# FACTORS AFFECTING THE RATE OF MATURATION OF CITRUS FRUITS

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Abstract. 'Washington navel' and 'Valencia' [Citrus sinensis (L.) Osb.] sample data (Brix/acid ratios) from routine processing plant tests were correlated directly with local climatic data throughout 7 seasons in California to yield linear relationships with r² factors of 0.96 or better for navels. The y-intercepts and slopes were found to strongly correlate with the accumulated maximum heat from the first 4 months after bloom, crop load, and tree growth with r² factors of 0.96. Equations were derived to enable (Brix/acid) ratio prediction. Average monthly limonin levels were also correlated with heat and crop loads for both 'navel' and 'Valencia' oranges. Other climatic factors and biological maturation processes are discussed.

The elucidation of the factors affecting maturation is important to any fruit industry, but especially in the citrus industry where the maturation is a very slow process, taking from 7 months to over a year for orange varieties. The primary indicator of maturity is the ratio of the % soluble solids to the % titratable acidity as citric acid, which must be at least 8:1 in California. Work has been done to try to determine what factors affect the development of this ratio for over 80 yr, with no real applicable results for the industry. The effects of rootstock, varieties, irrigation, dormancy, moisture stress, heat, location of the fruit on the tree, radiation, and other parameters were discussed in detail by Reuther (9). Recognizing the contributions from all these parameters to the maturation rate, heat and tree strength seem to be the dominant factors under normal conditions. To date, except for previous work by the author (5), no correlation has been attempted to relate the heat as it develops throughout a season to the corresponding maturation. This work proceeds further with correlations between the maturation parameters and climatic parameters, and also the corresponding relationships with known biological processes. Also, limonin, a bitter compound in processed citrus juices, is compared to climatic data. Little work of any kind has been done with seasonal limonin trends (1, 4).

Even though data presented here is characteristic of citrus grown in California, the correlation techniques can be applied to any citrus growing area to determine similar relationships.

#### **Materials and Methods**

When fruit is brought into a processing plant, 30-40 lb. samples are analyzed for quality, including the % soluble solids measured as Brix by refractometer or hydrometer, the % titratable acidity as citric acid determined by standard methods (6), and the ratio of the two, or the B/A ratio, as well as the yield, so that the owner can obtain a proportional credit for his fruit. Data from these tests were processed into monthly averages from 3 packinghouses,

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one sample a day for every day that fruit was received from that particular packinghouse, and correlated with the accumulated temperatures (F°) during a given period. Climatic data were obtained from Lindsay-Strathmore Irrigation District, which is centrally located in Tulare county's citrus belt. Crop data were obtained from the Tulare County Agricultural Commissioner's office. Limonin analyses were done by mixing reconstituted juice with diatomaceus earth, extracting with five 30-ml portions of chloroform using vacuum filtration, and evaporating the filtrate in a rotary evaporator. The flask from the rotary evaporator was rinsed with several 2 ml portions of chloroform and analyzed, using a Perkin Elmer series 10 HPLC pump, LC-75 UV detector set at 210 nm, and a model R100 chart recorder. A CN column and precolumn were used with a solvent system consisting of ethylene glycol monomethyl ether, 2-propanol, and n-heptane in the proportion of 10:15:25.

#### **Results and Discussion**

Accumulated degree-days have long been used to estimate the amount of heat applied to commercial agricultural crops (2). This practice assumes a sinosoidal temperature profile during a 24-hr period, with the average temperature between the maximum and minimum representing the overall average temperature. Use of the same groves year after year keeps such variables as rootstock, variety, soil types, and general agricultural practices essentially constant. It also minimizes the affect of acclimation of the trees to varying climatic conditions which is a major factor in comparing growth patterns in 2 different geographical locations.

Organic acid changes. Citrus maturation consists of basically a growth stage and a maturation stage. During the growth stage organic acids accumulate in the fruit. This has been attributed to early blockage of the citric acid cycle, where citric acid is produced, by either a lack of or inhibition of the aconitase enzyme which would normally break down the citric acid (7). Vines and Metcalf (12) demonstrated that in grapefruit, mitochondrial activity increases into maturity and then declines, while Ramakrishnan (7) demonstrated that in lemons no respiratory activity occurs after maturity. Bennet (USDA Fruit and Vegetable Laboratory, Pasadena, California, presented to the Citrus Products, Technical Committee, June 1984, Pasadena, CA) illustrated the probable activity of mitochondria in lemons by recording on video a microscope probe that slowly penetrated a living juice vesicle; the mitochondrial movement in channels of the juice cell cytoplasm were rather energetic. In oranges, lack of mitochondrial activity and/or lack of or inhibition of the aconitase enzyme is the most likely cause of organic acid build-up during the growth phase. As water accumulates in the vacuoles of the juice cell, these acids are absorbed along with the water by osmotic force, which separates them from the mitochondrial activity in the cytoplasm. Sinclair and Ramsay (10) illustrated that even though the concentration of the citric acid decreases during maturation, the absolute amount of the organic acids remains essentially unchanged. The decrease in acid concentration upon maturation is at least partially the result of an increase in the size of the orange and the accumulation of water in the fruit which dilutes out the acid concentration. Flavonoids have been found to undergo a similar dilution effect upon maturation (B. Carter, Ventura Coastal Corp., personal communication). However, the fruit growth data

from Bain (3) does not seem to account for all the reduction of acid concentration. The vacuoles appear to store the acids for use in the citric acid cycle during times of high respiratory demand, which may cause a further reduction of acids to occur. This change in acid concentration during maturation is the primary factor in the development of the B/A ratio.

Fig. 1 is a plot of the monthly average % titratable acidity of fruit samples from 3 packinghouses, versus the accumulated temperatures from July 1, just after bloom, up to the end of the harvesting month for 7 seasons for 'Washington navel. Fig. 2 is a similar plot for 'Valencia'. The smooth curves for the navel plots suggest an acid concentration peak sometime during September or October and a strong correlation with heat. The heat here is probably more directly related to fruit growth and an increased capability of the fruit to absorb and retain water, which in turn, as previously mentioned, is related to dilution of the acids. In Fig. 2, the 1981-82 season and the 1983-84 season are omitted because of the small crop and corresponding short season. Since the B/A ratio is inversely related to the acid concentration, a plot of the reciprocal of the % acid versus the accumulated heat might be more illustrative of its effect on the B/A ratio development. Such plots are found in Fig. 3 and 4. In Fig. 3 the linearity is readily apparent. In Fig. 4 the reciprocal of the % acid seems to remain somewhat constant until the latter portion of the season, where it in-

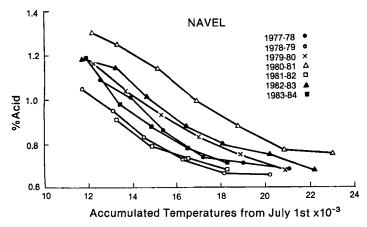


Fig. 1. Changes in % acid with accumulated temperatures from July 1 just after bloom for navels.

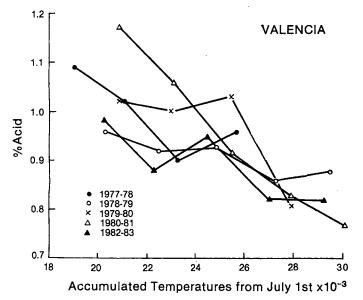


Fig. 2. Changes in % acid with accumulated temperatures from July I just after bloom for 'Valencia'.

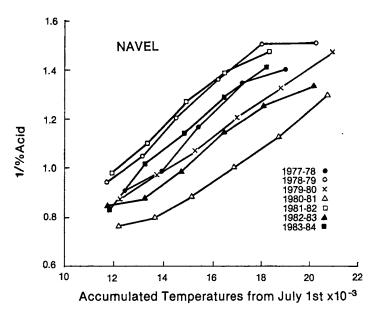


Fig. 3. Changes in the reciprocal of the % acid with accumulated temperatures from July 1 just after bloom for navels.

creases. Increased transpiration in the early summer and/or increased irrigation in late summer may partially account for this trend. In addition, temperatures in July and August probably exceed the compensation point where respiratory demand is high, which would then cause the fruit to draw on the citric acid reserve in the juice cell vacuoles, reducing the acid concentration.

Sugar development. The sugar accumulation in citrus fruit is primarily a function of temperature and light intensity in the photosynthetic process. Fruit that is grown at the top of the tree or on the sides where it is exposed to sunlight grow sweeter more quickly than shaded fruit. Even the stylar half of the fruit is sweeter than the stem half. Although citrus has been shown to be dormant at temperatures below 54°F and above 97°F (3), soluble solids have been shown to continue to increase even when fruit ceases to grow (8). And while the fruit itself can undergo photosynthesis in the peel, most of the carbohydrates are translocated from other parts of the plant. This translocation has its own set of kinetics, which further complicates the sugar development (14).

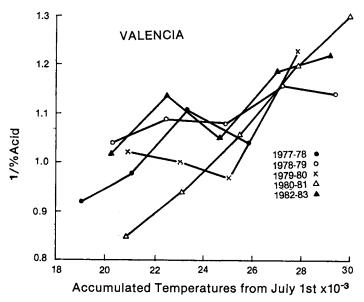


Fig. 4. Changes in the reciprocal of the % acid with accumulated temperatures from July 1 just after bloom for 'Valencia'.

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Fig. 5 and 6 are plots of the monthly average Brix from the previously mentioned fruit samples versus the same heat data in the previous figures. Again, the 1981-82 and 1983-84 'Valencia' data are omitted. In both plots the Brix during the 1978-79 freeze year was lower due to freeze damage.

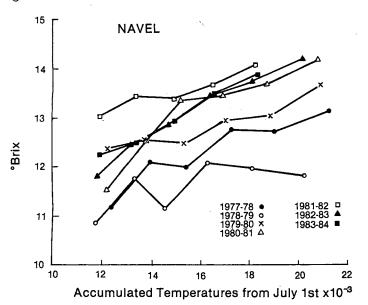


Fig. 5. Changes in Brix with accumulated temperatures from July 1 just after bloom for navels.

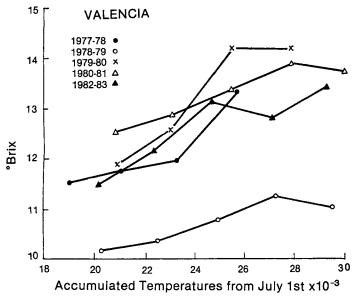


Fig. 6. Changes in Brix with accumulated temperatures from July 1 just after bloom for 'Valencia'.

B/A ratio development. The reason that the B/A ratio is such an important parameter is because the sweetness of the sugars and the sourness of the acids compete for the same receptor sites on the tongue. This means that the absolute amounts of the sugars or acids are not as important as their relative amounts. In Fig. 7 and 8 are the resulting B/A ratio plots taken from the data of the previous figures. In Fig. 7, strong linear relationships are evident which are primarily affected by the linearity of the plots in Fig. 3. The large deviation in March of the 1977-78 season was the result of an unusually high level of precipitation during that month and preceding months. The increased precipitation caused the fruit to absorb more water which further diluted the acids. When the soil moisture returned to normal, the

excess water in the fruit disappeared and the B/A ratio trend returned to previous levels. In Fig. 8, the B/A ratio is dominated by the sugar development in the first half of the season and by the acid reduction in the latter half of the season; probably due to increased transpiration, irrigation, and/or temperatures exceeding the compensation point.

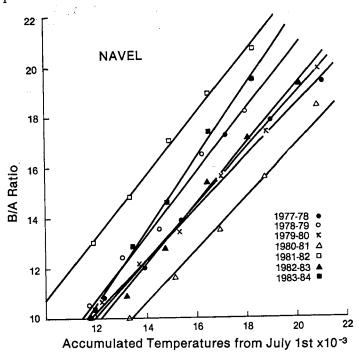


Fig. 7. Linear regressions of the change of the (B/A) ratio with the accumulated temperatures from July 1 just after bloom for navels.

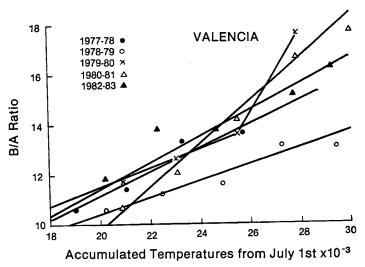


Fig. 8. Linear regressions of the change of the (B/A) ratio with the accumulated temperatures from July 1 just after bloom for 'Valencia'.

The strong linear relationships between the B/A ratio and the accumulated heat in Fig. 7 raises the question of what parameters might affect the slopes and y-intercepts. The y-intercepts were found to strongly correlate linearly with the accumulated maximum temperatures between May and August just after bloom with a r² factor of 0.96, excluding the freeze year, as follows:

$$Y = 0.006607M - 78.42 \tag{1}$$

where Y is the y-intercept from Fig. 7 and M is the accumulated maximum temperatures explained above. The slopes were found to correlate strongly with the crop load and tree

growth by performing a trivariant least squares analysis (excluding the freeze year) with a r<sup>2</sup> factor of 0.96 with the following result.

$$S = ((C - 2.045n)(-0.03076) + 1.2371) \times 10^{-3}$$
 (2)

where S is the slope from Fig. 7, C is the total harvested crop in tons per acre, and n is the number of the year beginning with 1 for the 1977-78 season. Each year the tree gains strength through growth which enables it to produce a larger crop. In order to determine the strain on the tree caused by the crop load, this added tree strength must be subtracted. The above equation suggests that the average grove can produce over 2 tons per acre more each year regardless of climatic factors. The adjusted crop load then determines the strength of the tree available for fruit maturation which determines the rate of the B/A ratio development. Combining equations 1 and 2, the B/A ratio can be calculated as follows.

$$R_n = HS + Y \tag{3}$$

where R<sub>n</sub> is the B/A ratio for navels, H is the accumulated average temperatures from July 1 just after bloom. Using equation 3, calculated values for the B/A ratio are compared to the measured values in Table 1. The 1983-84 season is omitted because the harvested crop data was not yet available. A statistical t-test comparison was made between the measured and calculated values resulting in a confidence level of 98%. Similar attempts to explain 'Valencia' B/A ratio trends were not successful due to one or more of the following reasons: less representative data, more than one crop being on the tree at one time, a warmer harvesting period, and biological differences.

Table 1. Measured and calculated Bric/acid (B/A) ratios for navel oranges.

Season	Month	Measured B/A ratios	Calculated B/A ratios
1977-78	Dec	10.8	10.8
	Jan	12.0	12.5
	Feb	13.9	14.1
	Mar	17.3	16.2
	Apr	17.9	18.0
1978-79	Dec	10.3	10.6
	Jan	12.4	12.2
	Feb	13.5	13.7
	Mar	16.5	15.6
	Apr	18.2	17.6
1979-80	Dec	10.6	10.5
	Jan	12.1	12.1
	Feb	13.4	13.8
	Mar	15.6	15.5
	Apr	17.4	17.4
	May	19.9	19.5
1980-81	Dec	9.0	8.6
	Jan	10.0	10.2
	Feb	11.8	11.7
	Mar	13.5	13.5
	Apr	15.6	15.5
	May	18.4	17.6
	Jun	19.7	20.1
1981-82	Dec	13.0	13.3
	Jan	14.8	15.0
	Feb	17.0	16.9
	Mar	18.9	18.9
	Apr	20.7	21.1
1982-83	Dec	10.0	8.9
	Jan	10.9	10.6
	Feb	12.8	12.3
	Mar	15.4	14.3
	Apr	17.1	16.3
	May	19.3	18.7
	Jun	21.9	21.2

Limonin changes. Even though limonin levels are not used as extensively in quality determinations of citrus fruit as is the B/A ratio, they are at least equally important to the quality of processed juice products in bitter varieties such as navel oranges and grapefruit (C. paradisi Macf.). As can be seen from Fig. 9, the limonin is high at the beginning of the season and decreases upon maturation. The November limonin levels are low due to the harvest of the early season Bonanza navel. These navels have a 1-2 month harvesting season and mature very quickly, reaching B/A ratios of 20+ in December. These navels are also low in limonin, and when mixed with the 'Washington navel' reduce the combined bitterness. From December on, a consistent decrease in limonin levels is observed. Unlike the acid concentration that dilutes throughout maturation with generally little absolute loss of acids, the limonin levels decrease to essentially zero, indicating that in addition to an expected dilution effect, an actual degradation of limonin occurs. The only correlation found between the trends of both the navel and 'Valencia' data seems to be that with large crops the limonin reduction is slowed down. This is probably due to a decrease in fruit growth characteristic of large crops, which decreases the dilution effect in the limonin levels.

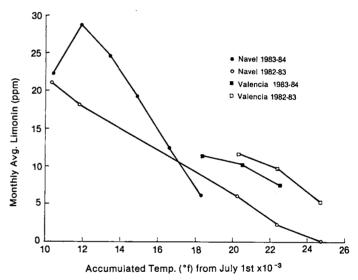


Fig. 9. L'monin changes with accumulated temperatures from July 1 just after bloom for navels and 'Valencia'.

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# TREE SPACING AFFECTS CITRUS FRUIT DISTRIBUTION AND YIELD<sup>1, 2</sup>

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Additional index words. tree density, tree size, harvesting.

Abstract. Fruit distribution within the canopy and yield per acre were affected by spacing of 'Pineapple' orange [Citrus sinensis (L.) Osb.] trees during 5 seasons. Trees were 18 yr old at the beginning of the study and involved spacings of  $20 \times 25$  ft,  $15 \times 20$  ft, and  $10 \times 15$  ft. Fruit distribution was determined by harvesting individually 4 ft zones vertically through the tree and by harvesting inside and outside fruit separately. A greater percentage of fruit was found in the upper parts of the tree at closer spacings. More inside fruit occurred on trees at wider spacings. Higher yields were obtained from trees at closer spacings in this experiment.

Tree spacing has become a very important consideration in citrus plantings. Generally, closer planted trees result in earlier net returns, but at the expense of earlier developing management problems. Growers want early net returns on their investment and maximum returns over the productive life of the planting. Selection of tree spacing to achieve these ends is complex and many of the considerations have been discussed elsewhere (1, 2, 3, 4, 5, 6, 7, 8).

Fruit distribution on the tree is important from the harvesting standpoint. For a given fruit density, fruit within 7 to 8 ft of the ground can be harvested without a ladder and can be harvested faster and easier than fruit more than 8 ft from the ground. Fruit within arm's reach of the outer tree canopy can usually be harvested at an easier and faster rate (fruit/hr) than fruit further inside the canopy because outer fruit is easier to reach and the fruit density (number of fruit per unit volume of canopy space) is generally higher.

The objective of this paper is to report on the effect of 3 different tree spacings on fruit distribution and yield.

### Materials and Methods

'Pineapple' orange trees on rough lemon (Citrus jambhiri Lush.) rootstock were planted in 1960 at spacings of 20 x 25 ft, 15 x 20 ft, and 10 x 15 ft at the Citrus Research and Education Center grove at Barnum City in Central Florida. These spacings are equivalent to 87, 145, and 290 trees per acre, respectively. The trees were frozen back to the soil banks in 1962, and the first season of recorded fruit production was 1967-68. Annual hedging was started in 1966 in the 10 x 15 ft spacing and 1971 in the 15 x 20 ft

<sup>1</sup>Florida Agricultural Experiment Stations Journal Series No. 6027. <sup>2</sup>Cooperative research by the U.S. Department of Agriculture, Agricultural Research Service, University of Florida, Citrus Research and Education Center, Lake Alfred, and the Florida Department of Citrus. spacing. The hedging width between tree rows was a nominal 7 ft near ground level and increased approximately 1 ft per 4 ft of height. Little foliage has been removed from the trees in the 20 x 25 ft spacing. In the 10 x 15 ft spacing, every fifth tree was removed in the row (10 ft spacing) in 1975 to form 4-tree units, resulting in 232 trees per acre. Cultural practices including overhead irrigation were per-

formed uniformly in all tree spacings (3).

Fruit distribution and yields were determined on the 3 tree spacings during five seasons, 1978-79 through 1982-83. These were the 12th through 16th seasons of fruit production. When the fruit was harvested, it was separated by height zones on the tree: 0 to 4 ft, 4 to 8 ft, 8 to 12 ft, and greater than 12 ft above ground. Further, within each height zone, fruit harvested beyond an arm's reach (approximately 3 ft) of the outside canopy was designated as inside fruit.

Four trees (4 replications) each were harvested each season at the 20 x 25 ft and 15 x 20 ft spacings. In the 10 x 15 ft spacings, 4 replications of the 4-tree units were harvested. Within each 4-tree unit, fruit records from the 2 center trees (hedgerow) were kept separate from the 2 end trees adjacent to the space resulting from the tree removal in 1975. It was assumed for this paper that the 2 center trees represented solid hedgerow trees (290 trees per acre); the 2 end trees represented a 10 x 15 ft planting with every third tree in the row removed resulting in 194 trees per acre.

Fruit yields were determined by weighing. Tree canopy height and width measurements were made in 1978-79, 1981-82, and 1982-83. Canopy width measurements were made approximately 4 ft above ground on the east-west (across row) and north-south (in row) directions.

# **Results and Discussion**

In the 20 x 25 ft spacing, the tree height averaged 14.8 ft high and the canopy diameter averaged 17.5 ft in both north-south and east-west directions. Fruit distribution in the first 3 seasons was fairly uniform at 25% to 35% in each of the 3 bottom zones (Fig. 1). In the last 2 seasons, fruit in the 8 to 12 ft zone had increased to 39% and 44%, respectively. Fruit above 12 ft high had increased to 27% of the total by the last season. Inside fruit fluctuated from 19% the first season to 26% the fourth season, then down to 7% the last season. Over the 5 seasons, outside fruit averaged 86% of the total.

Trees in the 15 x 20 ft spacing averaged 14.5 ft in height and the canopy width dimensions averaged 15 ft in the north-south and 14.3 ft in the east-west directions. Vertical fruit distribution was more variable than in the 20 x 25 ft spacing (Fig. 2). One possible reason for this was that hedging removed more tree canopy in the 15 x 20 ft spacing. During the 5 seasons, outside fruit averaged 87% of the total, with a range from 79% to 93%. There was no apparent reason for the high percentage (55%) of

fruit in the 0 to 4 ft zone in 1982-83.