원예과학기술지 19(4):459-464. 2001

Effect of IAA and Zeatin Riboside on Plantlet Induction from Leaf Disks of *Solanum tuberosum* L. and Variation of Regenerated Plants

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ABSTRACT Leaf disks from cultivar 'Kennebec' and one selection line (ND 860-2) were cultured on Murashige-Skoog medium with various combinations of indole acetic acid (IAA) and zeatin riboside. Shoots, roots and callus were induced at various combinations of plant growth regulator levels. The medium containing 3.5 mg · L^{-1} IAA and 4.0 mg · L^{-1} zeatin riboside produced the most plantlets. Rooted regenerants were grown in the greenhouse. The growth of regenerated plants obtained from the MS medium supplemented with 7.0 mg · L^{-1} IAA and 3.0 mg · L^{-1} zeatin riboside was significantly greater than those grown from nodal expalnts. In ND 860-2, a leaf chimera with chlorophyll deficient (light yellow) sectors was found in plants regenerated from leaf disks (grown on MS medium supplemented with 3.5 mg · L^{-1} IAA and 3.0 mg · L^{-1} zeatin riboside) but not in plants grown from nodal explants. The phenotypic variability was also observed in the tuber number, size and weight.

Additional key words: characteristics, organogenesis, phytohormone, somaclonal variation, tuber

Introduction

Plants may be propagated rapidly in vitro by using tissue of the leaf, stem or root as an explant. Of the many factors that influence organogenesis in vitro, the most important single factor seems to be plant plant growth regulator (Tran, 1981). Skoog and Miller (1957) showed that different types of organs or tissues can be obtained by varying the concentrations of auxins and cytokinins in the culture medium. Following the work of Skoog and Miller (1957) there have been many demonstrations of effect of hormonal interactions, particularly between auxins and cytokinins, on morphogenesis in a diverse range of plant material (Murashige, 1974).

Regeneration of plants from leaf disks has been accomplished with several potato cultivars (Webb et al., 1983). Shoots, roots, and callus were formed from leaf disks of six cultivars of potato when grown in vitro on the basal medium of MS supplemented wit IAA (5.69×10^{-6} M · L^{-1}) and BAP (4.44×10^{-6} M · L^{-1}). The potato can be propagated by the formation of adventitious shoots on explants excised form a compound leaf (petiole, rachis and leaflets) and the subsequent rooting of subcultured shoots (Roset and Borkelmann, 1976).

Recent work has been concerned with assessing the optimal hormonal composition of nutrient media for plantlet regeneration from leaf tissue. Plants have been obtained from isolated leaf mesophyll protoplasts of potato using the MS medium with 0.5 mg · L^{-1} zeatin and 0.1 mg · L^{-1} IAA (Kozukue et al., 1999; Shepard and Totten, 1977). Zeatin riboside was also found to be effective in regenerating plants from leaves of potato (Jacobsen, 1977). Jarrett et al. (1980) found kinetin, 2iP and BAP were essential for explant survival, while BAP (3.0 mg · L^{-1}) was most efficient in promoting shoot initiation. Auxin (IAA and IBA) did not stimulate shoot initiation, however, a low concentration (0.3 mg · L^{-1}) of NAA stimulated both explant survival and the number of shoots produced. In tomatoes, shoot regeneration was obtained by growing the leaf disks on MS medium supplemented with 2×10^{-5} M · L^{-1} IAA and 2×10^{-6} M · L^{-1} zeatin riboside (Gavazzi et al., 1987). Okazawa et al., (1967) found that a relatively low concentration of auxins in the medium was required for root formation in tuber callus, and IAA was markedly superior to any other auxin in root forming activity.

Plants regenerated from tissue cultures or isolated protoplasts derived from somatic tissue may show changes in phenotype. Such somaclonal variation has been suggested as a useful source of variability for plant breeders (Evans, 1989; Larkin and Scowcroft, 1981; Mohan Jain et al, 2000). The basis of variation may involve gene mutation or changes in chromosome number or structure which were either present in the mesophyll cells or induced during the regeneration process (Karp et al., 1982). Sree

※ Received for publication 9 November 2001. Accepted for publication 23 November 2001.

Ramulu et al. (1983) reported the variant plants from protoplast-derived plants of tetraploid potato cultivar 'Bintje' had alterations in growth and vigor, and leaf and stem characteristics. The morphologically abnormal regenerants showed abnormal ploidy levels at a high frequency.

In the case of regeneration from cultured explants, such as pieces of leaf, stem or tubers, reports indicated that variation was present (Mohan Jain et al., 2000; van Harten et al., 1981). Evans et al., (1986) suggest that the technique of regeneration from explant cultures of potato could be used to produce somaclonal variants. This method offers the possibility of effecting significant and permanent improvements in existing potato cultivars. Alteration in chromosome number occurs less frequently using this method (Kaeppler et al., 2000; Mohan Jain et al., 2000; Wheeler et al., 1985).

The objectives of this study were to define phytohormone concentrations for the formation of plantlets directly from leaf disks of *Solanum tuberosum* L. and subsequently to study of the variation of regenerated plants.

Materials and Methods

Plant material

Cultivar 'Kennebec' and one selection line (ND 860-2) of *Solanum tuberosum* L. ($2n = 4x = 48$) were used as the source material for leaf tissue culture throughout the study. Plants obtained from the meristem culture were cut to give stem nodal segments formed by a part of the stem bearing one leaf. Stem nodal segments were cultured aseptically on MS (Murashige and Skoog, 1962) basal medium containing 0.4 mg · L^{-1} thiamine HCl, 100.0 mg · L^{-1} myo-inositol, 20.0 g · L^{-1} sucrose, 0.2 mg · L^{-1} GA_3, 0.05 mg · L^{-1} NAA, 2.0 mg · L^{-1} calcium pantothenate and 8.0 g · L^{-1} Difco Bacto-agar at pH 5.6. Axillary shoots were formed within 14 days. Complete plantlets with expanded leaves including roots were obtained in 4 - 6 weeks. Plantlets were maintained in vitro by transferring nodal segments to fresh medium every 3 month.

Experiment 1. The effects of IAA and zeatin riboside concentration on leaf disk explant growth and plantlet induction

This experiment was studied in an arrangement of six levels of IAA (0, 0.1, 0.5, 1.0, 3.5, and 7.0 mg · L^{-1}) and seven levels of zeatin rivoside (0, 0.1, 0.5, 1.0, 2.0, 3.0, and 4.0 mg · L^{-1}). Leaves from in vitro grown plants were excised, and 5 mm disks were cut from them using a cork borer. The leaf disks were placed onto MS salts medium, with various combinations of IAA and zeatin riboside, containing a medium of MS salts containing 0.4 mg · L^{-1} thiamine HCl, 100.0 mg · L^{-1} myo-inositol, 30.0 g · L^{-1} sucrose and 8.0 g · L^{-1} Difco Bacto-agar at pH 5.8. IAA and zeatin riboside were filter-sterilized and added after autoclaving. The cultures were grown at 24°C and under 16 hrs light provided by one 40 Watt cool white and one 40 Watt Grow Lux fluorescent light bulb (54 μE m^2sec^{-1}). After 8 weeks, the formation of callus, roots, number of shoots, and explant weight were measured.

Experiment 2. Variation of regenerated plants

The occurrence of variations in *Solanum tuberosum* L. plants regenerated directly from leaf disks was compared with plants propagated from nodal cuttings. For shoot induction, the same method was used as described for experiment 1 except that 3.5 and 7.0 mg · L^{-1} IAA and 3.0 mg · L^{-1} zeatin riboside were used as the phytohormone component. After shoot differentiation, the shoots were excised and rooted on 0.5 strength MS medium containing 15.0 g · L^{-1} sucrose, 0.1 mg · L^{-1} IAA, 0.01 mg · L^{-1} kinetin and 7.0 g · L^{-1} Difco Bacto-agar at pH 5.8. In in vitro cultures, the cultures were grown at same condition as experiment 1. Rooted plantlets were rinsed in running water to remove the agar and planted into peat pots containing a commercial soil mix (Jiffy Mix). The plants were then placed on a heated bench (24°C) under intermittent mist for 48 hours. The plants were grown on a heated bench for 2 - 3 weeks and then transferred into 10 cm pots containing a commercial soil mix (Sunshine Mix #1). Plants were grown in the greenhouse until tuber development was completed. In the greenhouse, plants were grown under a daylength of 16 hours supplemented by 1000 watt metal halide lamps placed 1.5 m above the plants (150 μE $m^{-2}sec^{-1}$). The plants were fertilized with 200 ppm N-P-K with every watering after being transferred to 10 cm pots. To analyze the type and extent of phenotypic variation among the regenerated plants, the following characters were studied: plant height, leaf size and shape, tuber number, tuber weight and tuber size. A completely randomized design was used.

The data are reported as an average of 8 replications (Experiment 1) and 10 replications (Experiment 2) in each treatment. All data means were separated using least significant difference at 5 % level.

Results and Discussion

Various combinations of IAA and zeatin riboside were tested. Leaf disks of ND 869-2 and 'Kennebec' formed callus, roots and shoots with leaves after 8 weeks of culture. The leaf disks began to expand after their placement in culture and subsequently curled either toward or away from the medium surface. Callus was first produced on explants in the injured region within 4 weeks. Shoots with leaves were regenerated after 7 - 8 weeks, and roots were produced from the same leaf explants.

After 60 days in culture, none of the explants on phytohormone-free medium had callus, roots or shoots. In the absence of both phytohormones, all of the explants turned brown and died. IAA caused limited callus development and abundant root formation. Although root formation was induced by IAA, the frequency of roots formed were found to be dependent upon the

concentration of IAA used. Higher concentration of IAA gave better root formation. The best shoot regeneration medium contained 3.5 mg · L^{-1} IAA for ND 860-2 and 'Kennebec' (Table 1 and 2). Addition of zeatin riboside to the medium resulted in stimulation of explant growth and plantlet induction. Shoots were initiated from leaf disks on media which contained 1.0 mg · L^{-1} or higher levels of zeatin riboside. The best range of zeatin riboside levels for plantlet induction and explant growth was 3.0 to 4.0 mg · L^{-1}. Lam (1977) reported that the addition of zeatin as cytokinin resulted in the formation of more fully developed shoots.

For ND 860-2 (Table 1) and 'Kennebec' (Table 2), there was a significant IAA and zeatin riboside interaction for callus and shoot development. Zeatin riboside induced green callus, but in

Table 1. Effect of IAA and zeatin riboside concentration on explant growth and plantlet induction on leaf disk explants of ND 860-2.

IAA (mg · L^{-1})	Zeatin riboside (mg · L^{-1})	Shoot number	Formation[z]		Explant weight (g)
			Callus	Root	
0.0	0.0	0.00 g[y]	0.00 j	0.00 g	0.13 j
	0.1	0.00 g	0.00 j	0.00 g	0.12 j
	0.5	0.00 g	0.00 j	0.00 g	0.18 j
	1.0	0.00 g	0.00 j	0.00 g	0.24 j
	2.0	0.25 fg	0.00 j	0.00 g	0.26 j
	3.0	0.00 g	1.50 efgh	0.00 g	0.56 ij
	4.0	1.50 def	3.00 bc	1.25 def	1.80 def
0.1	0.0	0.00 g	0.00 j	0.00 g	0.14 j
	0.1	0.00 g	0.00 j	0.00 g	0.13 j
	0.5	0.00 g	0.50 ij	0.00 g	0.23 j
	1.0	0.00 g	0.00 j	0.00 g	0.31 j
	2.0	0.00 g	0.75 hij	0.00 g	0.33 j
	3.0	0.13 fg	1.75 efg	0.25 fg	0.73 hij
	4.0	1.13 defg	2.25 cde	1.25 def	2.01 def
0.5	0.0	0.00 g	0.00 j	0.00 g	0.19 j
	0.1	0.00 g	0.00 j	0.00 g	0.30 j
	0.5	0.00 g	0.00 j	0.00 g	0.31 j
	1.0	0.00 g	0.75 hij	0.25 fg	0.36 j
	2.0	0.38 efg	2.00 def	0.25 fg	0.93 ghij
	3.0	1.75 cde	2.75 bcd	1.25 def	1.92 def
	4.0	2.50 cd	3.00 bc	2.00 bcd	2.08 def
1.0	0.0	0.00 g	0.00 j	0.00 g	0.32 j
	0.1	0.00 g	0.00 j	0.00 g	0.33 j
	0.5	0.00 g	0.00 j	0.00 g	0.45 ij
	1.0	2.50 cd	2.25 cde	1.50 de	2.15 cde
	2.0	2.88 c	2.25 cde	1.75 cde	2.54 cd
	3.0	1.38 defg	2.25 cde	1.00 defg	1.62 efg
	4.0	1.38 defg	2.00 def	0.75 efg	1.55 efgh
3.5	0.0	0.00 g	0.00 j	2.00 bcd	0.40 j
	0.1	0.00 g	0.00 j	2.00 bcd	0.53 ij
	0.5	0.00 g	0.50 ij	1.75 cde	0.69 ij
	1.0	0.00 g	1.00 ghi	1.50 de	0.55 ij
	2.0	2.38 cd	3.00 bc	1.50 de	2.24 cde
	3.0	7.75 a	4.00 a	3.50 a	5.14 a
	4.0	8.00 a	4.00 a	3.75 a	5.05 a
7.0	0.0	0.00 g	0.00 j	1.25 def	0.35 j
	0.1	0.00 g	0.00 j	1.50 de	0.30 j
	0.5	0.00 g	0.00 j	2.00 bcd	0.41 j
	1.0	0.63 efg	1.25 fghi	1.75 cde	1.28 fghi
	2.0	0.75 efg	1.50 efgh	1.75 cde	1.28 fghi
	3.0	5.13 b	3.50 ab	3.00 ab	3.68 b
	4.0	5.13 b	3.00 bc	2.75 ab	2.95 bc

[z]Formation score: 0 = none; 2 = some; 4 = abundant.

[y]Mean separation within columns for each plant growth regulator level by LSD, 5% level.

Table 2. Effect of IAA and zeatin riboside concentration on explant growth and plantlet induction on leaf disk explants of 'Kennebec'.

IAA (mg · L^{-1})	Zeatin riboside (mg · L^{-1})	Shoot number	Formation[z]		Explant weight (g)
			Callus	Root	
0.0	0.0	0.00 k[y]	0.00 j	0.00 h	0.14 l
	0.1	0.00 k	0.00 j	0.00 h	0.14 l
	0.5	0.00 k	0.00 j	0.00 h	0.17 kl
	1.0	0.00 k	0.00 j	0.00 h	0.21 kl
	2.0	0.25 ijk	0.00 j	0.00 h	0.22 kl
	3.0	1.00 ghijk	2.00 efg	1.00 efg	1.35 hij
	4.0	2.63 cdef	2.75 bcde	1.50 def	2.38 cdefg
0.1	0.0	0.00 k	0.00 j	0.00 h	0.13 l
	0.1	0.00 k	1.00 hi	0.50 fgh	0.74 jkl
	0.5	0.00 k	0.50 j	0.00 h	0.20 kl
	1.0	0.75 hijk	1.25 ghi	0.50 fgh	1.00 ijk
	2.0	1.50 efghij	2.25 def	1.00 efgh	1.71 ghi
	3.0	2.75 cdef	2.25 def	1.75 cde	2.59 bcdef
	4.0	0.75 hijk	2.25 def	1.25 defg	1.28 hij
0.5	0.0	0.00 k	0.00 j	0.50 fgh	0.24 kl
	0.1	0.00 k	0.00 j	0.00 h	0.35 kl
	0.5	0.00 k	0.00 j	0.00 h	0.32 kl
	1.0	0.00 k	1.00 hi	0.00 h	0.55 ikl
	2.0	1.00 ghijk	2.50 cdef	0.50 fgh	1.33 hij
	3.0	2.75 cdefg	2.75 bcde	1.25 defg	2.12 cdefgh
	4.0	3.50 cd	3.00 bcd	1.75 cde	2.83 bc
1.0	0.0	0.00 k	0.00 j	0.25 hg	0.27 kl
	0.1	0.00 k	0.00 j	0.00 h	0.29 kl
	0.5	0.125 kj	1.00 hi	0.00 h	0.75 ikl
	1.0	2.38 cdefg	3.00 bcd	2.75 abc	2.22 cdefg
	2.0	2.13 cdefg	2.25 def	1.75 cde	1.81 efghi
	3.0	2.88 cde	3.00 bcd	2.25 abcd	2.11 cdefgh
	4.0	1.75 efgh	2.50 cdef	1.75 cde	1.95 defgh
3.5	0.0	0.00 k	0.00 j	2.00 bcde	0.50 jkl
	0.1	0.00 k	0.00 j	2.25 abcd	0.41 kl
	0.5	0.00 k	0.00 j	1.50 def	0.35 kl
	1.0	0.00 k	1.00 hi	1.50 def	0.62 jkl
	2.0	3.75 bc	3.25 abc	3.00 ab	2.74 bcd
	3.0	5.00 ab	4.00 a	3.25 a	3.37 ab
	4.0	5.88 a	4.00 a	3.00 ab	3.87 a
7.0	0.0	0.00 k	0.00 j	1.50 def	0.42 jkl
	0.1	0.00 k	0.00 j	1.50 def	0.41 kl
	0.5	0.00 k	0.00 j	1.50 def	0.33 kl
	1.0	1.38 fghijk	1.75 fgh	2.00 bcde	1.35 hij
	2.0	1.63 efghi	2.50 cdef	2.25 abcd	1.78 fghi
	3.0	2.88 cde	3.50 ab	3.25 a	2.63 bcde
	4.0	2.38 cdefg	3.25 abc	3.00 ab	2.63 bcde

[z]Formation score: 0 = none; 2 = some; 4 = abundant.

[y]Mean separation within columns for each plant growth regulator level by LSD, 5% level.

the absence of IAA (auxin) failed to promote organogenesis except at the highest concentration (4.0 mg · L^{-1}). Athough plantlets were induced at the highest concentration (4.0 mg · L^{-1}) of zeatin riboside alone, generally two phytohormones were required for regeneration of plantlets. Shoot development was enhanced by the addition of IAA, however, the combination of the high level (7.0 mg · L^{-1}) of IAA with zeatin riboside inhibited shoot formation.

Shoot differentiation appears to be a function of zeatin roboside (cytokinin) activity since IAA (auxin) alone did not initiate shoot development. Moreover, leaf explant responded differently to different levels of IAA. The shoot development was not affected by low concentration (<0.5 mg · L^{-1}) of IAA and zeatin riboside. This might be due to auxin : cytokinin ratio which is unfavorable to this process.

This experiment indicated that an equal ratio of IAA and zeatin riboside was effective for direct shoot induction from leaf disks. The difference in regeneration capacity and mode of regeneration at higher and lower concentrations may be explained on the basis of variation in the endogenous levels of these phytohormones in leaf tissue (Nehra and Stushnoff, 1989). Therefore, for the successful induction of normal plantlet formation from leaf disks in vitro, levels of exogenous and endogenous phytohormones need to be taken into consideration.

The shoots seemed to originate from the proliferating cells rather than the original cells of the leaf. The histological study of shoot induction in leaf disk propagating of potato needs further study. Plants regenerated from callus and cell suspension cultures may include a varying proportion showing structural or physiological abnormalities, according to the origin and age of cultures (Yeoman, 1986).

Plantlet formation from potato leaf disks was dependent on the phytohormonal composition of the medium employed and the genotype used. Phytohormone interactions tested in this experiment helped in the identification of phytohormone levels for use in plantlet regeneration.

One potato cultivar 'Kennebec' and one breeding selection, ND 860-2, were used for this study. Regenerated plants from the shoot induction medium containing either 3.5 mg · L^{-1} IAA+3.0 mg · L^{-1} zeatin riboside or 7.0 mg · L^{-1} IAA+3.0 mg · L^{-1} zeatin riboside were grown in the greenhouse to produce tubers. Plants propagated in vitro from nodal cuttings were grown under the same conditions as control plants. Regenerated plants showed evidence of phenotypic variation (Table 3). Significant differences were observed in the response of different cultivars and phytohormones. The growth of regenerated plants obtained from the MS medium supplemented with 7.0 mg · L^{-1} IAA and 3.0 mg · L^{-1} zeatin riboside was significantly greater than those grown from nodal explants. But leaf length and leaf shape (width/length) were not significantly different. In ND 860-2, leaf width of regenerated plants from the medium containing 7.0 mg · L^{-1} IAA and 3.0 mg · L^{-1} zeatin riboside was significantly greater than those obtained from the MS medium supplemented with 3.5 mg · L^{-1} IAA and 3.0 mg · L^{-1} zeatin riboside and nodal explants. These phenotypic changes may be due to changes in karyotype. Hermsen et al. (1981) reported that phenotypic variation observed in explant-derived shoots of diploids mainly concerned changes in the ploidy level and morphological changes which were probably the result of mutated genes or deletions.

Plant regenerated from leaf disks can show differences in a range of characters, which is somaclonal variation. Vegatative characteristics such as leaf shape were also different in some regenerated plants. They may have existed as genetic differences in leaf cells, thus, probably leading to expression of the trait. Wenzel et al. (1979) and Sree Ramulu et al. (1983) also reported variant plants with alterations in growth and leaf characteristics.

In ND 860-2, a leaf chimera with chlorophyll deficient (light yellow) sectors was found in plants regenerated from leaf disks (grown onto MS medium supplemented with 3.5 mg · L^{-1} and 3.0 mg · L^{-1} zeatin riboside) but not in plants grown from nodal explants. Perhaps the participation of variant cells, in addition to the normal cells, can lead to the formation of leaf chimeras. These plants had a higher tuber number (7 – 8) than normal plants.

The phenotypic variability was also observed in the tuber number, size and weight (Table 4). In ND 860-2, the tuber

Table 3. Observations of plant height and leaf morphology of ND 860-2 and 'Kennebec' plants regenerated directly from leaf disks as compared with plants propagated from nodal cuttings.

IAA (mg · L^{-1})	Zeatin riboside (mg · L^{-1})	Explant source	Plant height (cm)	Leaf width (cm)	Leaf length (cm)	Width/ length
ND 860-2						
0.0	0.0	node	25.50 b[z]	4.47 b	5.46 a	0.82 a
3.5	3.0	leaf disk	29.63 ab	4.44 b	5.21 a	0.85 a
7.0	3.0	leaf disk	32.25 a	5.14 a	5.63 a	0.92 a
'Kennebec'						
0.0	0.0	node	22.58 b	4.72 a	5.95 a	0.80 a
3.5	3.0	leaf disk	26.30 ab	4.81 a	5.81 a	0.84 a
7.0	3.0	leaf disk	28.80 a	4.89 a	5.73 a	0.86 a

[z]Mean separation within columns for each cultivars by LSD, 5% level.

Table 4. Observations of ND 860-2 and 'Kennebec' tuber characters from plants regenerated directly from leaf disks as compared with plants propagated from nodal cuttings.

IAA ($mg \cdot L^{-1}$)	Zeatin riboside ($mg \cdot L^{-1}$)	Explant source	Tuber number	Tuber width (cm)	Tuber length (cm)	Tuber weight (g)
ND 860-2						
0.0	0.0	node	4.60 b[z]	2.34 a	2.54 a	8.84 a
3.5	3.0	leaf disk	6.63 a	2.02 b	2.26 a	6.77 b
7.0	3.0	leaf disk	6.56 a	2.31 a	2.55 a	8.20 a
'Kennebec'						
0.0	0.0	node	5.11 b	2.20 a	2.41 a	7.39 a
3.5	3.0	leaf disk	7.10 a	1.84 b	2.07 b	4.91 b
7.0	3.0	leaf disk	7.80 a	1.99 b	2.21 b	5.44 b

[z]Mean separation within columns for each cultivars by LSD, 5% level.

number from plants regenerated from leaf disks was significantly greater than from plants from nodal explants. The tuber width and weight of regenerated plants obtained by growing onto MS medium supplemented with 3.5 $mg \cdot L^{-1}$ IAA and 3.0 $mg \cdot L^{-1}$ zeatin riboside was significantly reduced compared to those derived from in vitro regeneration on MS medium supplemented with 7.0 $mg \cdot L^{-1}$ IAA and 3.0 $mg \cdot L^{-1}$ zeatin riboside and those derived from nodal explants. In 'Kennebec' the tuber number of regenerated plants from leaf disks was significantly increased compared to those from nodal explants but tuber width, length and weight were significantly reduced.

Rietveld (1988) found that regenerated plants from tuber disks of 'Kennebec' had more elongated tubers, higher total tuber number and weight. In this experiment, this may have been true in tuber number, but not in tuber length and weight.

Regenerants from 'Red Pontiac' had tubers with a range of skin colors from pink to red (data not shown). But plants regenerated from nodal cutting plants had the usual red color. This cultivar is a chimera of 'Pontiac' and the tuber skin color would vary depending on the cell layer from which the plantlet was derived. Mohan Jain et al. (2000) and Sree Tamulu et al. (1983) suggested that variation may be due to somatic segregation of chimeras resulting from gene mutations or chromosome structural rearrangements in only part of the regenerated plant.

The results show a range of phenotypic variation among regenerants from leaf disks. This phenomenon has previously been reported in plants regenerated from protoplast or explant cultures. Plant regeneration from leaf explant cultures is quicker and easier than protoplast culture and can be used to study somaclonal variation. This somaclonal variation has been suggested as a useful source of variability for plant breeders (Kaeppler et al., 2000; Larkin and Scowcroft, 1981; Mohan Jain et al., 2000). However, it is not possible to draw firm conclusions from a single generation of plants grown in greenhouse where such characters may have been altered by the artificial conditions.

IAA와 Zeatin Riboside가 감자의 엽절편체로부터의 식물체 유기 및 재분화개체의 변이에 미치는 영향

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초 록

감자 품종 'Kennebec'과 육종 계통 ND 860-2의 잎 절편을 식물생장조절제 indole acetic acid(IAA)와 zeatin riboside의 다양한 조합이 포함된 Murashige-Skoog 배지에서 배양하였다. 신초, 뿌리 및 캘러스가 다양한 식물생장조절제 조합으로부터 유기되었으며 두 품종 공히 3.5$mg \cdot L^{-1}$ IAA와 4.0 $mg \cdot L^{-1}$ zeatin riboside를 포함한 배지에서 가장 많은 소식물체를 유기하였다. 뿌리가 유기된 재분화개체를 온실에서 재배한바 7.0$mg \cdot L^{-1}$ IAA와 3.0$mg \cdot L^{-1}$ zeatin riboside이 포함된 배지에서 재분화된 개체의 생육이 다른 처리구에 비해 유의성 있게 좋았다. ND 860-2의 경우 3.5$mg \cdot L^{-1}$ IAA와 3.0$mg \cdot L^{-1}$ zeatin riboside의 배지로부터 재분화된 개체의 잎에서 chlorophyll이 결핍되어 부분적으로 옅은 황색 부분이 나타나는 chimera를 발견하였으며 재분화개체로부터 tuber 수, 크기 및 무게 등 표현형의 변이가 나타났다.

추가 주요어 : 기관형성, 식물호르몬, 체세포변이, 괴경, 형질

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