Potato Stolon and Tuber Growth Influenced by Nitrogen Form

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Abstract: Potato tuber initiation and its growth are key processes determining tuber yield, which are closely related to stolon growth, and are influenced by many factors including N nutrition. We investigated the influences of different forms of nitrogen (N) on stolon and tuber growth in sand culture with a nitrification inhibitor during 2010 – 2011, and using two potato cultivars. Plants supplied with NO₃-N (N as nitrate, NO₃⁻) produced more and thicker stolons than those supplied with NH₄-N (N as ammonium, NH₄⁺) at tuber initiation stage. In the plants fed NO₃-N, the stolon tips swelled or formed tubers earlier and produced more tubers than in those fed with NH₄-N. However, no significant difference was observed among N forms in terms of tuber yield at harvest, this may have been because of the shoot growth rate at tuber initiation stage was lower in the plants fed NO₃-N. During the tuber bulking stage, the difference in shoot DWs among N forms began to decrease, and the shoot DW of plants fed NO₃-N was even heavier than those fed NH₄-N in some cases. The influence of N form on potato plant growth may therefore vary with the potato growth stage.

Key words: Ammonium, Nitrate, Potato, Shoot, Stolon, Tuber.

Potato (Solanum tuberosum L.) is an important crop widely used both for food and as an industrial source of starch. The tubers are the harvestable component of the crop, originating from stolon swelling. The processes determining the time of tuber formation and the number of tubers per plant play an important role in crop yield; therefore, factors and processes regulating tuber formation are always important topics in potato research (Struik et al., 1989; Pelacho and Mingo-Castel, 1991; Helder et al., 1993; Wurr et al., 1997; Brown, 2007; Chang et al., 2012). Previous research has shown that tuber size distribution within a plant is influenced by both timing of tuberization and the number of stolons tuberizing per plant; a key determinant of the marketable component of total yield (Helder et al., 1993; Wurr et al., 1997; Chang et al., 2012). Tuberization is sensitive to environmental conditions (Brown, 2007), therefore understanding the influence of environments on stolon growth and tuber formation is a precondition for agronomic management aimed at maximizing marketable tuber yield.

High temperature and long photoperiods have been suggested to stimulate stolon initiation and growth, whereas low temperature or short photoperiods lead to tuberization and tuber expansion (Ewing and Wareing, 1978; Struik et al., 1989; Pelacho and Mingo-Castel, 1991; Brown, 2007). Further research has revealed that temperature and photoperiod play roles in tuberization via internal phytohormones including gibberellin, cytokinin, jasmonate, and abscisic acid (Hannapel, 2007). In addition, planting depth (Pavek and Thornton, 2009), planting density (Van der Veeken and Lommen, 2009), and soil moisture (Haverkort et al., 1990) have impacts on tuber number of the plant, suggesting that they are involved in the switch from stolon growth to tuber formation. As an essential element, a plenteous supply of nitrogen (N) is the base for higher tuber yield; however, Krauss and Marschner (1982) found that continuous N supply in hydroponic culture delayed tuber formation. Sarkar and Naik (1998) obtained similar results in which a large amount of available N inhibited tuber formation.

Nitrate (NO₃⁻) and ammonium (NH₄⁺) are two forms of N for plant uptake. Different crops have different responses to N form. Some crops prefer nitrate and severe inhibition of plant growth occurs when plants are supplied with only ammonia; others grow better with an ammonium source (Marchsner, 1995; Elgharably et al., 2010; Gao et al., 2012). For most plants, a combination of NH₄⁺ and NO₃⁻ is preferred over application of a sole source of either form (Marchsner, 1995), and the optimal NO₃⁻/NH₄⁺ ratio varies with the crop (Claussen and Lenz, 1995, 1999;
Hassan et al., 2008; Elgharably et al., 2010). Although the effect of different forms of N on potato growth has been studied by several groups, there is discrepancy in their results (Lorenz et al., 1974; Davis et al., 1986; Cao and Tibbits, 1994; Osaki et al., 1995; Serio et al., 2004). MS culture medium, which is usually used for potato tissue culture and potato micropropagation, contains NO$_3^-$ and NH$_4^+$ at a 2:1 ratio, but no physiological basis for this ratio has been reported. Furthermore, there is little research regarding the response of stolon growth to N form, and closely related tuber formation and growth. Our objective was therefore to determine the influences of N form on potato stolon growth as well as tuber formation and tuber growth under sand culture conditions.

### Materials and Methods

1. **Plant materials and experimental design**

Seed potatoes used in this study were all virus-free mini seed tubers: Cultivars of potato were Kexin-1 with a mean tuber weight of 30 ± 3 g in 2010, Kexin-1 (10 ± 2 g) and Shepody (7 ± 3 g) in 2011. A pot experiment was conducted in a greenhouse at the Inner Mongolia Agricultural University. The temperature was between 21 and 29ºC, the light period was 12 – 15 hr, the light intensity was about 500 – 700 μmol photons m$^{-2}$ s$^{-1}$, and the average relative humidity was around 55%. Each 15 L pot contained 20 kg of river sand rinsed with distilled water. Seed potato tubers were planted 8 cm deep on 2 July, 2010 and 20 April, 2011.

After planting, the pots were irrigated once a week with distilled water. After potato emergence, plants were irrigated with nutrient solutions every 4 days during the early growth stage ($\leq$ 20 days after emergence (DAE)) and every 2 days during the late growth stage ($\geq$ 21 DAE). Three nutrient solutions (three treatments) were prepared; each solution contained 0.25 mmol L$^{-1}$ KH$_2$PO$_4$, 0.2 mmol L$^{-1}$ CaCl$_2$, 0.5 mmol L$^{-1}$ MgSO$_4$, 50 μmol L$^{-1}$ Fe-EDTA, 50 μmol L$^{-1}$ KCl, 25 μmol L$^{-1}$ H$_3$BO$_3$, 2 μmol L$^{-1}$ MnSO$_4$, 2 μmol L$^{-1}$ ZnSO$_4$, 0.5 μmol L$^{-1}$ CuSO$_4$, 0.5 μmol L$^{-1}$ Na$_2$MoO$_4$, and 3 mmol L$^{-1}$ of N, and one of three N forms: 100% NO$_3^-$-N, 100% NH$_4^+$-N, and NO$_3^-$ and NH$_4^+$ at a 1:1 ratio (NO$_3$-NH$_4$-N). The NH$_4$-N resource was delivered as (NH$_4$)$_2$SO$_4$, and the NO$_3$-N resource as Ca(NO$_3$)$_2$. Each treatment was conducted in 6 replicates. Nitrification inhibitor dicyandiamide (DCD) was added (5% of total ammonium N content) to the pots with NH$_4$-N and NO$_3$-NH$_4$-N, to impede transformation of ammonium to nitrate.

2. **Measurements**

To observe potato stolon growth and tuber development, we sampled potato plants at 22, 29, and 85 DAE in 2010, and at 17, 21, and 97 DAE in 2011. The two early sampling times were at the tuber initiation stage and the last ones were at the tuber bulking stage. Potato seedlings were separated into leaves, stems, roots, stolons, and tubers. After rinsing, stolon number, stolon length, and stolon tip diameter were recorded using ruler and calipers. After measurement, all plant parts were oven-dried at 105ºC for 30 min, and then at 70ºC for 24 hr for dry weight measurements.

3. **Statistics**

The data were subjected to analyses of variance (ANOVA at a 0.05 level of significance) using SAS software.

### Results

1. **Influence of N forms on potato plant growth**

Table 1 shows how potato plant dry weight (DW) at tuber initiation and tuber bulking stages varied with the N form under sand culture conditions. At the tuber initiation stage, shoot DW of the plants supplied solely with NH$_4$-N (0 : 100 plot) were heavier than in the plants supplied solely with NO$_3$-N (100 : 0 plot), in both cultivars across 2
For root DW, there was no difference in root DW between the 100 : 0 and 0 : 100 plots in Kexin-1 at both growth stages and in both years. However, the 50 : 50 plot had lighter root DW than either the 100 : 0 or 0 : 100 plot in 2011. In cultivar Shepody, root dry weight was heavier in the 0 : 100 plot than in the 100 : 0 plot.

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### 2. Influence of N form on potato stolon growth

Stolon growth was measured for cultivar Kexin-1 across 2 years. In 2010, potato plants in 100 : 0 plot produced longer stolons than those in 0 : 100 plot at the tuber initiation stage. Similar trends were observed in 2011, where stolon length of seedlings in the 100 : 0 plot was 50% longer than in the 0 : 100 plot. Stolon length in 50 : 50 plot was shorter than that in NO₃-N treatment, but it was longer than that in the 0 : 100 plot (Table 2).

Stolon number per plant and the percentage of tuber-bearing stolons were also recorded for cultivar Kexin-1 at the tuber initiation stage across 2 years. These were significantly influenced by N form. Plants grown with NO₃-N produced more stolons than those in 0 : 100 plot in both years. Stolon number of plants with combined N sources was greater than those in the 0 : 100 plot, but smaller than those in the 100 : 0 plot (Table 2). No regular trend was observed in percentage of stolons bearing tubers (Table 2).

Table 3 shows that in 2011, plants of cultivar Kexin-1 produced thicker stolons in the 100 : 0 plot than in the 0 : 100 plot at both tuber initiation stage and tuber bulking stage.

### 3. Influence of N form on potato tuber growth

Table 4 shows how N form can affect tuber formation and growth. When sampled at 17 DAE in 2011, no tubers were found in cultivar Shepody grown under any N form; although some tubers were found in the cultivar Kexin-1 in the 100 : 0 plot. Four days later, tuber bearing was observed in all plots in both cultivars. However, the tuber dry weight of plants in the 100 : 0 plot was significantly heavier than in the other plots. In 2010, at 22 DAE, no tuber was found in any plot. Seven days later, tubers were observed in both 100 : 0 and 0 : 100 plots with no significant differences in DW.
At 85 DAE in 2010 and at 97 DAE in 2011, there were no longer any significant differences among N forms in terms of tuber dry weights per plant in both cultivars (Table 4). This is coincident with tuber yield at harvest (Table 5).

In cultivar Kexin-1, the tuber number per plant was greater in the 100 : 0 and 50 : 50 plots than in the 0 : 100 plot (Table 2).

**Discussion**

In spite of studies by several groups, a consistent conclusion about the effect of different forms of N on potato growth has not been reached. One reason may be the transformation of NH$_4^+$ to NO$_3^-$ by nitrification in the soil. Our results under sand culture with nitrification inhibitor in NH$_4^+$ treatments showed that potatoes (cultivars Kexin-1 and Shepody) shoot DW at the tuber initiation stage was lighter in the 100 : 0 plot than in 0 : 100 and 50 : 50 plots in both 2010 and 2011. However, at later stages, the differences in shoot DW between treatments began to decrease, and the shoot DW in the 100 : 0 plot was even heavier than in the 0 : 100 plot in some cases (Table 1).

This suggests that the response of potato growth to N form is complex. It is likely that the influences of N form on potato plant growth varied with the potato growth stage. Before or at tuber initiation, NH$_4^-$N might benefit shoot growth more than NO$_3^-$N, but later, shoot growth of plants in the NO$_3^-$N plot began to increase and gradually the difference between the treatments with NO$_3^-$N and NH$_4^+$N may disappear.

A field experiment by Sveensson (1962) with cultivar Bintje showed that stolon number increased with increasing amount of applied NH$_4^+$N; however, how stolon number varied with the amount of NO$_3^-$N was not included there. Osaki et al. (1995) found by field trial that NO$_3^-$ stimulates potato stolon growth and branching more than NH$_4^+$N. In our experiments across 2 years with 2 cultivars, both stolon number and stolon length per plant were greater under sole NO$_3^-$N treatment than under sole NH$_4^+$N (Table 2), which agrees with the findings of Osaki et al. (1995). Our finding that plants in the 50 : 50 plot produced more and longer stolons than in the 0 : 100 plot further confirms that NO$_3^-$N is favorable for potato stolon growth.
growth more than NH₄-N.

The growth of tuber-bearing stolons has been reported to be suppressed after tuberization (Kloosterman et al., 2007). However, growth was not suppressed completely in our experiments with either NO₃-N or NH₄-N (Table 3). At tuber initiation stage, there was no significant difference between the two N forms in terms of average stolon length. However, after a certain period of growth, the average stolon length in each treatment increased, and was greater in NO₃-N plot than in the NH₄-N plot (Table 3).

In the present experiment, the plants fed NO₃-N bore tubers earlier than those fed with NH₄-N (Table 4). Plants fed with NO₃-N produced more tubers than those fed with NH₄-N (Table 2). NO₃-N is therefore an efficient N form for potato tuber setting. However, the tuber yield per plant at harvest in NO₃-N treatment was not higher than those in NH₄-N treatment because of the lighter tuber weight (Table 5). It is well known that tuber growth greatly depends on assimilates from shoots, and therefore it cannot be concluded yet that potatoes favor NO₃-N, although NO₃-N has been shown to be beneficial for tuber setting. As described above, there is some evidence that NO₃-N could improve shoot growth after tuber initiation, although shoot growth prefers NH₄-N before the tuber initiation stage (Tables 1, 6). There is a possibility, therefore, that NO₃-N produces higher tuber yield by improving shoot growth and thus providing more assimilates after the tuber initiation stage. In dry land fields, inorganic N pool is mainly NO₃⁻ in soils, and the NH₄⁺ applied could be easily transformed into NO₃⁻. These soil characteristics could be beneficial to tuber initiation. However, a certain amount of NH₄-N fertilizer with nitrification inhibitor may be necessary because NH₄-N improves shoot growth at the early stage of potatoes (Table 1).

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