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Tuber Initiation in Hydroponically Grown Potatoes by Alteration of Solution pH

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Abstract. **Because tuberization in potatoes *(Solarium tuberosum* L.) reportedly is inhibited when stolons are immersed in liquid, this study was conducted to determine the effect of intermittent pH reductions of the nutrient solution on tuber induction of potatoes in solution culture. Tissue-culture potato plantlets were transplanted into solutions maintained at pH 5.5. The pH of the nutrient solution was changed to 3.5 and 4.0 for 10 hours on each of three dates (30, 35, and 40 days after transplanting). For the pH 3.5 treatment, tubers were observed first on day 42 and averaged 140 tubers per plant at harvest on day 54. For the pH 4.0 treatment, tubers were observed first on day 48 and averaged 40 tubers per plant at harvest. At a constant pH 5.5, tubers were observed on day 52 and averaged two tubers per plant at harvest. Plants with the intermittent pH 3.5 had smaller shoots and roots with shorter and thicker stolons compared to constant pH 5.5. With the intermittent pH 4.0, plants were of similar size, but stolons were shorter and slightly thickener compared to those from pH 5.5. Mineral composition of leaf tissues at harvest was similar for the three pH treatments. These results indicate that regulation of solution pH can be a useful technique for inducing tuberization in potatoes.**

Tuber initiation in potatoes occurs readily in solid matrix culture (Tibbitts and Cao, 1994; Tibbitts and Wheeler, 1987) and efficiently with nutrient film techniques (Wheeler et al., 1990). Although tuberization is inhibited when roots and stolons are immersed in solution or subjected to continuous mist culture (Tibbitts and Cao, 1994), tuberization under these hydroponic procedures can be promoted under certain stress conditions. In solution culture, N deficiency promoted tuberization (Krauss and Marschner, 1982). In mist culture, N deficiency and interruption of mist for 12 h induced rapid tuberization (unpublished data). Tubenzation does occur in solution culture when the container becomes filled with roots and stolons. Vreugdenhil and Struik (1989) suggested that the lack of mechanical resistance on the stolons prevents tuberization, yet efforts to place restriction on stolons have not promoted tuberization. This poor tuberization in solution culture limits the value of solution culture techniques for growing potatoes in controlled-environment facilities.

In a solution culture experiment, a low pH was induced accidentally for several hours in the solution, and small tubers were observed on the plants a few days later (unpublished data). This incident suggested that tuber initiation on plants grown in solution culture might be induced by short-term reductions in solution pH.

Potato plants adapt well to acidic conditions (MacLean et al., 1967; Smith, 1938), with optimum growth obtained between pH 5 and 6 (Cao and Tibbitts, 1994; McLean and Brown, 1984). Potatoes grown at low soil pH matured earlier and had a higher tuber : top ratio than those grown at normal pH (Smith, 1938). The ambient pH has been reported to significantly affect nutrient uptake and growth of many crop plants, particularly in solution cultures (Islam et al., 1980; Jariel et al., 1991). At very acidic pH, growth can be depressed through pH-induced indirect effects of toxicity or deficiency of certain elements or from direct root injury by excess hydrogen.

Our objective was to establish if intermittent pH reductions in solution culture could induce tuber formation in potatoes without producing significant injury to the plants.

Materials and Methods

The experiment was conducted in a walk-in growth chamber at the Univ. of Wisconsin–Madison. A recirculating solution culture system was used to grow 'Norland' potato plants (Fig. 1). The culture containers were clear acrylic. They were 25 cm long on each side with a 15-liter capacity. Container surfaces were covered with an opaque polyethylene plastic sheeting with a white outer surface and black inner surface to exclude light and minimize solution heating. A plastic screen was placed halfway into the solution to confine stolons and tubers within the top of the container. Around opening was made in the cover , of the container and a foam plug was inserted to support plants.

The nutrient solution was a modified half-strength Hoagland solution (Hammer et al.,1978) providing only nitrate N. The solution was supplied continuously to each container at 120±10 ml·min^{-1} through a tube at the center of the bottom of each container and drained out from a tube at the top of the container that maintained the solution level 1.5 cm below the cover. The solution was aerated with an air diffuser at the bottom of the container. The solution was recirculated through a 19-liter opaque-plastic jar. The solution in the jar was maintained at a constant

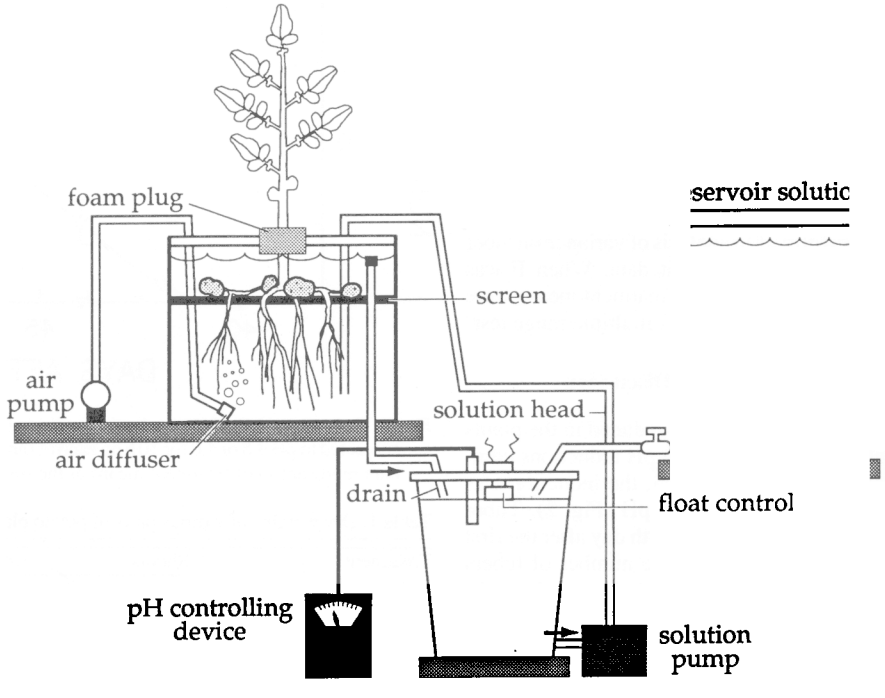

Fig. 1. Diagram of solution culture apparatus used in this experiment.

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level with an automatic float switch that controlled gravity flow from a 200-liter reservoir tank. Solution pH was maintained at 5.5 with an automatic pH controller using 0.5 N H_2SO_4. Conductivity of the solution was monitored every 3 or 6 days and adjusted when it varied more than 10% from the supply solution by releasing part of the solution in the plastic jar and adding concentrated stock solutions or distilled water, as required.

Plants were raised from micropropagated stem cuttings grown in sterile agar culture (Tibbitts and Wheeler, 1987). A uniform single plantlet, 21 days old, was transplanted into each culture container. Transplants were covered for 3 days with clear plastic cups to minimize transplant shock.

During the experiment, light period was 12 h provided with cool-white fluorescent lamps, and photosynthetic photon flux was 390 ± 20 $\mu mol \cdot m^{-2} \cdot s^{-1}$ at the top of the canopy. Temperature was 18 ± 0.3C during the light and 15 ± 0.6C during the dark periods. Relative humidity was constant at 75% ± 3% daily. The CO_2 level was ambient at ≈350 $\mu l \cdot liter^{-1}$.

Three times after transplanting (day 30,35, and 40), the solution recirculation was turned off for 10 h and the pH treatments (3.5,4.0, and constant 5.5) were established for that period. For the pH 3.5 and 4.0 treatments, the pH was lowered with an addition of 0.5 N H_2SO_4 and raised at the end of each 10-h period with 0.5 N KOH. The pH was adjusted to the appropriate level every hour during each of the 10-h periods. The design was a randomized complete block with three replications. Following the pH treatment at 30 days, tuber initiation and stolon morphology were examined visually, and tubers were counted every other day. Any enlargement of a stolon tip equivalent to twice the diameter of the stolon was recorded as a tuber.

Plants were harvested 54 days after being transplanted and separated into leaves, stems, tubers, and roots plus stolons. Plant material was oven-dried at 70C for 2 days for dry weight determinations. We sampled fully expanded green leaflets for mineral concentrations and analyzed them with inductively coupled plasma-emission spectrophotometry. Tissue N concentration was determined with the micro-Kjeldahl method (Jones et al., 1991). We performed an analysis of variance on tuber number and dry weight data. When F was significant at $P < 0.05$, treatment means were separated with Duncan's multiple range test.

Results and Discussion

Tuber initiation was induced in the plants subjected to intermittent pH reductions compared to constant pH 5.5; the induction was greatest with the lowest pH (Fig. 2). Tuber initiation started on the 10th day after the first pH 3.5 treatment, and the number of tubers increased significantly over the remaining 14 days. At harvest, 24 days after the first pH lowering, there were 140 tubers per plant. For the pH 4.0 treatment, tuber initiation was delayed 6 days compared to pH 3.5. The tuber initiation was later, and final tuber count (40 per plant) was significantly lower than with pH 3.5. For the constant pH 5.5, tuber initiation was not apparent until 2 days before harvest, and there were never more than two per plant. These results demonstrate that pH reductions at certain growth stages can induce tuber initiation in potatoes grown in solution culture.

The experimental variation in the time of induction and number of tubers was much greater with pH 4.0 than with pH 3.5 (Fig. 2). The data imply that pH 4.0 is close to a critical low pH for effective tuber induction in potatoes. In the previous accidental pH reduction (below pH 4.0), tuber initiation was seen at 4 days after pH reduction, compared to 10 days in this study. The plants in that study were at a more mature stage (≈ 40 days after transplanting) than in this study (30 days after transplanting). Thus, we suspect that in our study the first pH change was imposed before the plants were in an inductive state that would permit tuber initiation on stolons. However, it should be recognized that plant maturation rate, and thus effective time for acid treatment, would vary with environmental conditions.

Growth of shoots, roots, and stolons was affected by the intermittent pH reductions. Shoot and root dry weights were lower and tuber dry weight was higher with pH 3.5 than with pH 4.0 or 5.5 (Table 1). However, there was no evidence of root necrosis at pH 3.5. Stolon dry weight did not differ significantly among the three pH treatments, yet the morphology of stolon development differed. With intermittent pH reduction, primary stolons appeared to be shorter and thicker than with continuous pH 5.5. This effect was more pronounced at pH 3.5 than at pH 4.0. Also, the stolons were pigmented (purple) with pH 3.5, were partially pigmented with pH 4.0, and had no pigmentation with pH 5.5. These results suggest that short, thick, and pigmented stolons are associated with strong induction to tuberize. The purple pigmentation on stolons may be associated only with the red-skinned potato cultivars such as 'Norland'.

Brown spots (≈1 mm in diameter) were observed on fully expanded leaflets on the upper-middle canopy of potato plants 1 day after pH reduction to 3.5, although the leaves remained green and presumably photosynthetically active. Following the second and third pH reductions to 3.5, the fully expanded leaflets also showed some additional brown spots, but less than after the first pH change. At pH 4.0 and 5.5, plants grew normally without the leaf spotting symptom seen with pH 3.5.

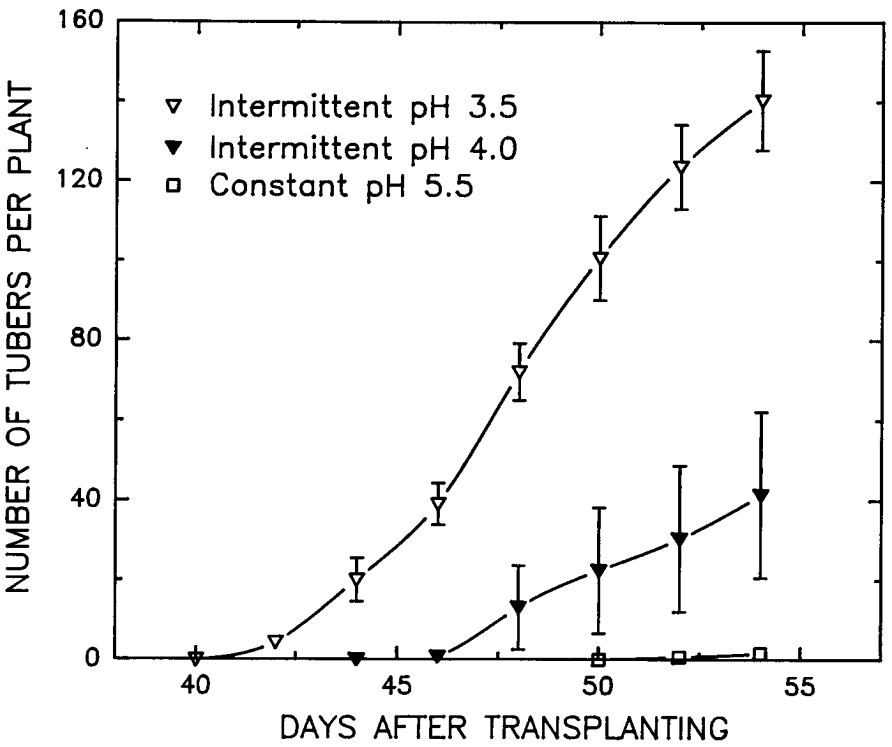

Fig. 2. Time course response of tuber initiation to three pH treatments. For the intermittent pH, plants were subjected to pH 3.5 or 4.0 for 10 h on each of three dates (30, 35, and 40 days after transplanting). Vertical bars represent SE of treatment means at the times noted.

Table 1. Dry weights of various parts of potato plants grown for 54 days with one of three pH treatments.

Treatment (pH)	Shoots	Stolons	Roots	Tubers	Total
			g/plant		
Intermittent 3.5[z]	26.7 b[y]	4.9 a	3.5 b	6.6 a	41.7 b
Intermittent 4.0[z]	54.9 a	5.7 a	5.2 a	1.0 b	66.8 a
Constant 5.5	48.5 a	4.2 a	5.0 a	<0.1	57.8 a

[z]Plants were subjected to pH 3.5 or 4.0 for 10 h on each of three dates (30, 35, and 40 days after transplanting).
[y]Mean separation in each column by Duncan's multiple range test at $P \leq 0.05$.

Mineral analysis of leaf samples at harvest time indicated that all tissue nutrients at pH 3.5 and 4.0 were within the normal range of concentrations for healthy growth of potatoes (data not shown); however, concentrations of certain nutrients were affected at pH 3.5. Tissue concentrations of Mn and Fe increased 40% and those of N and P decreased 10% with pH 3.5 compared to pH 4.0 and 5.5. Enhanced Mn accumulation in the leaf tissues with pH 3.5 may have resulted in the interveinal brown spots on leaves following the pH reductions. This presumed Mn toxicity symptom was consistent with a previous report on potatoes by Marsh and Peterson (1990). However, it is questionable if the changed nutrient concentrations in leaf tissues with pH 3.5 affected particular physiological processes to promote tuberization; pH 4.0-treated plants exhibited no significant nutrient changes, yet were also induced to tuberize. In a previous study, withholding N induced tuberization (Krauss and Marschner, 1982), but tuberization was stimulated only after N had been withheld for several days. In our study, N was always provided to the plants, and the treatment period with reduced pH was only 10 h.

Our results demonstrate that shot--term reductions of solution pH can significantly promote tuber initiation on potato plants grown with stolons immersed in liquid. Our data also suggest that for rapid and consistent tuber induction the intermittent pH levels need to be < 4.0. Apparently with the low pH treatments, assimilate partitioning to shoots and roots was restricted and directed toward stolon development and tuber initiation. This solution-pH control procedure may be use to produce many small tubers for plant propagation.

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