Seasonal changes in nutrient concentrations of navel orange fruit

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Received 21 June 1999; accepted 22 July 1999

Abstract

Concentrations of the macronutrients (K, Ca, Mg, Na, P and S) and micronutrients (Fe, B, Zn, Mn and Cu) in whole fruit, pulp, rind and albedo of fruit from young and mature 'Bellamy' navel orange trees were measured at fortnightly intervals during fruit development. The general order of abundance of the macronutrients were K > Ca > Mg = P > S > Na and the abundance of the micronutrients followed the order B > Fe > Mn = Zn > Cu in whole fruit. The concentrations of most elements in whole fruits and fruit parts decreased during fruit development. In whole fruit K concentrations decreased throughout fruit development whereas the Ca concentration increased during stage I and early stage II, and then progressively decreased. In the pulp these trends were reversed. The K/Ca ratio of whole fruit and rind initially decreased during stage I and then remained constant or increased slightly. In contrast, the K/Ca ratio of the pulp increased linearly during most of fruit development. Seasonal trends in whole fruit, pulp and rind K/Ca and Mg/Ca ratios were similar. Fruit from mature trees, with a history of frequent and severe outbreaks of the rind disorder albedo breakdown (crease) had higher albedo K/Ca and Mg/Ca ratios during stage I of fruit development, compared to fruit from young trees. Concentrations of Fe and B in whole fruits and fruit parts changed little during fruit development, contrary to other micronutrients, such as Mn, Zn and Cu, whose concentrations declined in whole fruits and most fruit parts over the same period. Indirect evidence for high B mobility in phloem of citrus is presented. © 2000 Elsevier Science B.V. All rights reserved.

Keywords: Navel orange; Fruits; Pulp; Rind; Albedo; Seasonal changes; Nutrient content; Nutrient ratios

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PII: S0304-4238(99)00093-X
1. Introduction

The mineral content of plant parts, in particular leaves, is used to identify nutrient deficiencies, excesses or imbalances within a crop. The nutritional status of citrus trees, particularly N, P and K, influences citrus fruit quality as well as crop yield (Moss, 1972, and references therein). Seasonal change in the mineral nutrient status of citrus leaves is well established (Jones and Parker, 1951; Labanauskas et al., 1959) and the effect of factors such as rootstock on leaf mineral nutrient composition have been reported in detail (Taylor and Dimsey, 1993). The macro- and micronutrient content of citrus fruits has been reported for mature fruit (Erickson, 1968), and the effect of factors, such as rootstock (Hass, 1948) and position of the fruit in the canopy have been assessed (Koo and Sites, 1956). Abdalla et al. (1986a,b) measured changes in floral N, P, K and micronutrients concentrations in the ovary from flower initiation to fruit initials. Descriptions of the seasonal changes in the nutrient composition of whole fruit from fruit set to maturity are few. Zidan and Wallace (1954) and Garcia-Martinez et al. (1973) measured changes in N, P, K, Ca and Mg concentrations of ‘Washington’ navel and Valencia orange fruits at monthly/bimonthly intervals during fruit development. However, these studies did not include measurement of micronutrients or changes in nutrient concentrations in the component parts of the fruit.

This paper presents some results from a study of fruit growth and nutrient accumulation in Bellamy navel orange fruit. In this study orange fruit were collected at fortnightly intervals from two adjacent sites planted with young and mature Bellamy navel orange trees. The young and mature trees differed in the incidence of the rind disorder albedo breakdown (crease). Seasonal changes in the concentrations of K, Ca, Mg, Na, P, S, Fe, Mn, B, Cu and Zn in whole fruit and the component parts, namely pulp, rind and albedo were measured. The results are discussed in relation to seasonal changes in nutrient concentrations of the structural parts of the fruit, and to their relationship with the incidence of albedo breakdown.

2. Materials and methods

2.1. Experimental design

The experiment was conducted on trees of Bellamy navel orange (a nucellar selection of Washington navel orange) grafted on trifoliolate orange rootstock (Poncirus trifoliata (L) Raf.). The plantings of Bellamy navel orange trees on one site were 4 years old and the plantings on the second site were 29 years old. The sites were located on adjacent blocks on a sandy loam soil, and the trees received
the same fertiliser (N fertigation and P banding) program and irrigation scheduling with adjustment for tree size. The blocks had the same row orientation. The most likely difference between the two sites would have been tree microclimate because of differences in tree size (3–5 m vs. ca. 1.5 m). Fruits were sampled each fortnight from 9 November 1994, to 20 June 1995, a sampling period of 8.5 months. Twelve fruits (one per replicate) were collected at each harvest from young and mature trees, except at the beginning of the season when up to eight fruits were bulked together to form one replicate.

Fruit development was divided into stages I, II, III (Bain, 1958). Stage I refers to the period of growth involving mainly cell division, stage II refers to the period of growth by cell extension and stage III is the period of fruit maturation following colour break. The trees flowered in mid-to-late October. In 1994 the transition from stage I to stage II occurred around 14th December (ca. day 35 from 9th November; ca. 55 days from full bloom) (Storey and Treeby, 1999).

2.2. Fruit analyses

In the field, the unshaded side of each fruit was marked with a felt tipped pen. All subsampling of fruit was designed to produce samples with equal amounts of tissue from the shaded and unshaded sides of fruit because preliminary work showed an effect of fruit side on the concentration of nutrients such as Ca. Fruits were lightly scrubbed with a paintbrush in tap water to remove surface contaminants, then washed in consecutive volumes of deionised water, containing a few drops of domestic detergent and acidified with a few millilitres of concentrated HNO₃, and finally rinsed in deionised water. The plant material was blotted dry with tissue paper. A single fruit was cut into two halves along the equatorial plane. One half or a representative subsample of the orange half was transferred to a pre-weighed glass digestion tube (whole fruit sample) and the tube, together with sample, was reweighed. All remaining samples were obtained from the second half of the fruit. Pulp, rind and albedo were carefully excised and subsamples (ca.1 g fresh weight) of all the three fruit parts were quickly transferred to pre-weighed digestion tubes and reweighed. Tubes were dried at 60°C for several days and reweighed to obtain the sample dry weight (200–400 mg).

The dried plant samples were wet digested in 5 ml 35 N HNO₃. The acid was added to the tubes and the tubes were allowed to stand overnight at room temperature before the samples were heated on an aluminum block digester (Windrift Instruments, Perth, Australia). The tubes were cooled, made up to 20 ml with deionised H₂O and the mixed solutions were allowed to settle overnight. Mineral elements were measured by inductively coupled plasma-atomic emission spectrometry (Spectroflame ICP, Spectro Analytical Instruments, Kleve, Germany). Samples and standard solutions were matrix-matched with respect to the
acid and its concentration. The elements K, Na, P, S, and Ca were measured with a simultaneous spectrometer and the remaining elements were measured with a sequential spectrometer. The elements Fe, Mn, B, Zn, S, and P were background corrected but corrections for peak interference and matrix effects were not necessary for any element.

Macronutrient and micronutrient concentrations in this study are expressed in units of millimoles or micromoles (g dry weight) of tissue, respectively, rather than the conventional nutritional units of % and mg kg$^{-1}$. The authors believe that such units are more appropriate in understanding aspects of plant nutrition addressed in this paper, e.g., relative concentrations of elements such as Fe and B. The Na data for fruit from young trees harvested on day 112 were clearly erroneous and are not presented.

2.3. Statistical analyses

Statistical analyses, including ANOVA, were carried out with Genstat 5, Release 3.1 (Rothamsted Experimental Station, UK), and Sigmaplot v4.01 (Jandel Scientific, USA) was used for producing the graphics. Data for young and mature trees were generally similar and, in most cases, combined and presented for whole fruit, pulp and rind. However, relevant differences between young and mature trees are described. Selected data for albedo tissue are presented.

3. Results

3.1. Macronutrients

Harvest date was a significant factor ($P < 0.05$) for all macronutrients in whole fruit, pulp and rind (Fig. 1), although in the case of Na there was clearly no major trend of increasing or decreasing concentration throughout the season. At fruit maturity, the relative order of concentration of the macronutrients in whole fruit was $K > Ca > Mg = P > S > Na$. Concentrations of K and P in whole fruit decreased from a maximum just after fruit set to a minimum at fruit maturity. The relationships between time and whole fruit K concentration, and time and whole fruit P concentration fitted linear curves with significant ($P < 0.001$) correlation coefficients ($R^2$) of 0.91 and 0.88, respectively (data not shown). Both nutrients showed a small rise in concentration toward the end of sampling. In contrast to K and P, the elements Ca, Mg and S increased in concentration on a whole fruit basis during stage I of fruit development. There was a four-fold increase in Ca during this period. During stages II and III the concentrations of all macronutrients, with the exception of Na, decreased.
Seasonal changes in the concentrations of Mg and S were similar in whole fruit, rind and pulp (Fig. 1). Sodium concentrations were largely the same in whole fruit, pulp and rind although during stage III Na concentrations were higher in the rind than in the pulp. Potassium and P concentrations increased in the pulp during stage I then decreased during stages II and III, whereas K and P concentrations in the rind decreased throughout the season. In contrast, Ca initially increased in the rind and then decreased in concentration after day 40, but the Ca concentration of the pulp changed little during stage I and then decreased more than four-fold by the final harvest.

Fig. 1. Seasonal changes in macronutrient concentrations of whole fruit (●), pulp (□) and rind (Δ) during fruit development. Data for young and mature trees combined. Vertical bars indicate LSD at $P = 0.05$ ($n = 24$) — F, whole fruit; P, pulp; R, rind.

Seasonal changes in the concentrations of Mg and S were similar in whole fruit, rind and pulp (Fig. 1). Sodium concentrations were largely the same in whole fruit, pulp and rind although during stage III Na concentrations were higher in the rind than in the pulp. Potassium and P concentrations increased in the pulp during stage I then decreased during stages II and III, whereas K and P concentrations in the rind decreased throughout the season. In contrast, Ca initially increased in the rind and then decreased in concentration after day 40, but the Ca concentration of the pulp changed little during stage I and then decreased more than four-fold by the final harvest.
The tree age by harvest date interaction was significant ($P < 0.05$) for seasonal changes in the Ca concentration of whole fruit and component parts (Fig. 2). During stage I the Ca concentration was higher in whole fruit and component parts of fruits that were harvested from young trees compared to fruits from mature trees. There was little difference between young and mature trees in the Ca concentration of whole fruit and pulp during stages II and III.

3.2. Micronutrients

Harvest date was a significant factor ($P < 0.05$) for all micronutrients of whole fruit, pulp and rind (Fig. 3). The general order of concentration of the micronutrients in mature fruit was $B > Fe > Mn = Zn > Cu$. In whole fruit, pulp and rind, Fe and B concentrations were relatively constant during fruit development in contrast to the decrease in concentrations of Mn, Zn and Cu. The concentrations of Mn, Zn and Cu decreased progressively with time in whole fruit, pulp and rind with the exception of Cu in the pulp that increased during stage I. Compared to the macronutrients (e.g. K), seasonal trends were, generally, the same in whole fruit, pulp and rind for all micronutrients. During stages II and
III the concentrations of B and Mn were greater in the rind than in the pulp, but Cu concentrations were higher in the pulp than the rind. Zinc concentrations were the same in whole fruit, pulp and rind post-stage I and decreased in concentration at the same rate, but remained relatively constant in the albedo (data not shown).

3.3. Macronutrient ratios

Seasonal changes in the K/Ca ratio of the pulp showed that the relative concentration of K compared to Ca increased throughout fruit development.

Fig. 3. Seasonal changes in micronutrient concentrations of whole fruit (●), pulp (□) and rind (△) during fruit development. Data for young and mature trees combined. Vertical bars indicate LSD at $P = 0.05$ ($n = 24$) — F, whole fruit; P, pulp; R, rind.
(Fig. 4). Over most of the season the increase in the pulp K/Ca ratio was linear. The K/Ca ratio decreased sharply and then either increased a little or remained constant in whole fruit and rind, respectively, from the beginning of stage II to final harvest. The seasonal changes in the Mg/Ca ratio were similar to the changes in the K/Ca ratio during fruit development for whole fruit, pulp and rind. However, the Mg/Ca ratios were much lower than the K/Ca ratios. The K/Mg and K/P ratios of whole fruit changed much less than the K/Ca and Mg/Ca ratios during stage I and, thereafter, the trend was slowly upwards. The K/Mg and K/P ratio of the pulp increased sharply during stage I and then remained relatively constant, whereas the seasonal increase in the rind K/P ratio showed no major discontinuities in contrast to the K/P ratio of the albedo (Fig. 4, inset). Even though the K/Ca and Mg/Ca albedo ratios decreased during stage I and early stage II, the ratios were substantially higher in fruit from mature trees than fruit from young trees (Fig. 5).

During stages II and III the Ca/Mn ratio remained constant but the Ca/B ratio decreased progressively showing that the B concentration of the fruit, in contrast to that of Mn, increased relative to the Ca concentration (Fig. 6).
3.4. Albedo vs. rind nutrient concentrations

From the end of stage I to the end of June the most abundant nutrient in the albedo was Ca (data not shown). During fruit development K and P concentrations were mostly lower in the albedo than the rind (Fig. 7). Calcium levels were about the same in the albedo and the rind. Rind and albedo Na concentrations were not correlated. The relationship between rind and albedo macronutrient concentration was essentially 1 : 1 for Ca, Mg and S. Variation about the regression line for a nutrient such as Ca was attributed to decreasing...
Fig. 7. Correlations between mean \((n = 24)\) rind and albedo nutrient concentrations. Asterisks indicate significance of \(R^2\).
regression slopes within each harvest, particularly during stage I and early stage II of fruit development (data not shown).

The correlations between rind and albedo micronutrient concentrations were relatively linear (Fig. 7) and in the case of Fe and Cu the ratio was essentially 1 : 1. The correlation between rind and albedo Zn concentrations was clearly curvilinear.

4. Discussion

4.1. Macronutrients

The pattern of decline of macronutrient concentrations in whole Bellamy navel orange fruits during fruit development is similar to trends found in other fruits, e.g., apple (Rogers and Batjer, 1954). A marked transition from increasing concentration to decreasing concentration was evident for Ca and has been observed in other fruits, e.g., avocado (Witney et al., 1990). Contrary to our results, Garica-Martinez (1973) reported an initial decrease in Ca concentration followed by an increase in Ca concentration of Washington navel and Valencia Late orange fruit four months after the first harvest date. Zidan and Wallace (1954) also reported a small increase in Ca concentration of Washington navel and Valencia orange fruit 2–3 months after fruit set. However, our findings concur with those of other researchers who have linked the increase in Ca concentration to the early phase of cell division as occurs in other fruits, for example apples (Wilkinson and Perring, 1964). Furthermore, the marked inflexion in the Ca curve for Bellamy navel orange fruit, which coincides with the beginning of stage II, suggests that the process of Ca import into fruits may not be simply passive and unregulated. The same physiological trigger that ends cell division and starts cell extension could also modulate Ca accumulation in the fruit.

Nutrients are not always distributed uniformly between structural parts of the fruit (Clark and Smith, 1990). In Bellamy navel orange fruit S and Mg concentrations were similar in whole fruit, pulp and rind but K and P concentrations were higher in the pulp than the rind and increased in concentration markedly during stage I. Sulphur, Mg, K and P, therefore, show different distribution patterns within the fruit even though all four macronutrients are characterised as being relatively phloem-mobile (Marschner, 1995). The high concentration of Ca in the rind and the low concentration in the pulp during most of fruit development can be attributed to the low mobility of Ca in phloem (Marschner, 1995). The pulp is hydraulically isolated from the vasculature of the outer parts of segments and rind (Koch and Avigne, 1990); therefore it is not a site of high water flows derived from xylem sap, as are leaves, and to a lesser extent, the rind of fruits.
4.2. Micronutrients

The overall seasonal trends in declining Mn, Zn and Cu concentrations are typical of fruits other than oranges (Clark et al., 1989). Manganese is a phloem-immobile micronutrient (Marschner, 1995), which probably explains the eight-fold decrease in Mn concentration in the pulp. Iron concentrations remained relatively constant during fruit development, whereas Zn and Cu concentrations decreased substantially over the same period, yet all three micronutrients are considered to have intermediate phloem mobility (Marschner, 1995). Defining nutrients as phloem-mobile, -immobile or of intermediate phloem mobility may explain some trends we observed in Bellamy navel orange fruits, e.g. Fe vs. Mn patterns of accumulation, but not all seasonal trends in micronutrient concentration can be explained by such simplistic concepts. Furthermore, the term ‘intermediate’ may include nutrients covering a broad range of phloem mobilities, e.g. Fe vs. Zn or Cu.

Boron was the most abundant micronutrient in whole fruit, pulp and rind of Bellamy navel orange fruit (Fig. 3) and is present at concentrations at least three-fold higher than those of Fe. There is a large range in B mobility between species which has been attributed to polyol production in source leaves and the formation of B-polyol complexes in the phloem (Brown and Shelp, 1997). The pulp, a non-transpiring sink, has a high concentration of B, which changes little during fruit development. Boron concentrations in fruits, as reported in this paper, are only a little lower than those in citrus leaves (Taylor and Dimsey, 1993), and similar to B concentrations in leaves and fruit tissues of other fruits such as those of apple known to have significant B mobility in the phloem (Brown and Shelp, 1997, and references therein). This uniformity in B concentration between leaves and sinks, such as fruits, is presented as evidence of significant movement of B within a plant via phloem (Brown and Shelp, 1997). Hu et al. (1997) demonstrated that B forms complexes with the ligands, mannitol, sorbitol and fructose. The major carbohydrates in citrus juice are glucose, fructose and sucrose, but it is also a rich source of the sugar alcohol, myo-inositol (White, 1990). Power and Woods (1997) speculated that more B complexing ligands in plants will undoubtedly be found in the future, and thus myo-inositol may prove to be a ligand involved in B transport in the phloem of citrus.

Some indirect evidence for B movement from leaves to fruit via the phloem was given by the seasonal changes in the Ca/Mn and Ca/B ratios of fruits (Fig. 6). Calcium and Mn are both phloem-immobile nutrients (Marschner, 1995). Thus, during stages II and III of fruit development the ratio of Ca–Mn remained constant but the concentration of B increased relative to both Ca and Mn over the same period. These results suggest that B and Mn move into fruits by different pathways, i.e. phloem vs. xylem, respectively. The cause of the increase in the Ca/Mn and Ca/B ratios during stage I remains unresolved but could be explained by
mobilisation of Ca from a limited pool stored in the wood of fruit trees (Ferguson, 1980).

4.3. Nutrient ratios

The seasonal changes in the K/Ca, K/Mg, Mg/Ca and K/P ratios of whole fruit and fruit parts can be partly explained by high K, P and Mg mobility in the phloem and by Ca delivery primarily in the xylem. Changes in the macronutrient ratios of the fruit during stage I mostly reflect changes occurring in the rind because the rind represents 80–90% of the fruit biomass during the period starting a few weeks after fruit set to the end of stage I (Storey and Treeby, 1999).

The moderately high concentration of Ca relative to K in mature fruit reported in this paper contrasts to many other fruits, such as peach (Batjer and Westwood, 1958) and apples (Quinlan, 1969), that have high K/Ca ratios. Contrasting seasonal changes in the K/Ca ratios of the pulp and rind probably reflect differences in pathways of water movement in orange fruits. The non-transpiring, hydraulically isolated pulp accumulated increasing levels of K relative to Ca over the season, whereas the K/Ca ratio of the transpiring rind decreased dramatically during stage I. The K/Ca ratio of the rind remained constant during stages II and III of fruit development when the transpiration rate of the fruit (normalised to fruit mass) may be constant (e.g. grapefruit — Huang et al., 1992), and epicuticular wax covers the surface of fruits (Storey and Treeby, 1994).

Seasonal changes in the Mg/Ca ratio of whole fruit, rind and pulp followed the same patterns of change as the K/Ca ratio (Fig. 4) as expected on the basis of their relative mobilities in the phloem and xylem. The marked increase in the K/P and the K/Mg ratios of the pulp during stage I of fruit development suggest that phloem-mobile nutrients may not always be accumulated at the same rate. The processes that differentially modulate phloem transport of nutrients into fruit/fruit parts are not clearly understood.

4.4. Albedo breakdown

Fruit disorders have often been associated with low Ca and high K/Ca ratios (e.g. bitter pit of apples — Faust and Shear, 1968). Orange rind showing symptoms of albedo breakdown (crease) is frequently low in Ca (unpublished data). The two field sites in this investigation were chosen because the adjacent blocks were on the same soil type, received the same fertilizer and water management schedules, but the mature trees had a history of frequent and severe outbreaks of albedo breakdown (Treeby et al., 1995). Both the K/Ca and Mg/Ca ratios were higher in albedo tissue of fruit from mature trees compared to young trees during stage I and early stage II (Fig. 5). Similarly, Tomala and Trzak (1994) found K/Ca and Mg/Ca ratios were twice as high in ‘Alexander Lucas’ pears.
affected with cork spot as those unaffected by the disorder. The low Ca status relative to K and Mg occurred in orange fruit from mature trees during the principal period of cell division and the early phase of cell extension when cell walls of albedo cells undergo substantial thickening (Storey and Treeby, 1999).

5. Summary

Information on the seasonal changes in nutrient concentrations of whole fruit and structural parts of the fruit is useful in understanding the nutrient requirements of the tree and the effect on yield and fruit quality. Stage I is a period of fruit development in which there are rapid and extensive changes in nutrient concentration. Differences between nutrients, e.g. K vs. Ca, and between structural parts, e.g. rind vs. pulp, may be determined by the relative contributions of xylem and phloem flows to fruit composition. Clearly, there are other factors that differentially modulate the transport of phloem-mobile nutrients such as K and P during stage I, but they are not fully understood. Boron concentrations are high in navel orange fruit and may be relatively phloem-mobile in citrus.

Acknowledgements

The financial support of the Horticultural Research and Development Corporation is gratefully acknowledged. The authors are grateful to Dr. R.R. Walker for his comments. Mses. Sue Maffei and Rachel Hall are thanked for technical assistance. The co-operation of the staff at the NSW Agriculture’s Research and Advisory Station, Dareton, is gratefully acknowledged.

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