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Annual and Seasonal Changes in Naringin Concentration of Ruby Red Grapefruit Juice

Roger F. Albach,* George H. Redman, and Robert R. Cruse

Juices from Ruby Red grapefruit (*Citrus paradisi* Macf.) from five different groves in the South Texas citrus belt were assayed for the flavonoid bitter component naringin during five successive seasons from 1968-1969 to 1972-1973. The Davis method was used for assaying naringin. Naringin concentration of juice from the same grove and trees fluctuated during the season and varied considerably between crop years. Climatic variations between crop years strongly influenced the Davis-test naringin value and thus bitterness in grapefruit juice extracted by simulated commercial methods. Location also influenced naringin concentration in some but not all crop years. Juice naringin concentration oftentimes increased during February, March, or April after the onset of rapid vegetative growth. Although seasonal trends tended to persist among all groves during any one crop year, the trends and absolute amounts of naringin values showed little consistency between crop years.

The bitterness of grapefruit is due to components of two chemical classes: limonoids and flavonoids (Maier, 1969). Limonin bitterness only develops after juice is extracted from the fruit, and the intensity is greater in juice from fruit harvested early rather than late in the harvest season (Maier et al., 1977).

Among the flavonoids that causes bitterness—naringin, neohesperidin, and poncirin—naringin is by far the most important (Hagen et al., 1965; Horowitz and Gentili, 1977). Because naringin bitterness is one of the main factors that determine acceptability of canned grapefruit juice, several surveys of its concentration have been made (Maurer et al., 1950; Kesterson and Hendrickson, 1953; Tatum et al., 1972; Dougherty et al., 1977; Dougherty and Fisher, 1977; Ting and McAllister, 1977). Horticultural and climatic factors which might influence naringin concentration were not considered. Maurer et al. (1950) and Kesterson and Hendrickson (1953) surveyed the naringin content of different varieties at about monthly intervals during the course of a single season. Maurer et al. suggested that the naringin content of juice from 12 different Texas-grown grapefruit varieties showed rises in juice naringin concentration toward the end of the season. Kesterson and Hendrickson failed to note these rises in the Texas survey and indicated that their own survey of five grapefruit varieties grown in Florida showed no significant decrease in juice naringin concentration until past peak maturity in April. They found no correlation between juice naringin concentration and °Brix, percent acid, or °Brix/acid ratio.

Maurer et al. found that in juice from 12 varieties, naringin concentration varied between 580 and 1880 ppm in mid-October down to a range of 130-390 ppm at the last analysis in late January. The Florida juice surveyed by

Kesterson and Hendrickson showed maxima of 300-370 ppm at any time from November through February for the different varieties. Minima of 150-190 ppm were confined to the last two analyses in April or May. These results contrast with those of Tatum et al. (1972), who found the lowest values mostly toward the beginning of the season and the highest values toward the end. They used the same nonspecific Davis test (Davis, 1947) as Maurer et al. and Kesterson and Hendrickson; their Davis-test naringin values of commercially prepared Florida grapefruit juice ranged from 425 to 746 ppm during one season.

All three of these reports refer to data from a single season and therefore have little predictive value for naringin levels in future crops from the same locality. Data for those seasons might not have been typical. Naringin content might be unusually high in certain years, as suggested by the expression "bitter grapefruit years" used by those knowledgeable in the art of grapefruit juice manufacture. Also, the method of juice extraction markedly affects the juice naringin concentration (Dougherty et al., 1977), and the juices in the three reports were not extracted by a standardized procedure.

Hagen et al. (1966) and Tatum et al. (1972) reported that the Davis test does not give a true measure of naringin concentration in grapefruit juice. These authors used sophisticated and time-consuming analytical methods based on thin-layer chromatography and spectral analysis to determine the concentration of the minor flavanones present in grapefruit juice. They independently concluded that the Davis-test method yielded apparent concentrations which were ~2.1 times the true naringin concentration. The authors reported, however, that the Davis method gave apparent naringin concentrations which were reasonably proportional to the true naringin concentration, and thus gave fair indexes of naringin concentration, and did accurately reflect changes in flavanone glycoside concentration. Hendrickson and Kesterson (1957) studied methods of assaying citrus flavanones and derivatives and

Subtropical Products Chemistry Research Unit, Science and Education Administration, U.S. Department of Agriculture, Weslaco, Texas 78596.

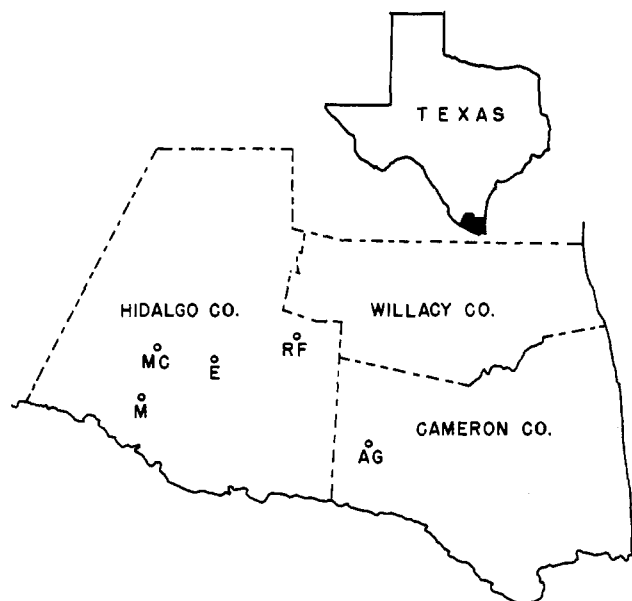


Figure 1. Map of South Texas citrus belt counties. Grove locations are designated as follows: Rio Farms, RF; Edinburg, E; Monte Cristo, MC; Mission, M; Adams Gardens, AG.

similarly conclude that the Davis test is most suited for routine assaying. In spite of its limitations, the Davis test has been used by the citrus industry for 25 years to determine relative flavanone bitterness in grapefruit juice and its limitations are well understood (Horowitz and Gentili, 1959).

The purpose of our research was to assess the variability of juice naringin approximations due to harvest date, crop year, and grove location. To accomplish this requires the use of uniform methods of sampling, extraction, and analysis.

MATERIALS AND METHODS

Eight trees each of Ruby Red grapefruit on Texas sour orange (*Citrus aurantium* L.) rootstock were selected and marked in each of five groves distributed within the citrus belt of the Lower Rio Grande Valley of Texas ($\sim 26^\circ$ north latitude). Their locations are shown in Figure 1. The groves were maintained by practices characteristic of good commercial grove management.

Samples of fruit were collected biweekly from each grove during five successive seasons: 1968–1969, 1969–1970, 1970–1971, 1971–1972, and 1972–1973. Initial sampling dates for each season are respectively Dec 12, Oct 16, Oct 27, Nov 2, and Sept 26. The final collection date was June 1 ± 10 days. Rains caused occasional deviations from the usual 2-week sampling intervals and partially account for variations in the dates of first samplings. Seven fruits were randomly harvested from each of the 8 trees to give a 56-fruit composite sample for each of the 5 groves.

Each sample was washed as in a commercial packing house, i.e., they were soaked in water, sprayed with water while brushed with bristled rollers, and then drained. Juice was extracted with a Model 091B FMC in-line test extractor. A no. 2 or no. 3 cup with a 0.64-mm (0.025 in.) prefinisher strainer screen were employed. The pulp outlets were set with the gauge at 124-kPa (18-psi) air pressure. Juice yield was measured from the weight ratio of juice to whole fruit. After deaeration of the juice, °Brix, total acid, pH, and pulp were determined by the standard methods of the industry (Praschan, 1975). Color was determined with a Gardner color difference meter; an LR-1 standard was used and the *a/b* ratio was calculated (Harding and Fisher, 1945).

Duplicate samples of deaerated juice were analyzed for naringin concentration by the Davis test and the means recorded. Because the juice is measured volumetrically for the Davis method, the naringin value should be expressed as percent weight by volume. Some workers (Tatum et al., 1972) have substituted parts per million (ppm) by approximating the density of juice to be 1 g/mL. We also made this approximation. Separate curves of Davis-test naringin values vs. harvest date were plotted for each crop year.

Starting with the 1969–1970 season, observations were made on the growth status of the trees from which the fruit were harvested. For 1969–1970, 1970–1971, and 1971–1972, these observations were made on harvest dates beginning around Feb 1; in 1972–1973 observations were begun at the early October harvest. Vegetative growth stages were classified as buds dormant, buds swelling, buds pushing, and shoot elongating. Flower growth stages were classified as “green ball”, white showing, elongated blossom, blossom open, and petal drop. Growth stages were subclassified as either general (typical for all trees) or isolated.

RESULTS AND DISCUSSION

Figure 2 shows that the naringin value varied with harvest date within the year, with grove, and with crop year. We conclude that annual climatic variations and/or cultural practices strongly influenced the Davis naringin value and thus probably the bitterness of the grapefruit juices which were extracted by simulated, uniform commercial methods.

The 1971–1972 crop year produced juice with the lowest naringin values, all below 170 ppm and all but the two February samples had less than 150 ppm. Three samples in May and June showed minimum values of 70–75 ppm. In the 1971–1972 crop year naringin values varied little between samples taken from different groves on the same date, the average difference being 30 ppm—considerably less than locational differences noted in the other crop years studied. This uniformity is probably due to the almost ideal growing conditions that prevailed at all groves during that year. The groves did not suffer from adverse weather stresses, which can be accentuated by untimeliness of cultural practices (e.g., irrigation to relieve water stress). The average decrease of naringin value from December to June was 45 ppm.

The 1970–1971 crop year gave the second lowest naringin values (between 135 and 205 ppm). Variation with location was only slightly greater than in 1971–1972. The maximum variation between groves on any one sampling date was 45 ppm. Other than short-term fluctuations, no upward or downward trends in naringin values were noted from December to June. With the exception of two harvest dates, naringin values were consistently the highest for the Edinburg grove. This indicates that location can influence naringin values and thus bitterness.

The 1969–1970 and 1972–1973 crop years were characterized by large differences between groves and widely separated extreme values. Naringin values ranged from 120 to 295 ppm for 1969–1970 and 135 to 285 ppm for 1972–1973. In both years naringin values dropped rapidly for all groves by mid-October when minima usually occurred. The 1969–1970 curves showed no upward or downward trend from December to June and fluctuated sharply. Fluctuations were less in the 1972–1973 crop year and the curves tended to fall during December to June by ~ 40 ppm. From late September to February the Edinburg grove consistently gave the highest naringin values. All groves showed a similar drop of 20–40 ppm 2 weeks after a moderate freeze on Dec 21, 1973.

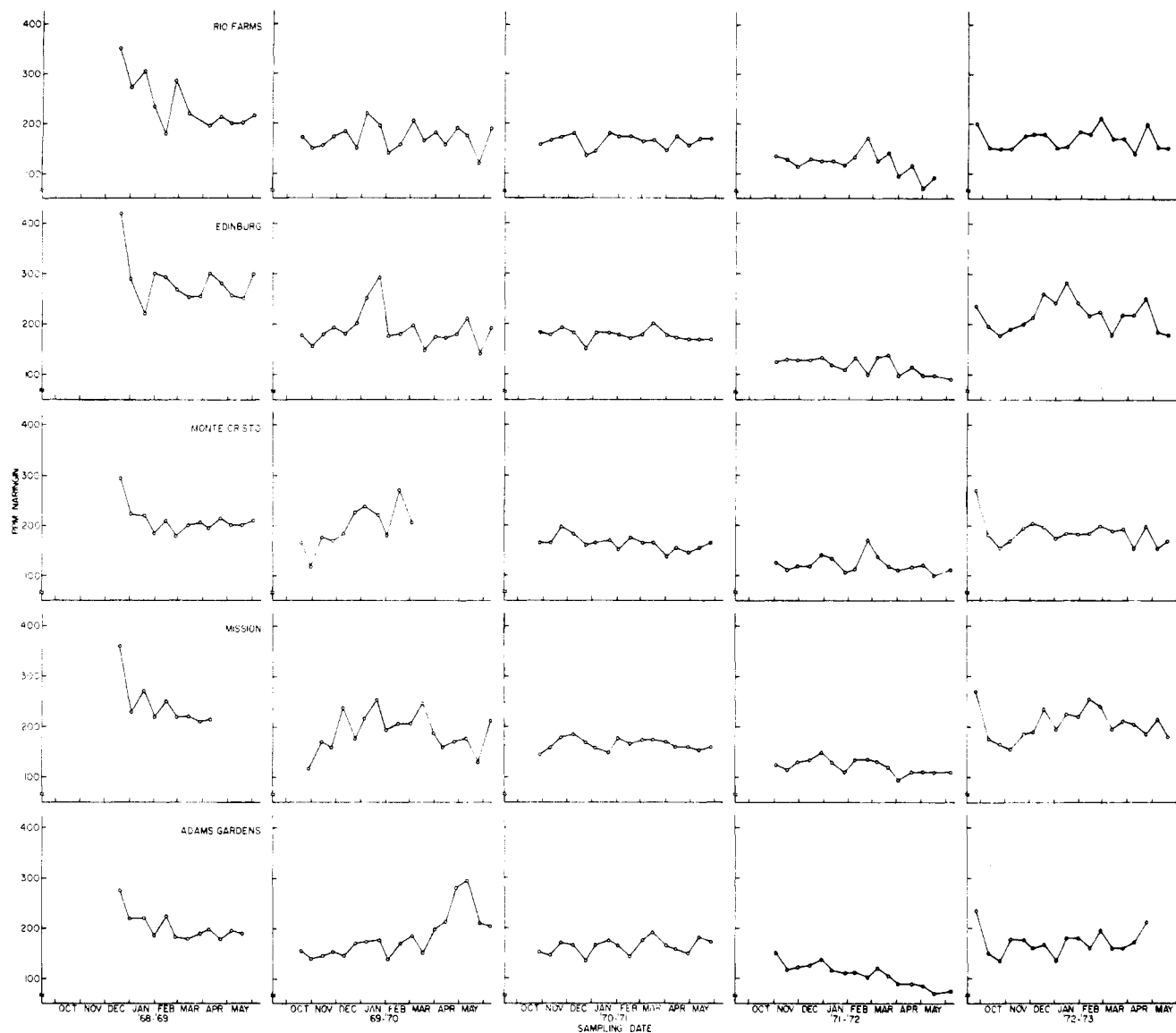


Figure 2. Davis naringin values (ppm) for five groves and five successive seasons.

Naringin values were highest during the 1968–1969 season, ranging from 420 ppm in mid-December (no samples were taken prior to that) down to 180 ppm. There were fairly wide and constant differences between groves. Again values for the Edinburg grove were the highest. On any one sampling date they were 50–80 ppm higher than the average for the other groves, except for one sampling date. The Adams Garden grove generally had the lowest naringin values. Of the five groves, Adams Gardens and Edinburg represented the extremes in growing conditions, but the significance of this difference remains to be determined. The Adams Garden grove has the heavier soil (U.S. Department of Agriculture, 1972) and is in a more humid location that experiences slightly cooler summers and warmer winters than the Edinburg grove (Orton et al., 1967). The 1968–1969 crop year was the driest of the 5 years studied (U.S. Department of Commerce, 1968–1973).

After the rate of fruit enlargement due to rapid water accumulation in juice sacs has moderated by mid-October, no consistent trend in juice naringin concentration can be anticipated; however, the late October values approximate an average level for the season more often than not.

On the basis of the 5 years of data, the Adams Garden grove would be most likely to have the lowest naringin values relative to the other groves for any future crop year. None of the remaining four groves are sufficiently unique

in juice naringin value to be differentiated in juice quality predictions.

Influence of Tree Growth Status. In many of the groves naringin values increased after the spring flush of growth in February, March, or April. The occurrence of these slight increases differed with grove and year and correlated well with the onset of vegetative spring growth.

Independent evidence for the association of vegetative growth with naringin accumulation has been presented by Maier (1969), Fisher (1968), and Albach et al. (1969).

Other Juice Quality Parameters. Unlike the naringin values, the other juice quality parameters examined did not vary significantly between groves or crop years.

Davis naringin values can only be compared for juices identically extracted. Commercial methods of extraction are primarily chosen to maximize juice yield and therefore result in significantly higher naringin values than we report (Albach et al., 1981). Moreover, Lime et al. (1958) have shown that pulp added to enhance color of juice from red-fleshed grapefruit varieties raises Davis-test naringin values and that the increase is proportional to the amount added.

Due to the known limitations of the Davis test as a measure of the bitter flavanone glycosides and its insensitivity to the presence of the bitter limonin, which is known to be an important factor in determining grapefruit

flavor scores (Dougherty and Fisher, 1977), the utility of the Davis test as a predictive measure of grapefruit bitterness is not without question.

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Effect of Potato Virus X on the Mineral Content of Potato Tubers

Subhash Chandra and Nell Irene Mondy*

The effect of infection with potato virus X (PVX) on the mineral content of five potato cultivars was studied. The cultivars, Katahdin and Chippewa, were grown during the first season, and Katahdin, Chippewa, Sebago, Peconic, and Bake King were grown during the second season. Tubers from plants that were infected with PVX were compared for mineral composition with those that were free of the infection. Infection with PVX resulted in a decrease in Ca, Fe, and Cu and an increase in P and Zn as compared with the controls. No significant changes were found in K, Mg, Mn, B, and Al content. Cultivars differed in their response to PVX. However, within cultivars there were consistent trends during each of the 2 years of the study.

Fresh potatoes are a good source of minerals. A 150-g serving of fresh potatoes will furnish 10% of the U.S. Recommended Dietary Allowance for iodine, 8% for copper and magnesium, 6% for phosphorus, and 2% for iron and zinc (True et al., 1978, 1979). Potato virus X (PVX) is an important virus affecting potatoes, and several researchers have reported an appreciable reduction in tuber yield with PVX infection (Emilsson and Gustafsson, 1956; Wright, 1970, 1977; Dowley, 1973). All the older varieties commonly grown in the United States and Canada such as Green Mountain, Irish Cobbler, and Russett Burbank are usually infected with this virus (Smith, 1977). Previous work from our laboratory has indicated that PVX-infected potatoes as compared to PVX-free potatoes are more susceptible to enzymatic discoloration, lower in crude lipid and phospholipids, and higher in phenols (Mondy and Koch, 1978). Differences in ultrastructural characteristics

have also been shown between PVX-free and PVX-infected potatoes (Mondy et al., 1980). However, relatively little is known concerning the changes in mineral composition of potato tubers due to PVX infection.

Kozłowska (1964) reported that PVX infection of potatoes resulted in a higher level of potassium content. Tubers of virus X infected plants contained an average of 18% more potassium than tubers of healthy plants (Slusarek, 1971). Slusarek also found that infection with virus X increased the potassium content of potatoes in the initial stages of their development, and the level of potassium decreased significantly toward the end of the vegetation period. Phosphorus is one of the chief components of nucleic acid which forms the framework of the virus particle, and phosphorus has been shown to be absorbed more rapidly by diseased plants (Kozłowska, 1963). Panjar (1960) reported higher amounts of phosphorus in PVX-infected potato sprouts. Accumulation of phosphorus in PVX-infected tomato fruit has also been reported (Singh and Mall, 1973). Wynd (1943) reported that virus diseases decreased the calcium content of plant tissues. Bergman

* Division of Nutritional Sciences and Institute of Food Science, Cornell University, Ithaca, New York 14853.