

MeCOPr, 107-87-9; BuCHO, 110-62-3; OHC(CH₂)₃CHO, 111-30-8; PrCH=CHCHO, 505-57-7; (hydroxymethyl)furfural, 25376-49-2.

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Received for review January 9, 1987. Revised manuscript received August 10, 1987. Accepted September 29, 1987.

Volatile Components of Mango Preserved by Deep Freezing

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Mango slices were stored for 14 months in a deep freeze at -15 °C and then analyzed for their volatile aroma components. Results were remarkably similar to those obtained for the fresh fruit, although the amount of the important constituent, car-3-ene, was slightly reduced. The plasticizer, di-2-ethylhexyl adipate, was also detected in the aroma isolate, originating by migration from the PVC film in which the fruit was wrapped while in storage.

In a previous paper we reported the results of analysis of the volatile flavor components of two cultivars of mango from Florida (MacLeod and Snyder, 1985). In addition, the effects of a limited range of storage conditions on the volatiles were also assessed, and it was found that storage of mango slices at -15 °C for 15 days provided results that were virtually identical with those for the fresh fruit. We now report data for mangoes stored similarly, but for a much longer period (over 1 year).

EXPERIMENTAL SECTION

All the mangoes used in this work (cultivar Tommy Atkins) were shipped by air freight to London from Miami,

FL. Some were analyzed when fully ripe, and results have already been reported (MacLeod and Snyder, 1985). The remainder were stored for subsequent analysis as follows. Two whole mangoes were placed separately in plastic bags, the bulk of the air was removed, and the bags were sealed. A further two were first wrapped in clingfilm (thin plasticized PVC film designed specifically for such food storage) and then kept in plastic bags as described above. The final two mangoes were first sliced, the stones removed, and the slices, with skin still attached, then wrapped in clingfilm and placed in two plastic bags as described above. All samples were then stored in a deep freeze at -15 °C for 14 months. After thawing, the aroma volatiles of the sliced fruit were then analyzed exactly as described previously (MacLeod and Snyder, 1985).

RESULTS AND DISCUSSION

On thawing, the mangoes that had been stored whole in the deep freeze for 14 months were found to be unac-

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Table I. Volatile Constituents of Mango cv. Tommy Atkins

component	t_R^a min	Kovats index (lit.)	stored		fresh	
			% rel abund	$\mu\text{g/g}$	% rel abund	$\mu\text{g/g}$
pentan-2-one	2.2	672	tr ^b	tr		
pyridine	2.3	695	1.1	1.0		
toluene	2.5		0.4	0.4	1.2	0.9
hexan-3-ol	3.0					
plus furfural		815	1.8	1.6		
<i>trans</i> -hex-2-enal	3.3	832	1.9	1.7		
<i>cis</i> -hex-3-en-1-ol	3.4	847	1.9	1.7		
α -pinene	5.8	932	22.0	19.9	22.2	16.6
camphene	6.1	948	1.8	1.6	0.4	0.3
sabinene	6.8	965	tr	tr	0.2	0.1
myrcene	7.0	983	3.0	2.7	1.3	1.0
2-pentylfuran	7.3	983	0.2	0.2		
β -pinene	7.5	988	1.7	1.5	2.0	1.5
monoterpene hydrocarbon	7.7		0.2	0.2		
α -phellandrene	8.0	1000	0.6	0.5	0.2	0.1
car-3-ene	8.5	1000	42.2	38.2	60.2	45.2
<i>p</i> -cymene	8.8	1015	tr	tr	0.1	0.1
β -phellandrene	9.0	1023	1.1	1.0	0.5	0.4
limonene	9.1	1022	2.1	1.9	1.7	1.3
γ -terpinene	10.5	1053	0.5	0.5		
α -terpinolene	12.0	1083	2.2	2.0	1.5	1.1
monoterpene	16.0		0.1	0.1		
? an ethylstyrene	16.2		0.1	0.1		
monoterpene	16.5		0.6	0.5		
a tetramethylbenzene plus 2,5-dimethylstyrene	17.9		0.2	0.2		
pentyl ester	23.9		0.3	0.3		
α -copaene	26.4	1410	0.3	0.3	0.8	0.6
β -elemene	27.1	1410	0.6	0.5		
guaiene	28.1		0.4	0.4		
β -caryophyllene	28.4	1452	3.2	2.9	1.2	0.9
α -humulene	29.8	1487	1.4	1.3	0.6	0.5
sesquiterpene hydrocarbon	31.0		0.1	0.1		
? β -selinene	31.2	1530	0.5	0.5		
? α -selinene	31.5	1534	0.2	0.2		
sesquiterpene hydrocarbon	32.0		0.1	0.1		
δ -cadinene	32.7	1546	0.2	0.2		
sesquiterpene	37.5		tr	tr		
? hexadecanol	40.0	1750	0.5	0.5		
ethyl hexadecanoate	50.5		0.2	0.2		
eicosane	54.5	2000	0.1	0.1		
heneicosane	57.2	2100	0.1	0.1		
docosane	61.5	2200	0.1	0.1		
tricosane	67.0	2300	0.1	0.1		

^aUsing a 25-m fused silica capillary column coated with bonded-phase BP1. ^btr = trace (<0.1).

ceptable and were not therefore further analyzed. The problem was that although the odor of the fruit tissue was good, its texture was not, and the now stringy pulp was much too soft and "mushy", doubtless due to cellular damage during freezing. The taste of the fruit was very poor, mainly due to the very unpleasant mouth-feel of the watery flesh. It was difficult to separate the fruit tissue from the stone and toughened skin, and it was decided not to attempt to analyze the aroma volatiles of the fruit, since clearly there was no commercial or other value to such a product. However, the mangoes that had been sliced and separated from the stone before similar storage were found, on thawing, to have perfect texture, like that of the fresh fruit. The firm tissue could be readily separated from the skin, and it was found to possess very good mango taste and odor. Clearly from an acceptance point of view, such storage provided a product that would not be rejected by the consumer. It is unclear why the sliced fruit survived the same storage processes so much better than the whole fruit.

The aroma volatiles of the thawed mango slices were collected and analyzed exactly as already described for the fresh fruit (MacLeod and Snyder, 1985), and results are given in Table I. The numbers of unidentified components not included in the table and their total percentage contributions to the samples were 14 (5.9%, w/w) for the

stored fruit and 15 (4.2%) for the fresh fruit. The majority of the unidentified components was present in such small amounts that mass spectra could not be obtained, even with packed GC columns and high sample loadings.

Comparing the results for the fresh and stored mango, it can be seen from Table I that they are remarkably similar, probably the main difference being the slightly larger number of minor constituents detected in the sample from the stored fruit. This was most likely due to slightly improved GC-MS in the later analysis. With regard to the total concentration of volatiles, this changed little on storage of the mangoes, increasing from about 72 to about 91 $\mu\text{g/g}$, under the circumstances an insignificant variation. Considering changes in individual aroma constituents, it can be seen from the table that in general these are minor, with the possible exception of a noticeable decrease in the amount of car-3-ene on storage, in both percentage and absolute terms. It is interesting to note in compensation that the amounts of virtually all other terpenes (mono and sesqui) increased slightly on storage, again in both percentage and absolute terms. It is highly unlikely that these very slight changes would make any difference to the flavor of the stored fruit compared with the fresh, particularly taking into account the gross differences in the aroma volatiles of different cultivars of mango that have previously been reported (MacLeod and Pieris, 1984; MacLeod

and Snyder, 1985). The conclusion, therefore, is that this method of storage, for an extended period, is a perfectly acceptable method, with regard to retention of flavor in the mango.

One other constituent was detected in the aroma isolate from the stored mango slices, but it is not listed in Table I since it is not a genuine mango volatile. It is di-2-ethylhexyl adipate, which eluted between docosane and tricosane on a BP1 bonded-phase fused silica capillary column, with a retention time of 65.0 min. This is the plasticizer used in clingfilm, and therefore this had clearly migrated from the wrapping to the flesh of the fruit, even when deep frozen. However, the amount detected was very small (about 0.2 $\mu\text{g/g}$, i.e. 0.2 ppm), and it is well-known that di-2-ethylhexyl adipate will migrate into closely wrapped foods, but at levels not constituting a health hazard.

ACKNOWLEDGMENT

We thank Ed Mitchell Mangoes, Miami, FL, for supplies of mangoes and W. G. Gunn and A. E. Cakebread for running the GC-MS.

Registry No. PrCOMe, 107-87-9; PrCH(OH)Et, 623-37-0; (E)-PrCH=CHCHO, 6728-26-3; (Z)-EtCH=CH(CH₂)₂OH, 928-96-1; Me(CH₂)₁₄CHO, 629-80-1; Me(CH₂)₁₄CO₂Et, 628-97-7; pyridine, 110-86-1; toluene, 108-88-3; furfural, 98-01-1; α -pinene, 80-56-8; camphene, 79-92-5; sabinene, 3387-41-5; myrcene, 123-35-3; 2-pentylfuran, 3777-69-3; β -pinene, 127-91-3; α -phellandrene, 4221-98-1; car-3-ene, 13466-78-9; *p*-cymene, 99-87-6; β -phellandrene, 555-10-2; limonene, 138-86-3; γ -terpinene, 99-85-4; terpinolene, 586-62-9; tetramethylbenzene, 25619-60-7; 2,5-dimethylstyrene, 2039-89-6; α -copaene, 3856-25-5; β -elemene, 33880-83-0; guaiane, 88-84-6; β -caryophyllene, 87-44-5; α -humulene, 6753-98-6; β -selinene, 17066-67-0; α -selinene, 473-13-2; δ -cadinene, 483-76-1; eicosane, 112-95-8; heneicosane, 629-94-7; docosane, 629-97-0; tricosane, 638-67-5; di-2-ethylhexyl adipate, 103-23-1.

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Received for review December 2, 1986. Accepted August 27, 1987.

Effects of Freezing and pH of Alfalfa Leaf Juice upon the Recovery of Chloroplastic Protein Concentrates

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Extracted alfalfa juice was frozen at $-25\text{ }^{\circ}\text{C}$ in a freezing chamber. After being subject to different freezing storage times, green concentrates were obtained by heat treatment, previously adjusting juice pH to 8.0, 8.5, and 9.0, leaving natural pH 6.0 as control. The results obtained show that at those alkaline pH values of the juice more total dry matter and nitrogen are recovered in the green concentrate than at the natural pH, although the concentrates have a lower protein content. Also, the recovery of green concentrate decreases as the freezing storage time of the juice increases, possibly due to a change in the conformation of membrane proteins, which is more intense as the storage time is higher.

Obtention of chloroplastic protein concentrates implies selective separation of the two protein fractions, chloroplastic and cytoplasmic, present in the juice. The primary separating methods are ultracentrifugation (Smith, 1966; Byers, 1971; de Fremery et al., 1973), heat treatment, organic solvents flocculation (Slade et al., 1945; Hove and Bailey, 1975; Bray and Humphries, 1978; Singh and Singh, 1985), and flocculation by means of polyelectrolites (Bray and Humphries, 1979; Fiorentini and Galoppini, 1980; Fiorentini et al., 1980).

Separation by means of heat of the two protein fractions is possible due to the fact that chloroplastic proteins coagulate faster at lower temperatures than soluble proteins. The main variants affecting the method are temperature, heating time, and the pH of the juice. Coagulation temperature of the chloroplastic proteins varies between 45 and 60 $^{\circ}\text{C}$ depending on the plant species (Subba Rau et al., 1969; Lexander et al., 1970; de Fremery et al., 1973; Edwards et al., 1975; Nagy et al., 1978). When temperature and/or heating time increase, denaturation of soluble proteins in the supernatant increases (de Fremery et al., 1973); therefore, the best heating techniques are steam injection followed by heat interchange.

With regard to the optimum pH value, some discrepancies are found in the bibliography. De Fremery et al. (1973) and Edwards et al. (1975) employ pH 6.0 and a temperature of 55-60 $^{\circ}\text{C}$ in pilot plant studies. However, a temperature of 50 $^{\circ}\text{C}$ and pH 6.0 are the optimum conditions for proteolysis to occur (de Fremery et al., 1972; Scalet et al., 1984). Arkcoll and Holden (1973) also showed that lipoxygenase activity in alfalfa leaves is maximum near neutrality. Betschart and Kinsella (1973) indicate highest solubility of proteins and better chloroplast rupture when pH is increased. Hood and Brunner (1975) recommend keeping an alkaline reaction during the process to intensify cytoplasmic protein solubility and inhibit proteolytic degradations. Likewise, Livingston et al. (1977, 1984) recommend coagulation at alkaline pH values to reduce the saponin content of the concentrate.

Therefore, we studied the influence of alkaline pH values of the juice upon the quantity recovered and on protein content of the chloroplastic concentrates, in comparison with the coagulation at the natural pH of the juice, 6.0. The maximum pH value chosen was 9.0, since at higher values organoleptic and nutritional alterations appear (Whitaker and Feeney, 1983): transformation of chlorophylls into pheophytins and pheophorbides occurs, with the consequent color change from green to brown (Miller et al., 1984) and the increase of nonenzymatic oxidation of polyphenols catalyzed by metals (Nashef et al., 1977; Finot, 1983; Friedman et al., 1984).

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