Relationship of Vegetable Color to Physical State of the Carotenes

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Some high-carotenoid vegetables show a distinct color shift when heated. The absorption spectra of fresh and heated carrot, sweet potato, squash and tomato were determined. The spectra of fresh vegetables having well-defined chromoplasts were nearly duplicated by aqueous suspensions of the major carotene. The color shift is attributed to degradation of chromoplasts and solution of the carotenes in other cellular lipids.

Some high-carotenoid vegetables show a distinct color shift when heated in water, while others do not. Carrots and sweet potatoes change from orange to yellow; tomatoes change from red to orange-red. No color change was observed in yellow summer squash. The color changes cannot be explained by a change in carotenoid content nor isomerization to *cis*-carotenoids (Purcell, 1962; DellaMonica and McDowell, 1965). These color changes were studied spectrophotometrically and microscopically.

EXPERIMENTAL

Vegetables. Mature carrots (Imperator), squash (Yellow Crookneck), sweet potatoes (Centennial and Goldrush) and table red tomatoes (Homestead) were obtained on the local market; consequently, their exact maturity states were unknown. In all cases, sections were obtained from the fleshy part of the vegetable.

Spectrophotometry. Carrot and sweet potato slices 1 to 2 mm. and 2 to 3 mm. thick, respectively, were placed in square 1-cm. borosilicate spectrophotometer cells and covered with water. The cells were placed in vacuum for 1 minute to remove entrapped gases from the tissue. When the vacuum was released, transparency of the slices increased. The cells were placed as near as possible to the phototubes in a Cary Model 15 Spectrophotometer. After balancing the instrument with neutral density filters in the reference beam, the spectrum was scanned from 600 to 400 mμ. Tomato slices 6 to 8 mm. thick and squash slices 4 to 5 mm. thick were treated the same way, except that they were placed in cells with a medium of 0.05*M* calcium chloride and 0.05*M* sodium chloride. This medium reduced disintegration of the slices during evacuation and heating without changing the spectra.

Heating. The slices were heated by immersing the spectrophotometer cells to about one half their depth in boiling water for two minutes. After cooling, the spectra of the heated slices were obtained.

Microscopy. Tissue mashes of fresh and heated vegetables were examined microscopically, enlarged 430× with a polarizing microscope.

Water-Dispersible Suspensions of Carotenoids. Crystallized bovine serum albumin, 14 mg., and recrystallized Hoffman-LaRoche β-carotene, 0.2 mg. in 2 ml. of ether, were intimately mixed in a tissue mortar. The ether was evaporated, and the residue was suspended in 5 ml. of water in the tissue mortar. Absorption spectra were obtained. Crystalline lycopene obtained from tomatoes was suspended in the same way.

RESULTS AND DISCUSSION

The spectral absorption maxima of fresh and heated carrot, sweet potato, squash, and tomato are shown in Table I. These data indicate that the absorption maxima of carrot and sweet potato, believed to be due to β-carotene, can be nearly duplicated by water-dispersible suspension of β-carotene. The absorption maxima of tomato at 560 and 480 mμ is nearly duplicated by the water-

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Table I. Spectral Absorption Maxima of Fresh and Heated Vegetables, Crystalline β-Carotenes, and Suspensions of β-Carotene and Lycopene

Vegetable	Major Carotene	Maxima, mμ
Carrot	β-Carotene	
Fresh		518,485,455
Heated		485,457,435
Sweet potato	β-Carotene	
Fresh		517,480,455
Heated		485,455
Squash	β-Carotene	
Fresh		480,450,425
Heated		480,450,425
Tomato	Lycopene	
Fresh		560,480
Heated		480,455,435
Crystalline carotene[a]	β-Carotene	538,492,462,435
Colloidal suspension[a]	β-Carotene	519,478,452,428
Water dispersible β-carotene	β-Carotene	523,485,457
Water dispersible lycopene	Lycopene	565,520,483

[a] Tachibana and Nakamura (1965).

dispersible suspension of lycopene with maxima at 565, 520, and 483 mμ.

The maxima of squash, heated carrot, and heated sweet potato are nearly the same as for β-carotene dissolved in vegetable oils, 485 and 457 mμ. The absorption spectrum of heated tomato has very little structure. The maxima at 560 mμ is broadened and ceases to be a distinct maxima, although absorption in this area is not changed. Absorption at 480 and 455 mμ are close to two of the maxima of lycopene in vegetable oil, 511, 478, and 450 mμ.

Microscopic examination of carrot and sweet potato showed numerous highly birefringent chromoplasts in the fresh tissue, with no yellow pigment outside the chromoplasts. Upon heating, the chromoplasts disappeared, and colored droplets formed around the inside of the cell wall. When prolonged heating caused breakdown of the cells, most of the debris appeared yellow with scattered yellow droplets. Fresh squash had no birefringent chromoplasts. The pigment appeared mainly in droplets near the cell wall. In the tomato, the chromoplasts disappeared on heating, but larger birefringent colored areas and scattered nonbirefringent droplets appeared upon cooking.

The data presented here and other published data indicate that a change in the physical state of the carotenoids is responsible for heat-caused color changes. Reeve (1943) and Weier (1944) stated that carotene dissolves in an unsaturated oil as dehydrated carrots deteriorate. Similarly, when fresh carrot and sweet potato are heated, chromoplasts disintegrate, and the carotenes dissolve in cellular lipids. Heating does not cause a color shift in squash because the carotene is not located in discrete chromoplasts, but dissolved in cellular lipids in fresh tissue. This supposition is supported by the data of Table II, which shows that the lipid-carotene ratio for squash is

Table II. Lipid and Carotene Contents and Lipid-Carotene Ratios of Vegetables Studied

Vegetable	Lipid, Mg./G.[a] (Seeds Omitted)	Carotene, Mg./G.[b]	Lipid/ Carotene
Carrot	1.54	0.11	14.0
Sweet potato	2.42	0.10	24.2
Squash	1.10	0.002	579.0
Tomato	1.10	0.12	9.2

[a] Watt and Merrill (1963).
[b] Typical analysis from this laboratory.

about 41 times greater than for carrots and 24 times greater than for sweet potatoes.

In the tomato, the color shift is not as pronounced as in carrot and sweet potato. This is believed to be due to the formation of lycopene crystals after heating. In addition to the lipid-lycopene ratio being somewhat lower than the lipid-carotene ratio of carrots (Table II), the solubility of lycopene is about 0.07 times as great as the solubility of β-carotene (Karrer and Jucker, 1950). It is therefore not surprising that lycopene crystals can form in the tomato after heating, while carotene crystals do not form in heated carrots.

This supposition was tested with tomato chromoplasts isolated by the method of Purcell *et al.* (1963). The absorption spectrum of the chromoplast suspension was essentially the same as that of the tomatoes from which they were isolated. Upon heating, the spectrum did not change significantly. An aqueous suspension of mineral oil stabilized with bovine serum albumin was added to the chromoplast suspension to a concentration of 1% mineral oil. This addition caused no qualitative change in the absorption spectrum, but when this mixture was heated the color changed to orange. Absorption near 560 decreased and distinct maxima with greatly increased absorption occurred at 511, 478, and 450 mμ.

Nearly identical behavior was obtained when an aqueous suspension of lycopene was treated in the same way.

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Received for review June 17, 1968. Accepted September 26, 1968. Published with the approval of the Director of Research, North Carolina Agricultural Experiment Station, as Paper No. 2608 of the Journal Series. Use of trade names of specific materials does not constitute a recommendation by the U. S. Department of Agriculture to the exclusion of others which may also be available. Research partially supported by Office of Economic Aid.