa*) Grown in the Seedling. *Khulna University Studies*, 9(2): 311-316.