TIMING OF NITROGEN UPTAKE PATTERN BY MAIZE USING ¹⁵N ISOTOPE TECHNIQUE AT DIFFERENT GROWTH STAGES

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Keywords: ¹⁵N isotope, Maize, N uptake, Nitrogen derived from fertilizer (NDFF), Nitrogen derived from soil (NDFS)

Abstract

It is necessary to know the right time for nitrogen (N) fertilizer application to meet the plant's need in order to reduce N loss. A glasshouse experiment was conducted at Universiti Putra Malaysia to determine the timing of N uptake at different growth stages of maize. Nitrogen was labeled as ¹⁵N urea with 10% atom excess (a.e). The total N uptake increased until eighth week of planting. After the eighth week of planting, total N decreased due to remobilization of nitrogen from leaf and stalks to grain. The maximum nitrogen use efficiency (NUE) was only 37% throughout the growing season. This indicated that the NUE was very low. NUE can be increased by understanding the right rate and time of nitrogen fertilizer application. The best timing for nitrogen fertilizer application was before the eighth week of planting which reduced the loss of N.

Introduction

The applied fertilizer that is not absorbed by plants can be more than 50%. Increasing NUE is one of the approaches to overcome this problem. Nitrogen use efficiency is a quantitative measurement of the actual uptake of fertilizer nutrient by the plant in relation to the amount of nutrient added to the soil as fertilizer. It is important to emphasize on the efficient use of fertilizer to obtain the highest possible yield with a minimum fertilizer application (FAO 1985), the term referred to as yield optimization.

In general, maize cropping season requires 120 days to reach physiological maturity and along this maturity, N losses are high. There is a lack of information about timing of nitrogen uptake especially in soils of Malaysia. Therefore, further research is needed to elucidate the timing of nitrogen uptake by maize. The precise method to estimate fertilizer efficiency is by using ¹⁵N isotope technique (Choudhury and Khanif 2001). There are several N sources for plant including soil, biological N fixation and fertilizer for plant. Nitrogen derived from fertilizer (NDFF) can be traced by using fertilizer with ¹⁵N isotope. Information obtained from tracer technique is very important in developing N-fertilizers that will release N according to the need of plants (Hashim *et al.* 2015). The most common and stable isotope used is ¹⁵N isotope (IAEA 2001). The objective of this study was to determine the timing of N uptake and NUE for maize using ¹⁵N isotope technique.

Materials and Methods

The study was carried out in a glasshouse at University of Putra Malaysia. Maize was planted in ceramic pots with a dimension of 33 cm diameter \times 32 cm height containing 20 kg of soil. The selected soil belongs to Munchong series (Typic Hapludox). The soil was analysed for physiochemical properties (Table 1) including soil pH which was measured using pH-water method

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(MacLean 1982). The CEC of the soil was determined by ammonium-acetate extraction method as described by Schollenberger and Simon (1945), organic C by the method as described by Walkley and Black (1934) and total N by the method as decribed by Bremner and Mulvaney (1982). The textural class of the soil was determined using USDA soil texture triangle (USDA 1983). The pre-germinated seeds of maize from a cultivar, namely Thai Super Sweet were sown at two seeds per pot. The experiment was carried out in CRD with four replications.

Soil properties	Values
pH	4.5
Texture	Clay
Clay (%)	68
Sand (%)	17
Silt (%)	15
Total C (%)	3.7
Total N (%)	0.12
CEC (cmol/kg)	8.7

Table 1. Physical and chemical properties of the selected soil used in the study.

Fertilizer was applied at the rate of 120 kg N/ha, 60 kg P_2O_5 /ha and 120 kg K_2O /ha. Nitrogen in the form of urea labelled with ¹⁵N was applied as a treatment. Fertilizer was applied 3 times throughout the growing season. Urea labelled with ¹⁵N used was ISOTECTM urea ¹⁵N₂, 10% atom excess (a.e). Triple superphosphate was used as the source of phosphorus (P) and muriate of potash fertilizer as the source of potassium (K) were applied 1 day after transplanting.

Plant was harvested at one week intervals during the ¹⁵N treatment period until 12th week. The harvested plants were separated into roots, leaves, stem and grain and dried at 60°C for 24 hrs. Total N was determined by Kjehdahl method (Bremner and Mulvaney 1982). The enrichment of ¹⁵N in plant was determined using emission spectrometry (IAEA 2001) at the Malaysian Nuclear Agency. The percentage of nutrient use efficiency (NUE) was calculated by dividing the N from ¹⁵N fertilizer with the applied fertilizer multiply by one hundred. The percentage of nitrogen derived from fertilizer (NDFF) and nitrogen derived from soil (NDFS) in plants was calculated to determine the percentage of N from fertilizer and soil (IAEA 2001).

Results and Discussion

The total N uptake in plant increased from the first harvest at the first week to the 8th week (Fig. 1). During the first week, the N uptake was very low (0.07 g N/pot) followed by the second week (0.49 g N/pot) and consistently increased and reached the maximum N uptake in the 8th week of planting (1.77 g Npot). After the 8th week, total N uptake started to decrease and reduced to 1.51 g N/pot, 1.23 g N/pot, 1.58 g N/pot, and 1.8 g N/pot at 9th, 10th, 11th, and 12th week, respectively.

The result indicated that the total N uptake by maize was affected by the stage of crop development (Fig. 1). During the early stage of growth until the 8th week, maize was at vegetative stage. After that, maize was at the reproductive stage (Ross *et al.* 2013). The low N uptake in the first week was due to the kernel which has stored all the essential nutrients for the growth, thus the N from soil was not critical in the first week. But after the emergence, the roots began to take up the nutrients from the soil and fertilizer. At the early weeks, roots were confined to a very small soil zone and the majority of soil volume was not effectively exploited by plant roots (Clarkson 1985). Nitrogen had mobility characteristics which allowed it to be utilized in one tissue then

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transported to another tissue by which it is to be used (Choudhury and Khanif 2001, Ross *et al.* 2013). After 8th week, N started to be remobilized from stalks and leaves to the grain during the grain-filling period. According to Salon *et al.* (2001), using ¹⁵N tracing experiment, the grain filling was a period where N uptake declined. N deficiency at any time of maize plant's life reduces yield, but if the deficiency occurred at the vegetative stage which was at the first week to 8th week of planting, yield losses may be severe. For this reason, understanding the relationship of N supply and uptake of N in maize is very important in developing successful N application to meet the plant N requirement. Nitrogen should be released during the time when uptake of N is at the maximum level for vegetative and reproductive growth (Ross *et al.* 2013).



Fig. 1. Total uptake of nitrogen (N) in maize plant at different weeks after planting.

Study conducted with standard practice of maize cultivation in which the N source was urea, the NUE for maize was 37% (Fig. 2). The highest NUE obtained from the 8th and 9th week during the vegetative stage was 37 and 36.6%, respectively. From this study, it showed that more than 50% N of applied fertilizer was not consumed by maize plants. Rather, it was either lost to the environment or remained in the soil. Similar finding for maize was reported by Cassman *et al.* (2002). According to Raun and Johnson (1999), NUE for cereal in worldwide was estimated at only 33%. The highest NUE recorded from the 8th and 9th week of planting was due to the size of maize plant and its root system.



Fig. 2. Nitrogen use efficiency from fertilizer by maize at different weeks after planting.

Low NUE obtained in this study was probably due to thigh N loss through several means like NH₃ volatilization, denitrification and leaching (Ladha *et al.* 2005). Leaching and denitrification

contribute to N losses from the agro-ecosystem, decreasing crop N use efficiency and increasing the potential for environmental problems (Bedard-Haughn *et al.* 2003). Nitrogen loss caused by NH₃ volatilization can lead to acidification and eutrophication while N₂O emission into atmosphere caused the depletion of the ozone layer (Cameron *et al.* 2013, Fowler *et al.* 2013). Thus, understanding N uptake and its distribution in plant tissue was very important for environmental concerns and quality of product (Rehman *et al.* 2011). Increasing NUE can minimize the impact of N fertilizer to the environment and it also increases crop production.

There were two sources for nitrogen plant uptake, which were from soil and fertilizer (Fig. 3). Generally, NDFF was higher in maize plant tissues, root, leaf, stem and grain throughout the growing periods but during the first week of growth, only 90% of N in plant was derived from soil (NDFS). Starting from the second week, N derived from fertilizer (NDFF) in the plant tissues increased with time. NDFF was in the range of 26 to 61% in roots, 42 to 60% in leaf, 52 to 65% in stem and 55 to 58% in grain.



Fig 3. Nitrogen source (NDFF) and (NDFS) in root (a), leaf (b), stem (c) and grain (d) at different weeks after transplanting.

In the leaf, the highest NDFF was 61% at the 9th week after transplanting and the lowest during the 1st week after transplanting which was 42% (Fig. 3b). In stem, the highest NDFF was recorded at 9th week after transplanting with the NDFF value was 65% and the lowest was 52% at the 12th week (Fig. 3c) while in the grain, NDFF was 55% at the 11th week and 58% at the 12th

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week after transplant (Fig. 3d). The lowest NDFF in leaf and root of maize was observed at the 1st week of transplanting (Fig. 3).

In this study, NDFF was higher in maize plant than NDFS, indicating that the plant absorbed more N from fertilizer than from the available N in soil. The highest uptakes from NDFF were at the 8th and 9th week after transplanting with the values range from 50 to 60%. This result revealed that only half of the fertilizer applied was taken up by plant. This was due to the fast release of N from urea which N was rapidly converted to NH_4 . A fraction of fertilizer was removed by plant while other fractions were lost to the environment (Hashim *et al.* 2015). This problem can be solved using enhanced efficiency fertilizer (EEF) which is able to release N slowly to the soil, allowing plant to uptake N at the right stage growth. The similar result obtained from other studies with ¹⁵N reported 20 to 50% of N losses from fertilizer in cereal production. The losses were due to the combined effects of denitrification, volatilization and/or leaching (Karlen *et al.* 1996).

Low NDFF resulted in economic loss and environmental pollution (Raun and Johnson 1999, Ladha *et al.* 2005). Raun and Johmson (1999) recorded for cereal production, the loss of N fertilizer estimated at \$15.9 billion annually. This loss affects the economic profit with the increase of production cost in agriculture. Depending on the fertilizer use, a 1% increase in N use efficiency would result in \$2.3 million saving in N fertilizer cost for cereal production worldwide (Raun and Johnson 1999).

The N fertilizer management is important not only for economic purpose but also for environment. Understanding the rate and timing of plant N uptake can improve NUE in maize. The N uptake of maize occurred up to 8th week of planting. After 8th week, plant consumed N from tissues through remobilization of N. The NUE of maize only reached 37% which is considerably low. Nitrogen derived from fertilizer (NDFF) in maize was low, ranging 50 to 60%. The data compiled from this study had provided information on efficiency of current urea used as N source and it allows in developing practical EEF to be made as efficient as possible. Supply of N fertilizer in the correct amount and time can reduce the N loss.

Acknowledgements

Financial support for this study was received from Long-Term Research Grant Scheme of the Ministry of Education, Malaysia under the project 'One BAJA: The Next Generation Green and Economical Urea'. Special thanks go to the science officer of Department of Land Management, University of Putra Malaysia for their expert works and guidance throughout the study.

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(Manuscript received on 11 December 2016; revised on 27 February, 2017)