to 55 days, and therefore the mice evidently did not live long enough to develop the lung metastases. The 3 mice which did show metastases at death lived 90 days. It was apparent during the experiment that the primary tumors of the mice receiving the 100-μg. doses of liver *L. casei* factor intravenously were growing much more rapidly than the tumors of untreated controls. Unfortunately, the wide fluctuations in the growth rate of these tumors do not allow any quantitative evaluation.

Whether these findings apply only to the particular strain of mice and method of assay used in these experiments needs further investigation. No conclusions should be drawn from these animal experiments as to the action of the liver *L. casei* factor on human cancer.

References

1. ANGIER, R. B., *et al.* *Science*, 1945, **102**, 227.
2. HUTCHINGS, B. L., STOKSTAD, E. L. R., BOHONOS, N., and SLOBODKIN, N. H. *Science*, 1944, **99**, 371.
3. LEUCHTENBERGER, R., LEUCHTENBERGER, C., LASZLO, D., and LEWISOHN, R. *Science*, 1945, **101**, 46.

Stability of Carotene in Dehydrated Carrots Impregnated With Antioxidants[1]

T. E. WEIER and C. R. STOCKING

Botany Division, College of Agriculture
University of California, Davis

Deterioration in quality of dehydrated carrots is usually associated with two different types of changes. One of these, resulting in a darkening of the carrots, occurs in an aqueous phase and is concerned with the union of amino acids and sugars—the so-called Maillard reaction. The other is made evident by the deterioration of the carotenoid pigment and is probably allied to the oxidative rancidity of the carrot oils. This report is concerned with the latter phase of deterioration.

We have shown in previous publications that, when carrots are dried, the carotene goes into solution in droplets of oil and that, upon storage, the carotene disappears, leaving colorless droplets which give a positive test for aldehydes with Schiff's reagent (*3*). It has been reported that carotene dissolved in oil hastens the oxidation of the oil (*1*). Furthermore, carotene is degraded concurrently with the oxidation of fats (*2*). These observations suggest that the oil droplets in the cells of dried carrots are undergoing oxidative changes and that the carotene is degraded as a result of these changes.

Blanching improves the stability of the carotenoids but does not stop their deterioration. Furthermore, leaching blanched carrots before storage accelerates the rate of pigment breakdown. (Compare Items 6, 7, and 8 with Item 15 in Table 1.) Tests with carrots which had been blanched but not dried showed that the rate of pigment breakdown could be retarded by the application of antioxidants (*4*). This paper reports the results of a survey of the action of some 30 tests of various antioxidants on blanched, dried carrots. The tests were carried out on carrots harvested during the summer.

Imperator carrots, about 4 months of age, were harvested during August, sliced (2–3 mm. thick), and blanched in steam for 5 minutes. The blanched slices were soaked for 5 minutes in solutions of various antioxidants. They were drained and dried in a tunnel dehydrator at approximately 60° C. to a moisture content of about 6 per cent and then ground on a Wiley mill to pass a 20-mesh screen, the smaller particles being sifted through a 48-mesh screen on a Rotap machine and discarded. The remaining grains were divided, half being stored in an incubator at 40° C., and half stored at room temperature. Both storage tests were carried out in darkness. Pigment concentration, in an acetone extract, was measured on the Evelyn colorimeter using the 440 filter.

TABLE 1

	Treatment	Carotenoids remaining in samples after storage at 40° C. (%)				
		48 hr.	72 hr.	96 hr.	120 hr.	168 hr.
(1)	.01% N.D.G.A.* + .3% $Na_2S_2O_5$ (in 40% ethanol) ..		95.8			91.5
(2)	.1% pyrogallol + .2% $Na_2S_2O_5$ ..	94.5				91.3
(3)	.1% ascorbic acid		101.0		93.0	89.5
(4)	.1% pyrogallol ..		94.5			85.1
(5)	.01% N.D.G.A. (in 40% ethanol) .		98.7		85.5	82.0
(6)	Blanch only		93.0		91.0	77.0
(7)	" "	94.0		92.0		75.0
(8)	" "	94.0		91.0		74.0
(9)	.1% piperidine ...	94.0		89.0		76.0
(10)	.2% pyrrole (in 40% ethanol) .	95.0		89.0		75.0
(11)	.1% hydroquinine	97.0				74.0
(12)	.2% lecithin (in 40% ethanol) .	94.0		87.0		71.0
(13)	Blanch	83.0		76.0		67.0
(14)	40% ethanol leach	93.0		77.0		57.0
(15)	Water leach	82.0		73.0		45.0

Other substances tested: hemin, tocopherol, tocopherol plus ascorbic acid, piperidine plus SO_2, phosphate buffer pH7, phosphate plus SO_2, diphenylamine, *p*-phenylene diamine dihydrochloride, N.D.G.A. plus ascorbic acid, gallic acid, maleic acid, oxalic acid, γ-tocopherol palmitate, and α-tocopherol succinate.

* N.D.G.A. = nordihydroguaiaretic acid.

The antioxidants tested are shown in Table 1. Since the procedure for impregnation of the antioxidants required a 5-minute soak in the solution of the antioxidant to be tested, standards for comparison were

[1] The subject matter of this paper has been undertaken in cooperation with the Quartermaster Corps Committee on Food Research.

prepared by soaking blanched carrots in distilled water or 40 per cent ethanol for 5 minutes (Items 14 and 15). Pigment breakdown was also followed in stored samples that had received no treatment other than blanching (Items 6, 7, 8). These latter experiments served as a general check on the series of tests.

A slower rate of pigment breakdown than that found for the water and ethanol-treated samples (Items 14 and 15) would indicate that the materials with which the carrots were impregnated were exercising some protective action. Practically, of course, any added substance must result in a slower rate of breakdown than that found after blanch only.

Those materials which protect the carotenoids in carrots stored at 40° C. are listed in the order of their potency in Table 1. Those compounds which do not retard the breakdown of the pigments are listed without inclusion of figures or placement in any order.

Item 13 may be of significance. The carrots of this test were blanched, dried, ground, and stored at 4° C. for about 8 weeks before being transferred to 40° C. for the regular storage test. No change in the pigment, measurable on the Evelyn colorimeter, occurred during cold storage. Rate of breakdown at 40° C., however, was much more rapid than that of samples stored at 40° C., immediately after preparation.

The erratic results for thiourea and sodium metabisulphite at 40° C. are not reported.

The samples held at room temperature were analyzed for pigment content after 2 and 4 months storage (Table 2). Since the samples were placed in storage during the fall, those stored early in September were subjected to higher temperatures than those stored at the end of the month. For this reason the table is divided into two parts.

We conclude that:

(1) The carotenoids in summer-harvested, blanched, dehydrated carrots may be stabilized by certain antioxidants.

(2) In general, those antioxidants which were effective at 40° C. were also effective at room temperature. The following exceptions may be noted: lecithin, in relation to the other substances, was not as effective at room temperature as at 40° C.; R (an antioxidant obtained from the Dairy Industry Division) and a combination of a phosphate buffer (pH 7.1) and .3 per cent $Na_2S_2O_5$, were, in relation to other substances, more effective at room temperature than at 40° C.

(3) The effectiveness of .1 per cent pyrogallol + .1 per cent $Na_2S_2O_5$ was markedly greater than that of any other substance or substances. Carrots treated with this mixture contained more of the original carotene content (79 per cent) after 4 months storage than other samples did after 2 months storage (Table 2). The combination is more effective than either substance alone. Color retention is good, and no unpleasant odor developed.

(4) There is a good correlation between odor and carotenoid degradation. Those samples with a high carotene content retained a pleasant smell. No tasting tests have as yet been tried.

TABLE 2

	Antioxidant	Carotenoids remaining in samples after room storage (%)		Comparison with 40° C. storage as shown by relative rank in Table 1
		2 mos.	4 mos.	
	A. *Stored between 6 September and 17 January*			
(1)	.1% ascorbic acid and .08% tocopherols (in 40% ethanol)	54	37	Not tabulated but similar to ascorbic acid alone
(2)	.1% ascorbic acid	57	34	3
(3)	.01% N.D.G.A. (in 40% ethanol)	46	34	5
(4)	.1% piperidine	43	31	9
(5)	R	54	30	Poor, not tabulated
(6)	.2% pyrrole (in 40% ethanol)	44	27	10
(7)	.1% piperidine + .5% $Na_2S_2O_5$	33	20	Poor, not tabulated
(8)	40% ethanol leach	20	12	14
(9)	Water leach	18	8	15
(10)	Blanch only	21	7	6
(11)	.2% lecithin (in 40% ethanol)	20	1	12
(12)	.07% tocopherols (in 40% ethanol)	27	1	Poor, not tabulated
(13)	.01% hemin	6	1	" " "
	B. *Stored between 26 September and 26 January*			
(1)	.1% pyrogallol + .2% $Na_2S_2O_5$	100	79	2
(2)	.1% pyrogallol	69	55	4
(3)	.01% N.D.G.A. + .3% $Na_2S_2O_5$	71	53	1
(4)	.1% hydroquinone	62	43	11
(5)	Phosphate buffer pH 7.1 + .3% $Na_2S_2O_5$	61	38	Poor, not tabulated
(6)	.1% diphenylamine (in 40% ethanol)	37	33	" " "
(7)	.5% $Na_2S_2O_5$	38	23	" " "
(8)	.1% *p*-phenylene diamine	26	17	" " "
(9)	Phosphate buffer pH 7.1	18	11	" " "
(10)	.1% gallic acid	7.1	7	" " "

In work on blanched, undried carrots it was found that the carotenoids were much more stable in winter than in summer. Similar results are being obtained with dried carrots. Pigment breakdown in winter-harvested carrots approximates that obtained for summer carrots treated with ascorbic acid. The protective action of ascorbic acid on the carotenoids in winter-harvested carrots is correspondingly lessened.

These results, which in no way refer to the suitability of these materials for use in foodstuffs, have been obtained from experiments on summer-harvested carrots. It is very possible that different results will be obtained from studies on winter-harvested material.

References

1. Olcott, H. S., and Mattill, H. A. *J. Amer. chem. Soc.*, 1936, **58**, 1627–1630.
2. Sumner, R. J. *Chem. Abstr.*, 1942, **37**, 961.
3. Weier, T. E. *Amer. J. Bot.*, 1944, **31**, 342–346.
4. Weier, T. E. *Hilgardia*, in press.