

Table III. Volatile Flavor Components of Tangerine Flavor Fraction

Myrcene	α -Sinensal
Limonene	Citronellyl acetate ^a
γ -Terpinene	Neryl acetate
Cymene	Geranyl acetate
β -Elemene	Decyl acetate ^a
Δ -Elemene	1,8- <i>p</i> -Menthadien-9-yl acetate ^a
Octanal	Octanol
Decanal	Linalool
Dodecanal	Thymol methyl ether
Neral	Thymol
Geranial	Elemol
Citronnellal	

^a Newly reported tangerine oil component.

Thus, GPC provided a method for separating a fraction of citrus color pigments which may be useful for enhancing color of food products, a flavonoid fraction, and a flavor fraction free of high boiling materials.

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Received for review July 29, 1976. Accepted November 23, 1976. Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture, and does not imply its approval to the exclusion of other products that may also be suitable.

Relationship of Alkane and Alkene Long-Chain Hydrocarbon Profiles to Maturity of Sweet Oranges

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Juice sacs from early-, mid-, and late-season orange cultivars were collected monthly over an 11-month growing period. Degrees Brix and acid values were assayed so that the maturity dates of the three cultivars could be ascertained. Long-chain hydrocarbons from the freeze-dried juice sacs were extracted with hexane and separated into alkanes and alkenes by AgNO₃-thin-layer chromatography. Alkane and alkene profiles were determined by gas chromatographic analyses. Relationships between the hydrocarbon profiles and the maturity dates of the three cultivars were determined.

Defining the legal maturity of citrus fruits for marketing has been important to the citrus industry. Because of the numerous varieties of citrus fruit and the differing times at which they mature, specific criteria were developed for regulating the time at which each could be legally harvested. In Florida, five standards are used to define legally mature fruit (USDA, 1969), viz. (1) color break, (2) minimum juice content, (3) minimum percentage of total soluble solids (TSS), (4) minimum acid content, and (5) total soluble solids/acid ratio (degrees Brix/acid).

Citrus fruit, unlike fruit from deciduous trees, ripen very little after being removed from the tree. Often the interior of the fruit will meet all legal requirements for maturity while the peel is still green. Under these conditions the peel, for aesthetic purposes, may legally be degreened by treatment with chemicals, e.g., ethylene. Ethanol, a metabolite in the fruit, has been shown to be quantitatively related to maturity of the fruit (Davis, 1970). The question

remains unanswered as to whether the legal standards of fruit maturity can be related to chemicals other than ethanol.

We have shown that lipids extracted from citrus juice sacs contain long-chain hydrocarbons. These were primarily saturated (alkane) and monounsaturated (alkene) in nature (Nagy and Nordby, 1971). Gas-liquid chromatography (GLC) showed that these hydrocarbons might be used as markers for chemical classification of citrus fruit (Nordby and Nagy, 1974). A further study showed alkanes and alkenes to be present in the epicuticular waxes of citrus peel and leaf (Nagy et al., 1975). In these two tissues, hydrocarbon profiles changed during tissue maturation.

Citrus juice sacs are held together by an epicuticular wax which can easily be extracted with such solvents as hexane or chloroform (Fahn et al., 1974; Shomer and Ben-Gera, 1975). Preliminary studies indicated that this wax contains essentially all of the long-chain hydrocarbons reported present (Nagy and Nordby, 1971) in the juice sac. Furthermore, we determined that these long-chain alkanes and alkenes could be uniformly extracted from either freshly sectionized fruit or from freeze-dried juice sacs.

The present study was undertaken to determine whether the composition of epicuticular hydrocarbons changes

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Table I. C₂₀ to C₃₃ Saturated Alkanes in Orange Juice Sacs (Relative Percent)

Hydro-carbon	Hamlin				Pineapple				Valencia			
	Mean	July 1971	May 1972	Change ^c	Mean	July 1971	May 1972	Change	Mean	July 1971	May 1972	Change
Br ^a 23	0.9	3.2	5.1	-8.1**	13.7	13.6	9.9	-3.7**	10.6	12.1	8.9	-3.2**
L ^b 23	18.6	19.9	15.7	-4.2**	18.2	17.7	17.9	+0.2	18.5	17.6	17.9	+0.3
Br 24	9.4	11.6	5.7	-5.9**	11.6	12.0	9.0	-3.0**	9.3	10.2	8.1	-2.1**
L 24	5.3	5.0	5.2	+0.2	5.0	4.9	5.1	+0.2	5.7	5.4	6.7	+1.3*
Br 25	14.2	14.7	12.9	-1.8**	16.4	16.7	16.6	-0.1	15.2	15.4	15.1	-0.3
L 25	17.7	16.1	20.7	+4.6**	14.9	14.5	17.0	+2.5**	17.2	15.9	17.5	+1.6**
Br 26	6.6	7.0	6.3	-0.7	7.2	7.9	7.2	-0.7	7.0	7.5	7.1	-0.4
L 26	1.8	1.5	2.4	+0.9**	1.2	1.2	1.3	+0.1	1.7	1.8	2.1	+0.3
Br 27	3.0	2.9	3.3	+0.4**	3.0	3.3	3.2	-0.1	3.5	3.5	4.1	+0.6**
L 27	3.1	2.1	4.5	+2.4**	1.6	1.6	1.7	+0.1	2.4	2.3	2.7	+0.4
Other Br	3.0	1.4	3.4	+2.0**	3.0	3.3	3.2	-0.1	4.0	3.8	5.0	+1.2**
Other L	6.4	4.6	14.8	+10.2**	4.3	3.3	7.9	+4.6**	4.9	4.5	4.8	+0.3

^a Br = branched; includes iso and anteiso structures. ^b L = linear structure. ^c **, 1% level of significance; *, 5% level of significance.

during fruit maturation, and whether the changes correlate with maturity as determined by Brix/acid ratios (B/A). Good correlation could provide the basis for a new approach for determining maturity.

EXPERIMENTAL SECTION

Hamlin, Pineapple, and Valencia oranges were obtained from USDA's Whitmore Experimental Farm (Crops Research Division, USDA, Orlando, Fla.). Fruit were collected monthly from late July 1971 to late May 1972 for Hamlins and Pineapples, and in late June 1972 for Valencias. Care was taken that the fruit were collected from all parts of the trees. Degree Brix and acid values were determined on three fruit from each cultivar by official analytical methods (AOAC, 1970). Thirty fruit were cut in half and the intact juice sacs (vesicles) were carefully separated from core, peel, seeds, and carpellary membrane with the aid of a citrus spoon. Juice sacs were freeze-dried to a powder with a moisture content no greater than 4% and stored at 5 °C until extracted for wax with hexane. A 20-g sample of powder was placed in a 500-ml flask and shaken 1 min with 50 ml of hexane and 5 ml of water. The hexane extract was decanted, and the wet powder was extracted twice more with 50-ml portions of hexane. The combined extracts were dried over Na₂SO₄ and reduced to dryness on a rotoevaporator. The residue dissolved in hexane-cyclohexane (1:1) was streaked on 250- μ m silica gel plates which had been impregnated with AgNO₃ and activated 1 h at 107 °C (Nordby and Nagy, 1971). The plates were developed at room temperature in chambers lined with filter paper in petroleum ether-ethyl ether (98:2). These conditions clearly separated the long-chain alkanes (av R_f 0.73) and the monounsaturated alkenes (av R_f 0.60) from the other lipids. The plates were sprayed with Rhodamine 6G and viewed under ultraviolet (UV) light. The two hydrocarbon bands were eluted with ethyl ether and stored in hexane for GLC. Gas chromatographic analyses of the long-chain hydrocarbons were performed with a Hewlett Packard Model 7610A gas chromatograph equipped with a flame ionization detector. Hydrocarbons were determined on a glass column (3.05 m \times 4 mm i.d.) packed with 3% SP-1000 (Supelco, Inc., Bellefonte, Pa.) on 100-120 mesh, Gas-Chrom Q. The injection port and detector were at 250 °C and the helium flow rate was 80 ml/min. The sample was injected on a column at 140 °C programmed at 2 °C/min to 220 °C then at 1 °C/min to 240 °C. Alkanes and alkenes were quantitated with an Autolab System IV electronic integrator. For each stage of fruit maturity (monthly sampling), two separate samples were obtained and each of these was, in turn, analyzed in triplicate by GLC.

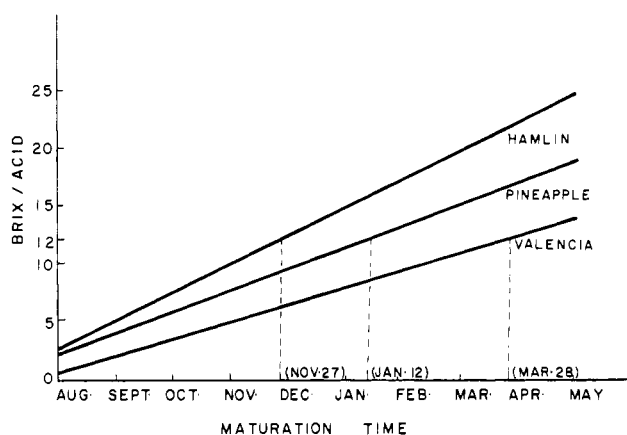


Figure 1. Brix/acid ratios vs. maturation time. Linear regression lines of Hamlin, Pineapple, and Valencia oranges.

RESULTS AND DISCUSSION

Brix/acid ratios were calculated each month over an 11-month maturation period for the Hamlin, Pineapple, and Valencia orange cultivars. When these ratios were plotted against maturation times, linear regression lines with correlation coefficients (r^2) of 0.99, 0.98, and 0.97 for Hamlin, Pineapple, and Valencia oranges, respectively, were obtained (Figure 1). A B/A of 12.0 is considered a reliable indicator of legally acceptable orange juice (USDA, 1969). A horizontal line drawn through this value intercepts the three regression lines at various maturity dates. For Hamlin, Pineapple, and Valencia, the intercepted dates are Nov 27, Jan 12, and March 28, respectively. These dates agree quite well with those on which these three citrus fruits mature (B/A ratio of 12.0) each year in Florida (Harding et al., 1940).

The level of hydrocarbons in the orange juice sacs remained fairly constant over the maturation period, as observed from the constant size of the GLC profiles. The approximate ratio of alkanes to alkenes in the epicuticular wax of juice sacs was 15:1, as reported previously (Nagy and Nordby, 1973). Table I lists the relative percentages of the major alkanes found in the juice sacs at the beginning (July 31, 1971) and end (May 31, 1972) of the maturation period. Initially (July 1971), the C₂₃ to C₂₇ alkanes comprised ca. 92 to 94% of the total alkanes. From 40 to 45% of these were linear. This percentage is typical for orange cultivars (Nagy and Nordby, 1973); differences among Hamlin, Pineapple and Valencia were slight. The rest of the C₂₃ to C₂₇ alkanes had iso- or anteiso-branched structures. Ratios of these two isomers of equal chain lengths were consistent with those published

Table II. Linear Regression Coefficients and Estimated \hat{y}^a Values for Branched C_{23} in Hamlin, Pineapple, and Valencia Oranges over Varying Maturation Periods

Maturation interval	Cumul. months	Line intercept (A_0)			Slope ($A_1 \times 10^{-3}$)			Rel % BR C_{23} (\hat{y})		
		Hamlin	Pine-apple	Valencia	Hamlin	Pineapple	Valencia	Hamlin	Pine-apple	Valencia
July 1971-Sept 1971	2	13.1	13.8	11.9	-5.2	+20.0	-21.7	12.9	14.9	10.7
July 1971-Oct 1971	3	13.0	14.1	11.6	+1.7	+3.4	-8.1	13.2	14.5	10.8
July 1971-Nov 1971	4	13.0	14.3	11.6	+1.4	-2.4	-8.6	13.2	14.0	10.6
July 1971-Dec 1971	5	13.2	14.1	11.5	-3.4	+2.1	-7.3	12.7	14.4	10.4
July 1971-Jan 1972	6	13.6	14.2	11.4	-10.6***	+1.2	-5.4	11.5	14.4	10.4
July 1971-Feb 1972	7	13.7	14.1	11.2	-13.4	+2.9* ^b	-2.0	10.9	14.7	10.8
July 1971-March 1972	8	13.9	14.3	11.1	-15.7	-0.8	+0.1	10.0	14.1	11.1
July 1971-April 1972	9	14.2	14.6	11.1	-19.4	-3.6	-0.4	8.8	13.6	11.0
July 1971-May 1972	10	14.6	15.0	11.3	-24.1	-8.7**	-2.0*	7.3	12.4	10.7
July 1971-June 1972	11			11.4			-3.8**			10.2

^a $\hat{y} = A_0 + A_1X$, where \hat{y} = relative percentage of branched C_{23} alkanes and X = maturation time. ^b * and ** = A_1 and following A_1 values significantly different (t values) from the preceding cumulative slope at the 5 and 1% levels, respectively.

for orange juice sacs (Nagy and Nordby, 1973). Differences among the branched structures of the three orange cultivars were minimal and, thus, are listed as combined branched alkanes (Table I).

The percentage of each hydrocarbon definitely tended to either decrease or increase with fruit maturation. The rate of the change, however, differed noticeably with each cultivar and was static over specific time intervals. Erratic fluctuations between consecutive months were absent. The mean percentage of each of the C_{23} to C_{27} alkanes for the 11-month sampling period is listed in Table I for each cultivar. Generally, differences between the mean and the initial values for each cultivar were minor. The most noticeable change was a 2.3% decrease of both branched C_{23} and C_{24} alkanes in Hamlin oranges. The changes between the alkane percentages for July 1971 and May 1972 varied significantly (Table I, Change column). They also varied in extent with orange cultivar. The changes were greatest for Hamlin, the earliest maturing orange (October-December), intermediate for the mid-season Pineapple orange (January-March), and least for the late-season Valencia orange (late March-June). Decreases in the percentages of branched C_{23} and C_{24} alkanes were consistently significant ($P < 0.01$). Percentages of the branched C_{25} and C_{26} also decreased, but the decreases were generally not significant (an exception was branched C_{25} for Hamlin). The increases in linear C_{25} were significant ($P < 0.01$) for all three cultivars, but those in linear C_{26} and C_{27} were significant only for Hamlin. Changes were significant ($P < 0.01$) in eight specific alkanes for Hamlin, three for Pineapple, and four for Valencia.

Table I also lists the total percentages of the other linear and branched alkanes in the C_{20} to C_{33} alkane spectrum. Within this minor combined group, increases in the relative percentages of C_{28} to C_{33} linear alkanes were noticeable (data not shown). Hamlin showed the greatest change with increases in C_{29} (3.5%), C_{31} (6.3%), and C_{33} (1.2%) between July 1971 and May 1972. The percentage of branched C_{23} changed the most over the 11-month maturation period for each of the three orange cultivars (Table I). To determine whether the percentages of these branched C_{23} alkanes changed linearly over any duration of the 11

months we calculated linear regression coefficients at cumulative monthly periods. Calculations were begun with values for the first 2 months (July-September 1971). Thereafter, values for the consecutive months were included (cumulative months). Line intercept (A_0), slope (A_1), and estimated percentage of branched C_{23} (\hat{y}) are listed in Table II for these cumulative periods.

The A_0 values for Hamlin and Pineapple were fairly constant over the July 1971-November 1971 and July 1971-February 1972 maturation periods, respectively. The A_0 values for Valencia, however, decreased throughout the July 1971 to April 1972 periods.

During these same maturation periods, the A_1 values for Hamlin and Pineapple were also reasonably constant. The initial high A_1 values for the July 1971-September 1971 period were due to unexplained, inconsistent high values obtained from the July fruit. Fruit obtained during July were very small, slightly larger than a golf ball. It might be that the fruit obtained at this very early maturity stage had not developed a uniform epicuticular wax layer on the juice sac. A_1 values for Valencia increased. The value for the July 1971-September 1971 period was also erratically high, probably because of the July fruit.

The estimated percentages of branched C_{23} (\hat{y}) were obtained from the formula $\hat{y} = A_0 + A_1X$, the values of X being the interval time period (in days). These values remained relatively static for Hamlin (mean ca. 13.0%) during the July 1971-November 1971 period, for Pineapple (mean ca. 14.6%) during the July 1971-February 1972 period, and for Valencia (mean ca. 10.7%) during the July 1971-April 1972 period.

For each cultivar, the maturation time at which the relative percentages of branched C_{23} began to change might be regarded as the "transition period". These periods are indicated by the boxed \hat{y} values in Table II. These \hat{y} transition periods corresponded reasonably well to the transition periods of line intercept and slope values. Beyond these transition periods, steadily increasing values of A_0 and decreasing values of A_1 were indicated (Table II). These transition periods also corresponded to the maturity dates calculated from the Brix/acid vs. time plot shown in Figure 1.

Table III. Estimated^a and Observed^b Relative Percentages of Linear Alkenes in Orange Juice Sacs at the Beginning (July 1971) and End (May 1972) of Growing Season and the Percent Change^c over This Period

Alkene	Hamlin					Pineapple					Valencia				
	July 1971		May 1972		Change ^c	July 1971		May 1972		Change ^c	July 1971		May 1972		Change ^c
	Est ^a	Obsd ^b	Est ^a	Obsd ^b		Est ^a	Obsd ^b	Est ^a	Obsd ^b		Est ^a	Obsd ^b	Est ^a	Obsd ^b	
23	16.7	15.9	1.6	1.3	-14.6**	19.7	13.8	6.1	4.2	-9.6	18.3	19.6	5.8	4.8	-14.8*
24	2.6	2.3	0.4	0.2	-2.1*	2.5	2.2	1.1	0.8	-1.4	2.3	1.8	1.4	0.8	-1.0
25	42.6	41.6	6.6	5.5	-36.1**	40.1	37.2	22.7	18.8	-18.4*	39.6	38.2	34.0	30.3	-7.9
26	4.3	4.4	0.9	0.4	-3.8*	3.3	3.8	2.6	2.4	-1.4	3.5	2.7	3.4	3.1	+0.4
27	26.2	28.1	6.6	4.4	-23.7*	27.3	31.2	20.7	21.3	-9.9	28.0	28.9	32.8	35.1	+6.2
29	5.1	4.7	16.4	13.2	+8.5*	4.7	6.2	14.3	14.8	+8.6*	4.7	4.8	9.4	10.7	+5.9*
31	tr ^d	0.9	46.5	47.8	+46.9**	0.5	3.0	25.7	29.4	+26.4*	1.9	2.4	9.6	11.7	+9.3*
33	tr	tr	16.1	21.8	+21.8*	tr	0.3	3.9	5.1	+4.8*	tr	tr	0.7	0.5	+0.5
Σ_{23}^{33} (odd numbered)	90.6	91.2	93.8	94.0	+2.8	92.3	91.7	93.4	93.6	+1.9	92.5	93.9	92.3	93.1	-0.8
Σ_{21}^{32} (even numbered)	9.4	8.8	6.2	6.0	-2.8	7.7	8.3	6.6	6.4	-1.9	7.5	6.1	7.7	6.9	+0.8

^a Estimated from linear regression lines. ^b Observed from GLC analyses. ^c Change in observed GLC values over this period. **, r^2 greater than 0.90; *, r^2 greater than 0.70. ^d Trace, less than 0.1%.

Table IV. Relative Percent Monoenes in Three Orange Cultivars at Three Maturity Dates

Monoene	Hamlin			Pineapple			Valencia			Maturity range
	Nov 27, 1971	Jan 12, 1972	March 28, 1972	Nov 27, 1971	Jan 12, 1972	March 28, 1972	Nov 27, 1971	Jan 12, 1972	March 28, 1972	
C ₂₃	11.4 ^a	8.9	4.9	14.3	12.2	8.8	13.4	11.5	8.3	8.3-12.2
C ₂₄	1.8	1.5	0.9	1.9	1.7	1.4	2.0	1.8	1.6	1.6-1.8
C ₂₅	30.0	24.2	14.4	33.2	30.5	26.2	37.3	36.5	35.1	30.0-35.1
C ₂₆	3.1	2.4	1.6	3.1	3.0	2.8	3.5	3.4	3.4	3.0-3.4
C ₂₇	19.5	16.3	11.0	24.7	23.7	22.0	30.0	30.7	31.8	19.5-31.8
C ₂₉	9.9	11.6	14.4	8.5	10.0	12.3	6.6	7.3	8.5	8.5-10.0
C ₃₁	16.8	24.4	37.0	10.6	14.4	20.6	4.9	6.1	8.0	8.0-16.8
C ₃₃	2.3	5.8	11.7	1.4	2.1	3.1	0.3	0.4	0.6	0.6-2.3

^a Values underlined are expected values at maturity for the respective orange cultivar.

These transition periods also corresponded to times at which other major alkenes showed percentage changes from a relatively static state to one of noticeable changing percentage patterns (increasing or decreasing in relative percentage). Significant changes in the percentages of alkenes within the epicuticular wax layer of juice sacs indicate that the fruit was undergoing a change from a static, equilibrium condition to one of senescent catabolism.

During the selected maturation time intervals the alkane percentages were relatively static (e.g., branched C₂₃ in \hat{y} column in Table II). In contrast, the percentages of the major alkenes in all three cultivars constantly changed during the 11 months. Alkenes either uniformly increased or decreased; no erratic fluctuations or static periods were evident.

Table III lists the relative observed and estimated percentages of the major linear monoenes. The C₂₀ and C₂₁ alkenes were found present at concentrations less than 0.01% and, therefore, were excluded from the table. The estimated values were obtained from linear regression plots for each alkene over 11 months. Branched monoenes comprised a relatively minor percentage of the total monoene fraction (Nagy and Nordby, 1972; Nagy et al., 1975) and, therefore, were excluded from this study.

Initially (July 1971), the observed percentage differences among specific alkenes of the three cultivars were no greater than 6 percentage points (observed C₂₃ between Pineapple and Valencia); however, by the end of May 1972 differences were as great as 36 percentage points (observed

C₃₁ between Hamlin and Valencia). Generally, the percentages of C₂₂ to C₂₈ alkenes decreased during maturation while those of C₂₉ to C₃₃ increased.

Over 90% of the monoenes were odd-numbered and their percentages changed the most over the maturation period. The percentage of total odd-numbered alkenes increased by ca. 3 percentage points for Hamlin and by smaller amounts for Pineapple and Valencia. Changes in relative percentages of the C₂₃ to C₃₃ alkenes appeared to follow specific patterns which were intrinsic for each cultivar. Changes in percentages for Hamlin were as much as four times the corresponding changes that occurred for Pineapple and as much as eight times those that occurred for Valencia. All changes, however, seemed to be linear as shown by the close agreement between the estimated and observed relative percentages in Table III.

Table IV shows the maturity dates obtained from Brix/acid plots of Figure 1 and regression estimates of the percentages of eight major monoenes on these respective maturity dates. The expected percentage of each monoene for each fruit at maturity is underscored. The ranges of these underscored values are also listed.

The relative percentage values for Hamlin and Pineapple were surprisingly similar at their respective maturity dates. The largest difference (4.2 percentage points) was in C₂₇. The values for Valencia at its estimated maturity date differed from those for the other cultivars by as much as 12.3 percentage points (Hamlin C₂₇, 19.5%, vs. Valencia C₂₇, 31.8%). Differences were also noticeable in C₂₅, C₃₁, and C₃₃. A comparison of the relative percentage values

Table V. Ratios of Alkenes Present in Orange Juice Sacs at Early-, Mid-, and Late-Season Maturity Dates

No.	Alkene ratio	Hamlin			Pineapple			Valencia			Maturity range
		Nov 27, 1971	Jan 12, 1972	March 28, 1972	Nov 27, 1971	Jan 12, 1972	March 28, 1972	Nov 27, 1971	Jan 12, 1972	March 28, 1972	
1	23/29	<u>1.1^a</u>	0.8	0.3	1.7	<u>1.2</u>	0.7	2.0	1.6	<u>1.0</u>	<u>1.0-1.2</u>
2	23/31	<u>0.7</u>	0.4	0.1	1.3	<u>0.8</u>	0.4	2.7	1.9	<u>1.0</u>	<u>0.7-1.0</u>
3	25/23	<u>2.6</u>	2.7	3.0	2.3	<u>2.5</u>	3.0	2.8	3.2	<u>4.2</u>	<u>2.5-4.2</u>
4	25/26	<u>9.7</u>	9.5	8.8	10.8	<u>10.3</u>	9.4	10.8	10.6	<u>10.3</u>	<u>9.7-10.3</u>
5	25/29	<u>3.0</u>	2.1	1.0	3.9	<u>3.1</u>	2.1	5.7	5.0	<u>4.1</u>	<u>3.0-4.1</u>
6	25/31	<u>1.8</u>	1.0	0.4	3.1	<u>2.1</u>	1.3	7.6	6.1	<u>4.4</u>	<u>1.8-4.4</u>
7	26/24	<u>1.7</u>	2.1	1.9	1.6	<u>1.7</u>	2.0	1.8	1.9	<u>2.1</u>	<u>1.7-2.1</u>
8	27/29	<u>2.0</u>	1.4	0.8	2.9	<u>2.4</u>	1.8	4.5	4.2	<u>3.8</u>	<u>2.0-3.8</u>
9	27/31	<u>1.2</u>	0.7	0.3	2.3	<u>1.6</u>	1.1	6.1	5.0	<u>4.0</u>	<u>1.2-4.0</u>
10	31/33	<u>7.3</u>	4.2	3.2	7.4	<u>7.0</u>	6.7	19.8	16.3	<u>14.3</u>	<u>7.0-14.3</u>

^a Values underlined are values at maturity for the respective orange cultivar.

for the three cultivars at maturity showed that the monoene percentages fell within specific ranges for C₂₄, C₂₅, C₂₉, C₃₁, and C₃₃. It is not possible, however, to employ this criteria to determine whether a fruit from a specific orange cultivar is mature. For example, the Nov 27 and March 28 C₂₇ monoene percentage values for Pineapple orange were both within the estimated maturity range for this alkene.

In previous studies on citrus, ratios of hydrocarbons were shown to be quite useful for chemotaxonomic purposes (Nagy and Nordby, 1972). The ratios of the eight major monoenes analyzed in this study were calculated, and the ten most promising ratios are presented in Table V. An orange cultivar, for example, with a C₂₃/C₂₉ ratio between 1.0 and 1.2 would be ripe. Further evidence for this cultivar's ripeness would be obtained if the C₂₃/C₃₁ ratio was found to be between 0.7 and 1.0. Monoene ratio no.'s 3, 5, 6, 7, and 10 can be used for early and mid-season fruit. If the orange cultivar in question had a 25/26 ratio (no. 4) less than 9.7 or greater than 10.3 and C₂₃/C₂₉ and C₂₃/C₃₁ ratios less or greater than their respective maturity ranges, the cultivar should mature before Nov 27 or after March 28. This same reasoning would apply for ratio no.'s 8 and 9 but with a less degree of certainty. If the 31/33 ratio (no. 10) for an orange cultivar was above 8.0, this value would indicate the fruit was late maturing.

Evidence has been presented that the hydrocarbons in the wax coating of citrus juice sacs change in composition during maturation of the fruit. Alkane profiles did not change appreciably until about 1 month after the fruit became legally mature (Brix/acid). Thus, over an 11-month growing period changes in alkane composition were greater for Hamlin, an early-maturing fruit, than Valencia, which matured in the ninth month of this period. Whether this so-called transition period is typical of citrus in general is still to be determined. The relative monoene percentages

underwent changes at an approximated linear rate throughout the entire 11 months. Relative percentage values for oranges that ripen at various times of the year fall within certain ranges. Values falling outside these ranges would indicate an orange was a very early or very late maturing fruit. Ratios of one monoene to another give narrower maturity ranges than relative percentage values. The data presented show the relative relationship of citrus wax hydrocarbons to maturity. Applications of these relationships might lie in the areas of maturity determination of new hybrids, analysis of adulterants blended into pure juice products, e.g., mandarin in orange juice, and chemotaxonomy of citrus.

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