Effects of Selected Herbicides on Provitamin A Content of Vegetables

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The carotene content of Danvers and Chantenay carrots grown in soil treated with the herbicides linuron or CIPC and that of Butternut squash grown in soil treated with amiben or dinoseb was significantly higher than the carotene content of control vegetables. Also dinoseb-treated Butternut squash contained a higher percentage of carotene as α-carotene than did amiben-treated or control squash. Herbicide treatment did not increase carotene content of Hubbard squash or that of two spinach varieties.

The effects of pesticides on fruits and vegetables have been extensively investigated. However, very little has been reported on pesticide effects on nutritional composition of fruits and vegetables. Garman *et al.* (1953) found that some fungicides increased the sugar content of apples. An increase in protein content of simazine-treated crops was noted by Ries (1968). Schuphan (1960) reported that residues of aldrin and dieldrin in the roots of carrots had no effect on ascorbic acid or sugar content. A decrease in free tyrosine in potatoes grown in soil treated with pentachloronitrobenzene was found by Sweeney and Simandle (1968). Increased carotene content of carrots grown in soil treated with telone has been reported by Emerson *et al.* (1969). Wu *et al.* (1970) found that soil treatment with either telone or nemagon increased the carotene content of carrots.

Interest in possible effects of pesticides on nutritional value of foods has made desirable further investigations in this field. Work described here was carried out to determine possible effects of selected herbicides on provitamin A (carotene) content of vegetables.

EXPERIMENTAL

Vegetables selected were known to be good sources of carotene. They were Danvers and Chantenay carrots, Bounty and 668.9 spinach, and Butternut and Hubbard squash. Herbicides used and levels of application were those recommended for carrots, spinach, and squash by the Crops Protection Branch, Agricultural Research Service, U.S. Department of Agriculture. They were CIPC [isopropyl *N*-(3-chlorophenyl)carbamate] or linuron [3-(3,4-dichlorophenyl)-1-methoxy-1-methylurea] for carrots. For spinach CDEC (2-chloroallyl diethyldithiocarbamate) or endothal [7-oxabicyclo[2.2.1]heptane-2,3-dicarboxylic acid] was used. For squash they were amiben (3-amino-2,5-dichlorobenzoic acid) or dinoseb (4,6-dinitro-*O*-*sec*-butylphenol).

All experiments were arranged in randomized block design with plots of each treatment for each variety being replicated four times. Herbicides were applied to the soil before planting at the following rates per acre: CIPC, amiben and CDEC, 4 lb, linuron 1.5 lb, dinoseb 6 lb, and endothal 2 lb.

Each vegetable was sampled on four or five different days. Samples were collected at random from each plot and composite samples prepared each day for carotene determinations. Extractions were made immediately after harvest and the extracts transferred to petroleum ether, concentrated under reduced pressure, and stored at 0° F. Determinations were replicated at least four times on each sample extract.

Determinations of α- and β-carotene and their stereoisomers were made by the method of Sweeney and Marsh (1970). Vitamin A values, determined by Zechmeister (1962) for the various carotene stereoisomers, were used in calculating biological values. Differences between means were tested for significance by application of Student's t test.

RESULTS

Carrots. Carotene content usually increased with increased maturity of the vegetable (Table I). However, for each sampling date, herbicide-treated carrots were significantly higher, usually at the 0.1% level, than were control carrots. Mean values for herbicide-treated carrots for all five harvest dates combined exceeded those for control carrots by the following percentages: Danvers, linuron 20%; Danvers, CIPC 30%; Chantenay, linuron 50%; and Chantenay, CIPC 40%.

Spinach. In contrast to results obtained on carrots, carotene content of spinach decreased with increasing maturity of the vegetable (Table II). Spinach grown in soil treated with CDEC or endothal was not higher in carotene content than was spinach grown in soil not treated with herbicides. Herbicide-treated Bounty spinach was lower in carotene content than was control spinach. For the 668.9 variety, mean values for herbicide-treated and control sample did not differ greatly.

Squash. Total carotene content of squash did not appear to be related to date of harvest. Butternut squash grown in soil treated with either amiben or dinoseb were generally higher in carotene than were control samples (Table III). There were also differences in percentages of α-carotene and β-carotene that appeared to be related to herbicide treatment. Butternut squash grown in soil treated with amiben generally contained a lower percentage of its carotene as α-carotene and a correspondingly higher percentage as β-carotene than did control or dinoseb-treated squash. Carotene content of the Hubbard squash, which contained no α-carotene, did not appear to be affected by the herbicides used.

Stereoisomers. None of the herbicides used on the vegetables appeared to have significant effects on the formation

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Table I. Effect of Herbicides on Carotene Content of Two Carrot Varieties[a]

Harvest date	Treatment	Danvers α-Carotene %	Danvers β-Carotene %	Danvers Total μg/100 g	Chantenay α-Carotene %	Chantenay β-Carotene %	Chantenay Total μg/100 g
8-4-70	Control	44.1	55.9	4910	41.9	58.1	4760
	Linuron	43.1	56.9	6220[c]	43.1	56.9	6470[c]
	CIPC	41.4	58.6	8580[c]	42.8	57.2	7380[c]
8-6-70	Control	49.1	60.9	6430	45.8	54.2	3470
	Linuron	43.1	56.9	7090	44.1	55.9	5440[c]
	CIPC	42.0	58.0	7000[b]	42.0	58.0	5260[c]
8-11-70	Control	45.0	55.0	7060	37.0	63.0	4460
	Linuron	40.6	59.4	9020[c]	41.8	58.2	6990[c]
	CIPC	47.7	52.3	10290[c]	40.4	59.6	7370[c]
8-12-70	Control	42.3	57.7	6880	41.6	58.4	6140
	Linuron	42.9	57.1	7610[b]	39.5	60.5	9420[c]
	CIPC	37.8	62.2	8210[c]	41.2	58.8	7500[c]
8-18-70	Control	38.4	61.6	14160	39.2	70.8	6260
	Linuron	41.8	58.2	17850[c]	39.0	61.0	8620[c]
	CIPC	41.8	58.2	17070[c]	43.7	56.3	7200[c]
Mean	Control	43.8	56.2	7890	41.1	58.9	5020
	Linuron	42.3	57.7	9560[c]	41.5	58.5	7390[c]
	CIPC	42.1	57.9	10220[c]	42.0	58.0	6940[c]

[a] Values are means of four replications. [b] Significantly different from control at 5% level. [c] Significantly different from control at 0.1% level.

of carotene stereoisomers (Table IV). α-Carotene was detected only in carrots and Butternut squash. The stereoisomers of α- and β-carotenes are known to have lower provitamin A values than do the all *trans*-carotenes (Zechmeister, 1962). The fact that the herbicides used did not cause significant changes in the ratio of cis isomers to all trans isomers is therefore important from a nutritional standpoint.

DISCUSSION

Nonchlorophyll containing vegetables, such as carrots, accumulate increased amounts of carotene with maturity. In green vegetables such as spinach, carotene is also synthesized as the plant matures. But since carotene helps prevent oxidative destruction of the vegetable by chlorophyll, the carotene molecule is altered in the process (Claes and Nakayama, 1959). Consequently, early harvested spinach would tend to be higher in carotene content than would that harvested at a later date. For squash all of the vegetables on a vine do not mature at the same time. Therefore, a relationship between harvest time and carotene content would not be expected.

The chemistry and mode of action of the various herbicides

Table II. Effect of Herbicides on Carotene Content of Two Spinach Varieties[a]

Harvest date	Treatment	Bounty β-Carotene μg/100 g	668.9 β-Carotene μg/100 g
6-9-70	Control	9250	5730
	CDEC	4260[e]	3910[e]
	Endothal	4820[d]	4830[d]
6-17-70	Control	5610	3650
	CDEC	4670[e]	4070
	Endothal	4826[e]	4810[e]
6-23-70	Control	4310	2870
	CDEC	4610[e]	3190[c]
	Endothal	3480[e]	5170[e]
6-30-70	Control	3900	3240
	CDEC	3340[c]	3250
	Endothal	2530[e]	3430
Mean[b]	Control	4830	3870
	CDEC	4220	3610
	Endothal	3910	4560

[a] Values are means of four replications. [b] Significance of differences between means not determined. [c] Significantly different from control at 5% level. [d] Significantly different from control at 1% level. [e] Significantly different from control at 0.1% level.

Table III. Effect of Herbicides on Two Varieties of Squash[a]

Harvest date	Treatment	Butternut α-Carotene %	Butternut β-Carotene %	Butternut Total μg/100 g	Hubbard β-Carotene μg/100 g
9-22-70	Control	32.4	67.6	2590	830
	Amiben	33.2	66.8	2120[d]	1020[d]
	Dinoseb	37.1[d]	63.0[b]	2990[c]	830
9-24-70	Control	40.6	59.4	1670	1220
	Amiben	31.6[d]	68.4[d]	2920[d]	1000[d]
	Dinoseb	38.6	61.4	2210[d]	890[d]
9-29-70	Control	31.4	68.6	1630	560
	Amiben	30.2	69.8	1940[c]	860[d]
	Dinoseb	36.7[d]	63.4[d]	2730[d]	790[d]
9-30-70	Control	35.6	64.3	2470	650
	Amiben	30.8[c]	69.2[d]	3500[d]	940[d]
	Dinoseb	34.3	65.7	3050[d]	900[d]
Mean	Control	35.0	65.0	2090	820
	Amiben	31.5[c]	68.5[c]	2620[b]	960
	Dinoseb	36.7	63.4	2740[d]	850

[a] Values are means of four replications. [b] Significantly different from control at 5% level. [c] Significantly different from control at 1% level. [d] Significantly different from control at 0.1% level.

Table IV. Carotene Stereoisomers in Herbicide-Treated Vegetables

Variety	Treatment	Neo-α-carotene B %	All-*trans*-α-carotene %	Neo-α-carotene U %	Neo-β-carotene B %	All-*trans*-β-carotene %	Neo-β-carotene U %	Biological[a] value %
				Carrots				
Danvers	Control	1.3	38.8	0.7	5.4	52.2	1.6	76.5
	Linuron	1.3	42.8	0.7	3.8	49.8	1.6	75.4
	CIPC	2.4	42.6	0.5	4.1	49.2	1.3	75.0
Chantenay	Control	2.4	37.2	0.9	3.2	55.2	1.1	77.5
	Linuron	1.8	37.0	0.4	5.4	53.6	1.8	77.2
	CIPC	1.4	38.1	0.3	3.2	55.6	1.4	78.2
				Squash				
Butternut	Control	1.2	32.4	1.2	6.6	53.2	5.4	76.4
	Amiben	1.4	29.1	1.1	6.7	56.6	5.1	77.8
	Dinoseb	1.8	30.8	1.3	5.9	55.1	5.1	76.9
Hubbard	Control				8.6	84.2	7.2	91.5
	Amiben				9.4	85.1	5.5	92.2
	Dinoseb				8.1	85.9	6.0	92.5
				Spinach				
Bounty	Control				9.4	80.3	10.3	89.2
	CDEC				7.5	82.4	10.1	90.2
	Endothal				10.4	78.9	10.8	88.6
668.9	Control				9.4	79.7	10.9	88.8
	CDEC				8.2	80.2	11.6	88.8
	Endothal				10.0	78.7	11.1	88.3

[a] Biological values based on biopotency values of Zechmeister (1962).

are not completely understood. It is known that phenylureas, such as linuron, and phenylcarbamates, such as CIPC, inhibit photosynthesis. This inhibition appears to be related to phytotoxicity (Woodford and Evans, 1963). Phenylcarbamates also inhibit cell division but this occurs in both susceptible and resistant species.

In work reported here increased carotene content was obtained for carrots grown in soil treated with either linuron or CIPC. Wu *et al.* (1970) have speculated that increased carotene content of carrots treated with the soil fumigants, telone or nemagon, was related to decreased respiration. Wassink and van Elk (1961) have reported that CIPC appeared to inhibit respiration of germinating pea seeds. This would appear to be further evidence of a relationship between carotene content and respiration rate. However, a relationship between respiration and carotene content would be difficult to establish because many herbicides have been reported to stimulate respiration when applied at a low level and to inhibit respiration when applied at higher levels (Bishop, 1958; Simon, 1953).

In squash, the Butternut variety showed significant increases in carotene as the apparent result of herbicide treatment. Hubbard squash, a much larger variety, had no increase in carotene with herbicide treatment at the level used.

In spinach it is possible that the effect of chlorophyll in decreasing carotene might mask any increase resulting from herbicide treatment.

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