

## Paclobutrazol Enhances Minituber Production in Norland Potatoes

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**Abstract.** The effect of two plant growth regulators, paclobutrazol and kinetin, on minituber yield in greenhouse-grown “Norland” potatoes was investigated. Plants were treated with paclobutrazol at 450 mg/L, kinetin at 10 mg/L, or a combination of paclobutrazol at 450 mg/L + kinetin at 10 mg/L as single foliar applications at early stolon initiation. A set of plants sprayed with water served as the control. The experiment was conducted twice. In both cases, paclobutrazol nearly doubled the number of usable tubers/plant without affecting total tuber yield. Kinetin had no effect either on tuber number or tuber weight. Kinetin applied as a combination with paclobutrazol decreased the effectiveness of paclobutrazol on tuber number by 13–20%. Paclobutrazol treatments prolonged tuber dormancy by approximately 3 weeks. The results suggest that paclobutrazol treatment would be effective in enhancing potato minituber production under greenhouse conditions.

Minitubers are virus-free intermediate products used for field planting in pre-nuclear seed potato production. Potato plantlets raised in *in vitro* culture, stem cuttings, microtuber or minituber pieces are used as planting materials for minituber production under greenhouse conditions. Commercial production of minitubers using *in vitro* plantlets is limited by the high cost of production. Although the exact cost of minituber production is generally confidential, it is estimated that the cost varies from \$0.75 to \$1.00/minituber (D. R. Waterer, personal communication, 1995). The cost of minituber production can be reduced by increasing the number of minitubers/unit area.

The role of plant growth regulators on potato tuberization has been studied widely both *in vitro* and *in vivo*. Tuberization can be controlled by altering the hormonal balance of the plant. For example, cytokinins (Hussey and Satacey 1984, Palmer and Smith 1969, Pavlista 1993, Pavlista 1994, Pelacho and Mingo-Castel 1991, Wang and Hu 1982) and auxins (Mangat et al. 1984) were found to promote tuberization, whereas gibberellins (Menzel 1980, Vrengenhil and Struik 1989) and ethylene (Vrengenhil and van Dijk 1989) inhibited tuberization. Mauk and Langille (1978) assumed that cytokinins act as triggering agents for tuberization in potatoes, although Koda and Okazawa (1983) and Jameson et al. (1985) found that cytokinins had little effect on tuberization. Hammes and Nel (1975) suggested that tuberization in potato is controlled by a balance between endogenous gibberellins and the tuber-forming stimulus. Plants grown in noninductive environments, long days, constant high temperature, are characterized by a high content of gibberellins (Pont Lezica 1970, Railton and Wareing 1973). The threshold ratio between gibberellin and tuber stimulus may be achieved through the endogenous application of plant growth regulators to potato plants. Gibberellin biosynthesis inhibitors such as chlorocholine chloride (Dyson 1965, Gunasena and Harris 1971, Menzel 1980), B 995 (Bodlaender and Algra 1966), and tetcyclacis and triazole (Balamani and Poovaiah 1985, Langille and Helper 1992) have produced promising results for tuberization.

Paclobutrazol [(2*R*,3*R*+2*S*,3*S*)-1-(4-chlorophenyl) 4,4-dimethyl-2-(1,2,4-triazol-1-yl)-pentan-3-ol] (PTZ) is a triazole compound that inhibits shoot growth in a wide range of plant species including potatoes (Balamani and Poovaiah 1985), and its growth-retarding properties are largely attributed to its interference with gibberellin biosynthesis (Davis et al. 1993, Hedden and Graebe 1985). Recent studies show that PTZ significantly enhanced the tuberization in potato *in vitro* (Harvey et al. 1991, Simko

**Abbreviations:** PTZ, paclobutrazol; KIN, kinetin.

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1991, Simco 1993). Moreover, combined treatments of PTZ with kinetin (KIN) produced a synergistic effect on tuberization (Simko 1993). Various methods of application such as soil drench, foliar spray, and painting on the stem or leaves have been tested for PTZ (Barrett and Bartuska 1982, Greene and Murray 1983, Steffens et al. 1983, Wample and Culver 1983). Advantages of whole plant treatment include lower cost of production/tuber and the ability for growers to adopt this technology more easily over tissue culture techniques. However, information on the effect of PTZ and its interaction with cytokinins on minituber production in potatoes is lacking. The present study was conducted to investigate the effect of the foliar application of PTZ and its interaction effects with KIN (a cytokinin compound) on minituber number and weight of "Norland" potatoes under greenhouse conditions.

## Materials and Methods

Seed tubers (Elite III) of potato (*Solanum tuberosum* L. cv. Norland) purchased from a local seed grower were used for this study. Tuber pieces (15–20 g) from the eye area at the apical ends were planted in plastic pots (12.5 and 15.0 cm in diameter in experiments 1 and 2, respectively). Each pot was filled with Redi Earth, an artificial growing medium, and placed in a greenhouse. Potato plants were grown at  $23 \pm 2^\circ\text{C}/18 \pm 2^\circ\text{C}$  (day/night), and a 16-h daylength with  $600\text{--}700 \mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$  light intensity was maintained using Sylvania Cool White and Sylvania Gro-Lux fluorescent lamps. Plants were fertilized weekly with 20:20:20 (N:P:K) at 250 mg/L commencing the 2nd week of planting.

Based upon results of our preliminary studies, optimal timing and concentration of treatments were determined (unpublished data). At early stolon initiation, potato plants were foliar treated with PTZ (25%; ICI Chipman, Stoney Creek, ON, Canada) at 450 mg (a.i.)/L, KIN (6-furfurylaminopurine; Sigma Chemical Co., St. Louis, MO, USA) at 10 mg (a.i.)/L, and a solution of PTZ + KIN (450 mg/L PTZ and 10 mg/L KIN in the spray solution) using a hand atomizer to foliar runoff. Each solution was made using distilled water and contained a surfactant, Citrowett Plus (octylphenoxy-polyethoxy ethanol 50%, BASF, Canada Inc., Rexdale, ON) at 0.12% (v/v). The control plants were treated with water and surfactant to foliar runoff. Each treatment was repeated four (experiment 1) or five (experiment 2) times. Each replicate included five plants. The study was conducted twice (experiments 1 and 2).

The effects of treatment on haulm length, haulm dry weight, total tuber fresh weight/plant, and the number of tubers/plant were evaluated by executing ANOVA in a completely randomized design. Preplanned group comparisons were made for all observed variables to compare the PTZ treatment effect with the control, combined effect of PTZ plus KIN with their individual effects and PTZ versus KIN. The effect of treatments on seed tuber dormancy was also evaluated.

Following curing for 1 week under room temperature conditions ( $23 \pm 2^\circ\text{C}$ , in the dark), 20 tubers (experiment 1) or 30 tubers (experiment 2) from each treatment were stored in a cold cabinet at  $4^\circ\text{C}$ , relative humidity  $85 \pm 2\%$  in the dark for dormancy evaluation. The dormancy period was assessed by weekly observation of tuber samples. The dormancy of each stored tu-

ber was deemed to have ended when at least one vigorous sprout 2 mm long was present.

## Results

In both experiments treatments had a significant effect on the number of tubers/plant (Table 1). PTZ significantly increased the number of tubers/plant over the controls (Table 1). Depending upon the pot size, PTZ increased the number of tubers/plant by 65–83% over the controls. KIN had no effect on tuber number in either experiment. PTZ combined with KIN consistently produced a similar effect on the number of tubers/plant compared with the average effect of PTZ and KIN applied separately (Table 1).

Treatment had no significant effect on tuber fresh weight/plant compared with the control in either experiment (Table 1). PTZ applied with KIN produced a similar effect on tuber fresh weight when compared with the average effect of PTZ and KIN applied separately. PTZ and KIN produced a similar effect on total fresh weight of tubers/plant in both experiments (Table 1).

The preplanned treatment mean comparisons revealed that on average, treatments had no significant effect on haulm length of "Norland" potatoes compared with the controls in both experiments (Table 1). The effect of PTZ on haulm length was similar to the KIN treatments in experiment 1, but PTZ reduced haulm length significantly over the KIN treatment in experiment 2, in which larger pots were used (Table 1).

In both experiments, on average, chemical treatments had no effect on haulm dry weight compared with the controls (Table 1). The difference in haulm dry weight among treatments, however, was observed when plants were grown in the 15-cm pots in experiment 2. In experiment 2 PTZ significantly reduced haulm dry weight (Table 1). PTZ applied together with KIN reduced haulm dry weight compared with the average effect of PTZ and KIN applied separately (Table 1). PTZ significantly reduced haulm dry weight compared with the KIN treatments only in experiment 2.

Tuber dormancy responses from plants treated with KIN were similar to the control plants in both experiments (data not shown). On average, tubers from the control and the KIN treatments ended dormancy 100–103 days after harvest. Tubers from the treatments with PTZ, and KIN plus PTZ ended dormancy 122–126 days after harvest (data not shown).

## Discussion

The hormonal control of potato tuberization is a complex developmental process. However, methods of altering the hormonal balance of the plant

**Table 1.** Effect of paclobutrazol, kinetin, and combination paclobutrazol + kinetin on haulm length, haulm dry weight, number of minitubers per plant, and minituber fresh weight per plant of potato cultivar Norland under greenhouse conditions in 12.5-cm pots (experiment 1) and in 15-cm pots (experiment 2).\*

Treatment	Haulm length (cm)	Haulm dry weight (g)	No. of tubers/plant	Tuber fresh weight/plant (g)
<b>Experiment 1</b>				
Cont. (water)	17.0	1.6	2.3	47.5
PTZ (450 mg/L)	18.0	1.5	3.8	42.5
KIN (10 mg/L)	20.0	1.8	2.0	55.9
PTZ + KIN (450 + 10 mg/L)	17.0	1.8	3.3	48.2
<b>Experiment 2</b>				
Cont. (water)	33.4	2.5	6.0	118.6
PTZ (450 mg/L)	28.1	1.9	11.0	121.7
KIN (10 mg/L)	39.6	3.0	5.6	127.8
PTZ + KIN (450 + 10 mg/L)	28.7	2.0	8.8	122.6
<b>CV% for</b>				
Experiment 1	10.8	15.9	20.5	20.9
Experiment 2	14.8	11.1	16.7	8.5
<b>Contrasts</b>				
<b>Experiment 1</b>				
Cont. vs. Treatment	NS	NS	NS	NS
Cont. vs. PTZ	NS	NS	**	NS
PTZ + KIN vs. 2 (PTZ + KIN)	NS	NS	NS	NS
PTZ vs. KIN	NS	NS	**	NS
<b>Experiment 2</b>				
Cont. vs. Treatment	NS	NS	**	NS
Cont. vs. PTZ	NS	**	**	NS
PTZ + KIN vs. 2 (PTZ + KIN)	NS	**	NS	NS
PTZ vs. KIN	**	**	**	NS

\* PTZ, paclobutrazol; KIN, kinetin; NS, nonsignificant at  $p = 0.05$ ; \*\*, significant at  $p = 0.01$ ; (PTZ + KIN), paclobutrazol and kinetin combination.

and inducing tuberization are known (Tovar et al. 1985). The role of plant growth regulators has been studied widely in *in vitro* systems. Cytokinins are considered to be tuber-inducing hormones because the cytokinin content is higher in induced short day potato plants than in plants grown under long day conditions (Forsline and Langille 1975, Mauk and Langille 1978). However, it is still uncertain whether the increase in cytokinin content following transfer of the plants to inducing daylength is a precondition for, or a consequence of, tuber initiation (Sattlemacher and Marschner 1978). Several studies have shown that cytokinins promote tuber formation in potatoes *in vitro* (Hussey and Stacey 1984, Palmer and Barker 1972, Palmer and Smith 1969, Wang and Hu 1982). In contrast, *in vitro* studies by Tizio and Brian (1973) and Simko (1993) reported that KIN had no effect on tuberization in potatoes. Pelacho and Mingo-Castel (1991) suggested that under longer photoperiods, cytokinins would be metabolized, inactivated, or alternatively, an unidentified inhibitor could be formed. This hypothesis agrees with results of the present study in which

foliar-applied KIN had no effect either on plant growth or tuberization (Table 1). Reduced KIN uptake into whole plants or insufficient dosage of KIN may also have resulted in a lack of response to the KIN treatment.

A consistent reduction in shoot growth in potatoes was reported by Balamani and Poovaiah (1985) under greenhouse conditions and by Simko (1991, 1993) *in vitro* and in many other species. The present study revealed that despite the fact that PTZ decreased haulm length in experiment 2, the effect was not significant at  $p = 0.05$  (Table 1). The reason for this differing response was unclear.

PTZ nearly doubled the number of tubers/plant compared with the controls but had no effect on the total fresh weight of tubers/plant in either experiment (Table 1). This suggests that PTZ increased the tuber number at the expense of tuber size. According to Balamani and Poovaiah (1985), PTZ increased the dry weight of tubers/plant. It was not clear, however, whether this effect was the result of an increase in tuber number/plant or an increase of the average tuber size. Leaves treated with PTZ

alone or with KIN were darker green in color compared with control plants. This may be because the chlorophyll content was higher in the treated plants. The chlorophyll content per unit area was consistently higher in the PTZ-treated potato plants (Balamani and Pooviah 1985).

PTZ applied with KIN had no beneficial effect over the treatment with PTZ and KIN applied separately on any of the parameters measured in either experiment when plants were grown at a 16-h daylength (Table 1). In contrast, Simko (1993) reported a synergistic effect of PTZ with KIN on tuberization in potato *in vitro* at a 14-h daylength. In addition to the major differences between Simko's (1993) study (*in vitro*) and the present study (greenhouse), daylength difference may also have contributed to the lack of plant response to a combined application of PTZ and KIN.

Finally, pot size appeared to be an important factor in several of the responses including tuber number and haulm dry weight. The majority of these differences appeared to be due to the differential response to the PTZ treatments under different pot size. The reason for this difference in response in larger pots is not clear. Larger pots may have facilitated both above and below ground biomass production in potatoes resulting in larger plants with more canopy area and larger root system compared with the smaller pots. The larger canopy area and larger root system may have enhanced the uptake of the foliar-applied and soil-applied compound through runoff from the treatments, resulting in an enhanced effect of PTZ on plant and tuber growth.

Results also revealed that PTZ prolonged seed tuber dormancy by 3 weeks over the control. This effect would be of benefit for seed export, since air freight cost of potatoes is uneconomical. This is of particular relevance when potato cultivars such as "Norland" with shorter dormancy are shipped to foreign countries with longer than 3 months shipping period. Untreated tubers will typically begin sprouting at the end of this time. Under this situation, unnecessary sprouting in storage of the shipment may be avoided through prolongation of seed dormancy by the PTZ treatment.

In conclusion, our results indicate that PTZ is an effective plant growth regulator that can be used to increase minituber production in potatoes under greenhouse conditions. Pot size is an important factor, and containers should be at least 15 cm in diameter. However, more studies are in progress to determine the consistency of the effect on tuber production in other popular potato cultivars such as "Russet Burbank" and "Shepody" and to determine the carryover effect of PTZ on subsequent generations.

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