

THE EFFECT OF CALCIUM SALTS ON THE COMPONENTS CAUSING BROWNING OF AVOCADO FRUIT

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OPSOMMING

Fuerte avokados was gedoop in 0.18 M oplossings van calciumchloried ($\text{CaCl}_2 \cdot 6\text{H}_2\text{O}$) of calciumfosfaat (CaHPO_4) of calciumarsenaat ($\text{Ca}_2(\text{AsO}_4)_2$) of calciumnitraat ($\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$) om vas te stel of hierdie behandelings enige invloed gehad het op die verbruinings komponente van die avokados. Dis gevind dat die calcium behandelings wel respirasie en polifenoloksidase, asook die totals fenoliese inhoud, leuco-antosianiene en flavonale onderdruk.

SUMMARY

Fuerte avocados were dipped in 0.18 M solutions of calcium chloride ($\text{CaCl}_2 \cdot 6\text{H}_2\text{O}$) or calcium phosphate (CaHPO_4) or calcium arsenate ($\text{Ca}_2(\text{AsO}_4)_2$) or calcium nitrate ($\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$) to determine whether these treatments had any effect on the browning components in avocados. It was found that calcium treatments suppressed both respiration and Polyphenol oxidation and also reduced the content of total phenolic, leuco-anthocyanins and flavonols.

INTRODUCTION

The role of calcium in plants has been widely studied. Calcium is present in the cell wall (Gielink, Sauer & Ringoet 1966 & Rathore, Bajaj & Wittwer 1972), the cytoplasm, the nucleus, the mitochondrions (Ratloe, Bajaj & Wittwer 1972) and the chloroplasts (Stocking & On gum, 1962, Mix & Marschner, 1974 and Larkum 1968). In a study on the total uptake of ^{45}Ca , Rathore, Bajaj and Wittwer determined that 60% of the ^{45}Ca accumulated in the leaves, 25% in the stem, and 15% in the roots.

Calcium plays a part in the physiology of cell walls, membranes (Ruis 1972), chromosomes, and enzymes (Jones & Lunt 1967 & Tingwa & Young 1974 & Wills & Tirmazi 1979). In the cell walls calcium is incorporated into protopectin as calcium pectate and may fulfill a structural role in the middle lamella and the rest of the cell wall (Setterfield & Bayley 1961).

An increasing number of fruit disorders have been found to show a significant relationship with the calcium contents of the fruit or with the applications of calcium or both. Bitter-pit, lenticel spot and cork spot in apples, blossom-end rot in tomatoes and

peppers are some of the more common physiological disorders in fruit related to the calcium status of the fruit. (Rathore, Bajaj & Wittwer 1972)

Flavonoids generally referred to as tannins (Lewak 1967 & Goldstein & Swain 1959) are associated with browning of plant tissues (Whitehead 1979) and are found in the vacuole (Lewak 1967 & Goldstein & Swain 1959), in the chloroplasts, in the chloroplasts (Goodwin & Mercer 1972) and etioplasts (Swain & Hillis 1959) or in specialized organelles.

The browning is due to structural changes in the phenolic compounds caused by polyphenol oxidation (Pefferi, Cuttrera 1974 & Padfield 1979). The oxidations of phenolic compounds change rapidly during plant growth (Tolbert 1973) and in higher plants it occurs in the cell or the cell wall (Coombs, Baldry, Bucke and Lang 1974). The Polyphenol oxidase responsible for this oxidation is present in the plastid thylakoids (Engelbrecht 1982, Mayer, Harel 1979 & Tolbert 1973) the mitochondrions (Shomer, Ben Shalom, Harel & Mayer 1979), micro bodies (Sharon, Kahn 1979), peroxisomes and microsomes (Ruis 1972 & Flurkey, & Jen 1978).

The enzymatic browning of fruit is caused by the catalytic oxidation of endogenous O-dioxy phenols, followed by polymerization or condensation (Prabha & Patwardhan 1980).

The browning potential of avocado, banana, peach and mango fruit is related to the total polyphenol oxidase activity and the phenol content of the fruit (Kahn 1975, Kahn 1976, Kahn 1977 & Golan, Kahn & Sadovski 1977).

This paper reports on studies undertaken to examine the role of calcium in the development of disorders and to see whether it would have any effect on the browning components in avocados. It was thought that dipping avocados in calcium salts might preserve firmness, reduce respiration, polyphenol oxidation and the content of total phenols, leuco-anthocyanins and flavonols.

MATERIALS AND METHODS Dipping of Avocados

Mature Fuerte avocados were selected within a narrow range of mass and firmness and divided into five groups. Group one (control) contained 15 avocados and the other groups 12 each. The method was the same as that used by Wills and Tirmazi (1979) for dipping tomatoes in calcium and other mineral solutions. Each fruit in the control group was dipped in distilled water at normal pressure for 30 seconds after which the pressure was increased to 38.2 kPa and maintained for 4.5 minutes. After the pressure was returned to normal, the fruit was left in the solution for a further 5 minutes, then removed to dry. The increase in weight after dipping was determined. The same procedure was used for treating the other four groups of avocados, which were dipped in 0.18 M solutions of calcium chloride ($\text{CaCl}_2 \cdot 6\text{H}_2\text{O}$), calcium phosphate (CaHPO_4), calcium arsenate ($\text{Ca}_3 (\text{AsO}_4)_2$) or calcium nitrate ($\text{Ca} (\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$) respectively. The same calcium concentration was used in all four solutions. When dry, all fruits were kept in cold storage at 7°C for 30 days. Batches of 3 fruits from each group were analyzed on days 1 (control only), 5, 15, 22 and 30 after treatment.

RESPIRATION

The respiration rate of the mesocarp tissue was measured with a Gilson differential respirometer and was expressed as $\text{mm}^3\text{O}^3\text{kg}^{-1}$ of fresh tissue over a 1 h period.

Total phenolic content, leuco-anthocyanins and flavonols. Methods of Swain and Hillis (1959) were used for the quantitative determination of the total phenols, leuco-anthocyanins and flavonols in fractions of each sample. The different substances were isolated from the extract and measured at different absorbances, total phenols at 720 nm, the leuco-anthocyanins at 550 nm and the flavonols at 500 nm. The respective concentrations were calculated and expressed per gramme of avocado mesocarp.

POLYPHENOL OXIDATION

Polyphenol oxidation activity was measured by a method used by Kahn (1977). The samples were extracted in 0.1 M sodium phosphate buffer adjusted to pH 6.5, and the activity of the extract was determined at an absorbance of 410 nm at 25°C. The activity was expressed as absorbance at 410 nm per minute and specific activity per mg protein.

RESULTS

During the 30-day storage period the fruit dipped in calcium arsenate had the lowest mean respiration rate. Progressively higher rates were shown by fruit dipped in calcium chloride, calcium phosphate and calcium nitrate. Respiration rate for the controls was about the same as that for calcium phosphate treated fruit (Fig. 1).

Total phenol content fluctuated greatly during the 30 days of storage (Fig. 2). After day 15 the content increased in all samples and after day 22 it continued to increase to those treated with calcium phosphate and calcium nitrate but decreased by various amounts in the other treatments. It was lowest in those treated with calcium arsenate.

The leuco-anthocyanin content increased during the first 15 - 22 days and then decreased by day 30 to less than the initial amount in all the treated fruit. Again it was lowest in those treated with calcium arsenate (Fig. 3).

Flavonol content, on the other hand, decreased until day 15 and then increased. The highest final concentrations occurred in the controls and in fruits treated with calcium chloride and calcium arsenate (Fig. 4). Polyphenol oxidation activity increased up to day 15, then decreased irregularly to day 30, reaching the lowest level in fruit treated with calcium arsenate (Fig. 5).

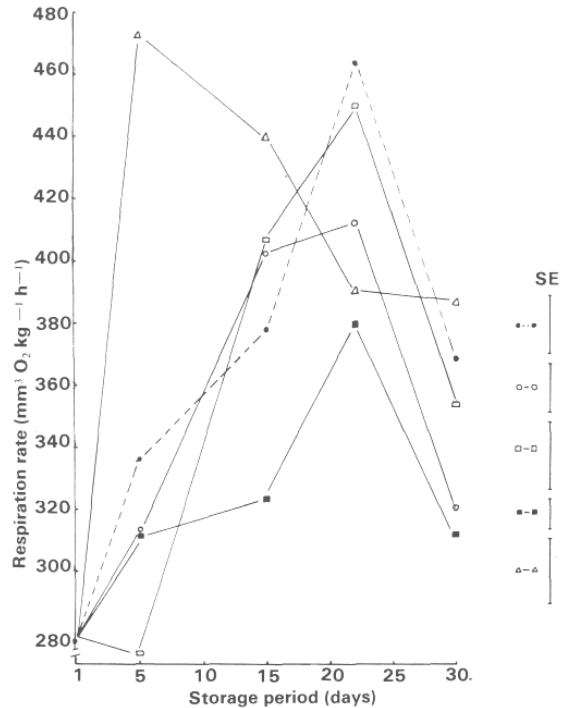


Fig. 1. Changes in the respiration rate of mature avocado fruit during a 30-day storage period at 7°C. ●-●- control; ○-○- calcium chloride; □-□- calcium phosphate; ■-■- calcium arsenate; △-△- calcium nitrate.

DISCUSSION AND CONCLUSIONS

These results show that the total phenols, leuco-anthocyanins and flavonols contained in mature avocado fruit were but little affected by the various treatments applied. Nor were significant differences observed between the effects of different treatments.

Polyphenol oxidation is an important factor in browning of avocado mesocarp (Kahn 1977). The results for polyphenol oxidation obtained in the present study agree with those of Sharon and Kahn (1979) except that greatest activity was measured on day 5 and not on day 4.

The decrease in respiration rate in these treated avocado fruits agrees with the results obtained by Wills and Tirmazi (1979) and Tingwa and Young (1974). The pre-climacteric and climacteric phases were retarded in all the treated fruit except in those treated with calcium nitrate.

The results obtained in this study thus show that these treatments applied do retard and suppress those processes that contribute to browning of avocados. Application of these treatments is thus likely to extend the shelf life of avocado fruit.

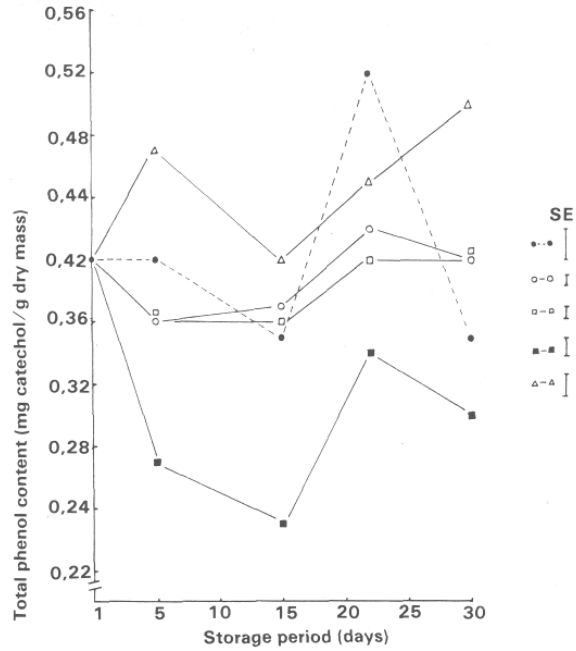


Fig. 2. Changes in the total phenol content of mature avocado fruit during a 30-day storage period at 7°C. ●-●-● control; ○-○-○ calcium chloride; □-□-□ calcium phosphate; ■-■-■ calcium arsenate; △-△-△ calcium nitrate.

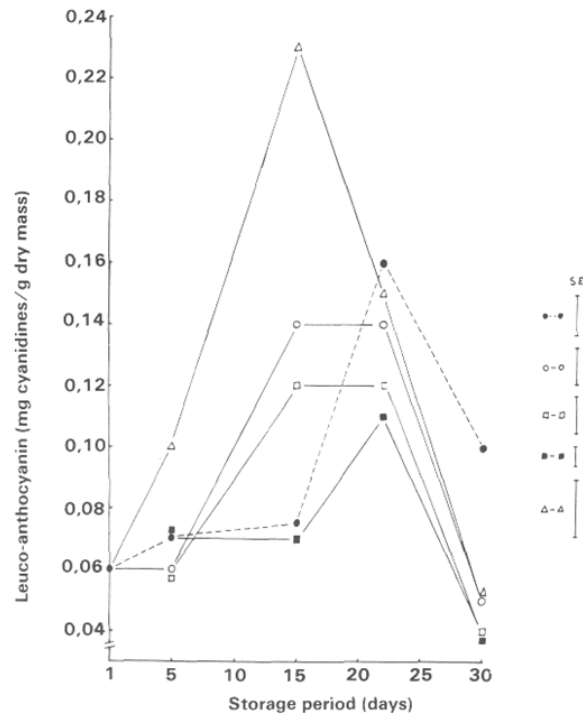


Fig. 3. Changes in the leuco-anthocyanin of mature avocado fruit during a 30-day storage period at 7°C. ●-●-● control; ○-○-○ calcium chloride; □-□-□ calcium phosphate; ■-■-■ calcium arsenate; △-△-△ calcium nitrate.

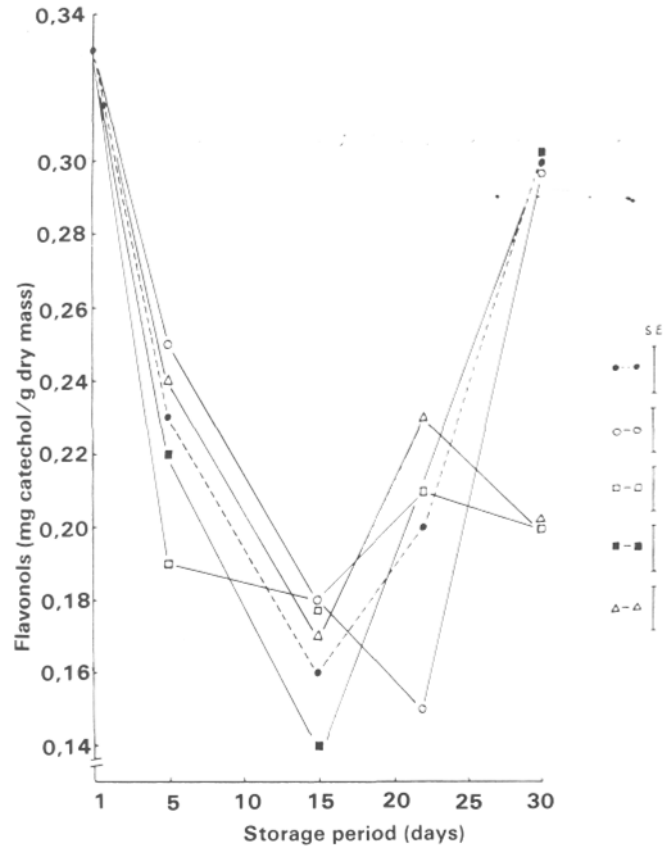


Fig. 4. Changes in the flavonols of mature avocado fruit during a 30-day storage period at 7°C. • - - • control; ○ - - - ○ calcium chloride; □ - - - □ calcium phosphate; ■ - - - ■ calcium arsenate; △ - - - △ calcium nitrate.

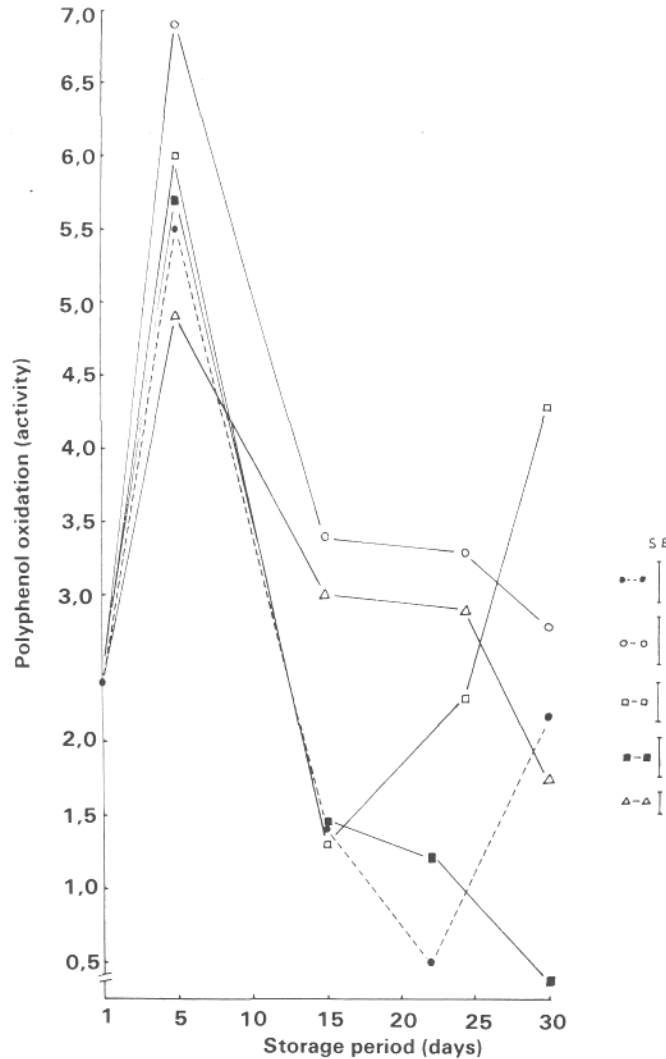


Fig. 5. Changes in the polyphenol oxidation of mature avocado fruit during a 30-day storage period at 7°C. • - - - • control; ○ - - - ○ calcium chloride; □ - - - □ calcium phosphate; ■ - - - ■ calcium arsenate; △ - - - △ calcium nitrate.

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