1985 for the two sets was 1.096, as compared to 1.091 for the unselected offspring (Table 1), a significant difference (t = 5.74, df = 210).

The mean specific gravity of the selected offspring in 1987 was 1.092, as compared to 1.087 for the unselected offspring (Table, 1), again a significant difference *(t* = 7.94, df = 207).

In both cycles of selection for tuber characteristics, the mean specific gravity of the selected clones was significantly higher by 0.005 than that of the unselected clones. This difference indicates that selection for physical tuber characteristics in this population did not adversely affect specific gravity. In fact, at least in this early stage of the breeding program, such selection led to higher specific gravity in the selected population than the unselected population. However, as indicated by the range of mean specific gravity values, there were undoubtedly several clones in the unselected population that had a specific gravity higher than some of the selected clones. Since the determination of specific gravity for individual clones can be prohibitively time-consuming as larger populations are tested, we are encouraged to note that selection can be made on visual observations in the field without negative impact on specific gravity.

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Promotion of in Vitro Leaf Growth of Inner Scales Excised from Dormant Onion Bulbs

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Abstract. **Inner scales excised from dormant bulbs of the short-day 'Texas Grano 1015Y' onion *(Allium cepa* L.) were cultured in vitro and leaf growth was examined. Light promoted leaf growth, but no differences in leaf growth were observed for media pH between 4 and 7. Leaf growth rate in darkness was highest at 24C, reduced at 15C, and greatly reduced at SC. Kinetin promoted leaf growth at 1, 10, and 100 µM. IAA was effective at 1 and 10 µM, but not at 0.1 and 100 µM. GA_3 promoted growth at 0.1** µM. **No inhibitory effects of ABA on leaf growth could be detected. Chemical names used: 1-H-indole-3-acetic acid (IAA), abscisic acid (ABA), gibberellic acid (GA_3), 6-furfurylaminopurine (Kinetin).**

Dormant onion bulbs are almost impervious to external stimuli, and the length of dormancy is affected by cultivar and conditions under which bulbs are grown and stored (Thompson et al., 1972). Onions stored at 10 to 15C tend to sprout earlier than those stored at higher or lower temperatures (Thompson et al., 1972). Auxin, gibberellic acid (GA), and cytokinin levels have been found to be low and inhibitors high in dormant bulbs. The growth promoter vs. inhibitor condition reverses itself when leaf growth is initiated (Aung and Peterson, 1974; Isenberg et al., 1974; Thomas, 1969). Attempts to break dormancy artificially in whole bulbs by applying GA_3, $GA_{4/7}$, 1-naphthaleneacetic acid (NAA), and N^6-benzyladenine (BA) have been unsuccessful (Kate, 1965; Thomas, 1969). However, bulbs with the outer scales removed sprouted earlier (Kate, 1965), and excised inner scales will sprout when cultured in nutrient agar or sand (Jaffe and Isenberg, 1968; Mahotiere et al., 1976a, 1976b).

Our study was conducted to determine effects of medium pH, light, temperature, and plant growth regulators on leaf growth of inner scales excised from dormant onions and cultured in vitro.

Preparation of explants. Three layers of the innermost scales, with the basal plate attached, were carefully removed from medium-size (5.0 to 8.8 cm in diameter) bulbs of 'Texas Grano 1015Y', a short-day onion. The initial size of the explant was ≈ 3 cm high and 1 cm in diameter. For growth measurements, the length of the second outer leaf of the explant was considered to be zero when the scales were excised. Experiments were carried out using bulbs within 1 month of harvest (25 Apr. 1986). Bulbs were stored at 24C until used. The tissue was surface-sterilized with 70% ethanol for 1 rein, 10% commercial bleach (5.2% sodium hypochlorite) solution for 2 rein, and then rinsed with sterile distilled water for 5 min. The basal end of the explants were placed 1 cm deep in medium. Nine explants were used for each treatment.

Preparation of media and culture conditions. To examine the effect of medium pH and light, the basal medium consisted of full-strength Murashige and Skoog (1962) inorganic salts, 30 g sucrosefliter, and 7 g agar/liter. The pH was adjusted to 4, 5, 6, or 7 with 0.1 N NaOH or HCl. Eight milliliters of medium were placed in 25 × 150-mm culture tubes and autoclave at 121C for 15

Table 1. Effect of medium pH and light vs. dark culture conditions on leaf growth of excised inner scales from dormant onions after 21 days in vitro culture.

	Leaf length (cm)[z]			
	Medium pH			
Illumination	4	5	6	7
Yes	5.6	6.4	8.1	5.9
No	8.6	6.1	9.0	8.8
Significance				
pH		NS		
Illumination		*		

[z]Means of nine cultures of inner scales. Initial leaf length was considered to be O cm.
NS,*Nonsignificant or Significant at $P = 0.0.5$, respectively, according to F test.

Table 2. Effect of plant growth regulator concentration on leaf growth on excised inner scales from dormant onions after 14 days in culture.

	Leaf length (cm)[z]			
	Growth regulators			
Concn (µM)	Kinetin	IAA	GA_3	ABA
0	1.8	1.8	1.8	1.8
0.1	3.5	4.3	4.9	4.0
1	7.6	7.9	3.4	4.5
10	9.5	7.6	3.6	3.0
100	12.1	5.7	3.9	1.6
Significance	**	*	*	*

'Means of nine cultures of inner scales. Initial leaf length was considered to be O cm.
*,** Significant at $P = 0.05$ or 0.01, respectively, according to F test.

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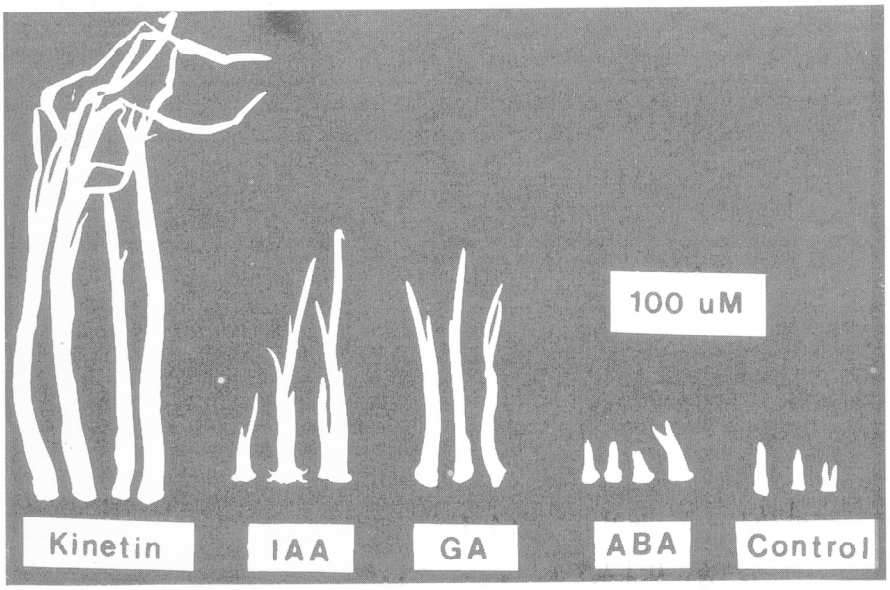

Fig. 1. Effects of 100 µM kinetin, IAA, GA_3, and ABA on leaf growth of excised inner scales from dormant onions after 14 days of in vitro culture.

min. Explants were maintained in either darkness or under continuous fluorescent light (15 $\mu mol \cdot s^{-1} \cdot m^{-2}$) at 24C. In an initial experiment, leaf growth was observed daily for 21 days, and 2 weeks was selected as the appropriate period to determine treatment effects on leaf growth.

To examine the effect of temperature on leaf growth, explants were kept in incubators at 5, 15, or 24C in darkness. The same medium described above (pH 5.5) was used in this experiment.

The medium examining the effects of plant growth regulators consisted of ABA, GA_3, IAA, or kinetin at 0.1, 1, 10, and 100 µM, respectively, and 0.7% agar at pH 5.5 (no salts or sugar). The explants were kept in darkness.

Leaf length was measured from the second outer scale after the first outer scale was removed.

Medium *pH and light. No* differences in leaf growth for media pH between 4 and 7 were observed in either dark or light culture conditions (Table 1). Although light generally promoted leaf growth, illumination was not required for leaf growth. Roots also grew in light, and root initiation coincided with greening of the leaves. In general, root proliferation was associated with enhanced leaf elongation. Bulbing was not observed in any of the treatments. Other results also indicated that leaf elongation could occur using excised scales (Jaffe and Isenberg, 1968; Mahotiere et al., 1976a, 1976b).

Temperatures and shoot growth. Removal of the outer scales had a dormancy-breaking effect, as previously reported (Jaffe and Isenberg. 1968: Mahotiere et al., 1976a, 1976b).Leaf growth was greatest at 24C (4.9 cm), followed by 15C (2.6 cm) and 5C (0.9 cm). Leaf growth response in vitro was different from intact onion bulbs during storage, which showed greater growth at 15C than at 24C (Thompson et al., 1972). By exposing scales to 10C before culture, Mahotiere et al. (1976b) noted the promotion of shoot growth in culture at 20C. Since a previous report indicated that sucrose promoted leaf growth (Mahotiere et al., 1976a), sucrose in the medium might have partly promoted leaf growth in the pH and light or temperature treatments.

Plant growth regulators. Leaf growth was significantly promoted by kinetin at 1 µM concentration or higher (Table 2, Fig. 1). Leaf growth was positively correlated with kinetin concentration. IAA promoted leaf growth at 1 and 10 µM, but not at either lower or higher concentrations. GA_3 enhanced growth at 0.1 µM, but not at the other concentrations and ABA was effective only at 1 µM. We found no inhibitory effects of ABA on leaf growth.

Root growth was evident in IAA-treated cultures, but not in kinetin-treated explants. Elongated and shrunken outer scales, suggesting a movement of water and nutrients to the growing point, were noticeable in kinetin-treated explants. However, the outer scales remained fresh and did not elongate in the control or ABA-treated explants. Promotion of shoot growth by kinetin in this study seems to support a report of increased cytokinin levels in onion bulbs when leaf growth is initiated (Isenberg et al., 1974).

GA_3 was less effective than kinetin in promoting onion leaf growth in this experiment, although increased GA levels have been observed in sprouting bulbs (Abdel-Rahman and Isenberg, 1974; Aung and Peterson, 1974),

A previous report indicated promotion and inhibition of shoot growth by kinetin and ABA, respectively (Mahotiere et al., 1976a); however, the inhibitory effect of ABA was not evident in our study. Conceivably, the low growth rate of the control prevented detection of inhibition by ABA. The level of ABA has been shown to be highest immediately after harvest and to be lowest when bulbs sprout (Jaffe and Isenberg, 1968; Thomas, 1969).

Several previous investigators observed that initial leaf elongation up to 2 to 3 cm occurred, but they did not examine further leaf growth (Jaffe and Isenberg, 1968; Mahotiere et al., 1976a, 1976b). However, we observed actual leaf growth over a longer time - interval (4 vs. 14 days) and at different levels of growth regulators than previously reported.

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