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Article

The effect of different calcium infusion methods on the texture and consumers' acceptance of ripe jackfruit (*Artocarpus heterophyllus)* pulps.

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Abstract: Ripe jackfruit pulps were infused with calcium solution at two different calcium concentrations, i.e. at 0.5% and 1.0% (w/v), using two methods: immersion and vacuum-assisted infusion. The calcium-treated ripe pulps lost their texture slower than the control (untreated pulps). The cell walls and middle lamella of pulps treated with calcium by vacuum-infusion were still intact after 14 days of storage, while control and immersed samples showed sign of middle lamella dissolution. The texture analysis supported these findings when pulps infused with calcium by vacuum-treatment were firmer than the other samples evaluated. Based on sensory evaluation, the ripe jackfruit pulps, vacuum-infused with 1.0% calcium has a shelf life of 14 days when stored at 8°C.

Keywords: jackfruit; calcium infusion; texture; minimal process; consumers' acceptance

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1. Introduction

Jackfruit (*Artocarpus heterophyllus)* is a popular tropical fruit. It is marketed locally as whole-fruit or minimally processed products whereby the fruitlet are separated from the whole-fruit. The current shelf-life of the jackfruit bulbs is only 1 day at ambient temperature and 3-4 days at 10-15^{o}C (Abdullah, 1988). Latifah (2010) found minimally processed jackfruits pulp stored in modified atmosphere packaging can be kept for up to 10 days when stored at 10^{o}C. Jackfruits destined for export market are usually sent abroad as whole fruit. Transportation through air will give the produce 5-6 days of market quality when they arrived to their destination. However, the air freight is very costly and the edible part of the fruit is only 50-60% of the total cost. Furthermore, the size and shape of the fruits are not consistent, making the design of packaging very difficult (Anon, 2004).

Today's consumers are demanding for produce that are wholesome, nutritional and convenient that still retain their natural characteristics as much as possible. The thick skin and latex on jackfruit makes the minimal processing potential even more attractive (Mardi, 2004). In view of the current trend, concerted effort has been made to develop new methods for minimally processed fruit and vegetable products.

Minimally processed products are defined as those processed by appropriate unit operations such as washing, peeling, slicing and packaging, including chemical treatments which may have a synergistic effect when used in combination (Beuchat, 2000). Traditionally, heat treatment was used to prolong the shelf-life of the products. Although thermal processing can be beneficial in term of food preservation and the development of flavours, it can also instigate damaging effect on the products, especially in term of colour and texture (Martinez-Monzo *et al.*, 2000). Other treatments that had been used in conjunction with minimal processing include irradiation, pulsed electric field and high pressure processing.

Minimally processed jackfruit will boost the fruit's potential both locally and internationally. The unique flavour and crisp texture, coupled with the diverse ways it can be incorporated into a meal, will make minimal processed jackfruit an important export commodity. A method that can preserve the fresh fruit's characteristics; enhanced its shelf stability yet keep cost at a minimum will achieve this purpose.

2. Experimental Section

2.1 Sample

In this study, methods of infusion of calcium infused into the fruit matrix were evaluated with respect to textural attribute, microscopy study and sensory evaluation. Methods of infusion evaluated were immersion in calcium solution for 18 hours and vacuumed-infusion, whereby the jackfruit pulps were immersed in calcium solution and treated with vacuum for 15 minutes. Two different calcium concentrations were studied, i.e. at 0.5 % and 1.0 %. Initially calcium concentrations of 1.5 % and 2.0 % were also considered, but the resulting products tasted bitter. Hence, concentrations of 0.5 % and 1.0 % were used in this experiment.

Ripe jackfruit pulps were separated from the whole fruit and de-seeded. The ripe seedless pulps were either immersed or vacuumed in distilled water containing 0, 0.5 % and 1.0 % calcium chloride dihydrate (Sigma, USA). These samples were drained and placed in polystyrene trays, wrapped with cling wrap (Reynold Clear Plastic Wrap) and kept at 8^{o}C in the refrigerator. An average of 10 - 15 ripe jackfruit pulps were set in each tray. Samples were taken at day 0, 3, 7, 10 and 14 for texture, microscopy examination (light microscope and transmission electron microscope) and sensory evaluation.

2.2 Calcium Infusion

Calcium infusion was done by immersion in calcium solution for 18 hours at 8 - 10^{o}C or vacuum-infusion 15 minutes at room temperature. All treatments were done in calcium chloride solution at 0, 0.5 % and 1.0 % (w/v) concentration.

Calcium chloride dihydrate was used to prepare solutions at concentration of 0.5 % and 1.0 % (w/v) in distilled water. The calcium solution was used at room temperature and calcium chloride was dissolved completely before jackfruit pulps were added to the solution.

2.2.1 Immersion

De-seeded jackfruits were placed in one layer in a deep plastic tray (45cm x 24 cm x 11cm). Calcium chloride solution was poured onto the fruits at a ratio of 1:2 (w/v), making sure all the pulps were completely immersed in the solution. The tray was covered tightly with plastic wrapper and kept in a fridge at 8 – 10^{o}C for 18 hours. The samples were rinsed with distilled water (3 x 400 ml) and drained for another 15 minutes. The jackfruit pulps were arranged onto polystyrene plates, wrapped with cling plastic wrap and kept at 8 - 10^{o}C before further tests.

2.2.2 Vacuum-Infusion

Seedless ripe jackfruit pulps were placed in small plastic trays (16.5 cm x 10.5 cm x 7 cm) in two layers. Calcium chloride solutions were poured into the trays at a ratio of 1:2 (w/v), making sure all the pulps were wholly submerged in the solution.

The plastic tray was put into desiccators and securely covered. Vacuum was applied using a Gast vacuum pump (Model DOA-P136-BN) until no gas bubbles can be observed in the solution. The vacuum pump was switched off. The samples were left in vacuum for 15 minutes.

When the vacuum was released, the pulps were left in solution for another 5 minutes before samples were drained and rinsed with distilled water. The ripe jackfruit pulps were arranged onto polystyrene trays, wrapped with cling plastic wrap and kept at 8 - 10^{o}C before further tests.

2.3 Firmness Analysis

Firmness analysis was done using Texture Analyzer (Stable Micro System version 1.05, UK) equipped with 5 kg load cell and samples were cut with probe HDP/BSK blade set with knife. Test speed was set at 2 mm/s. The probe was programmed to cut through the samples at a distance of 17mm. The data acquisition rate was set at 200pps. Force unit was in Newton (N). Higher force required to cut through the samples correlate to firmer texture of the sample.

Samples of jackfruit with thickness of between 8 to 15 mm were cut into strips of 1 cm width for texture analysis. The strips were placed on the platform and the blade was raised 1 cm above the sample surface. The blade cut through the middle of the strip. A total of at least 10 strips per treatment were averaged to get the data for texture.

2.4 Transmission Electron Microscopy (TEM)

The preparation of samples for Transmission Electron Microscope study was done based on the Technical Manual of TEM obtained from the School of Biological Sciences, USM. The ripe jackfruit samples, which were infused with calcium chloride dihydrate at different concentration either by immersion in solution for 18 hours or subjected to vacuum, were

inspected under the transmission electron microscope to study the structure of cells wall and the middle lamella.

The samples were cut into small cubes (approx. 1 mm x 1 mm x 1 mm) and placed into McDowell-Trump fixative solution in 0.1 M phosphate buffer, pH 7.2. The jackfruits were post-fixed in 1 % Osmium tetraoxide prepared in 0.1 M phosphate buffer, pH 7.2 and washed in distilled water. The samples were dehydrated with acetone and immersed in a mixture of acetone and Spurr's resin to harden. The samples were cut into ultra-thin sectioning, stained with uranyl acetate and lead citrate before they were checked under the transmission electron microscope.

2.5 Sensory Evaluation

Sensory evaluation was carried out on the stored untreated ripe jackfruit pulps and those treated with calcium. The aim of the sensory evaluation was to determine the panellists' acceptability of the ripe jackfruit pulps which were treated with calcium and the changes during storage. A 7 point hedonic scale rating was conducted on days 0, 3, 7, 10 and 14. A scale of "1" indicates "most dislike" and "7" "most like". The attributes evaluated include colour, firmness, texture, taste and overall acceptability. The ripe jackfruit pulps samples were cut into strips of 2 cm in width and thickness between 5 -8 mm was presented to the panellists. Each sample was given a 3-digit random code. Samples were not evaluated when it was found to be unsuitable for consumption. Results were recorded based on the average score given for each attribute for the samples.

2.7 Statistical analysis

Analyses of variance on data in this study were conducted using the SPSS 11.0 Window software. The one way analysis of variance (ANOVA) was done to determine if there was any significant difference and Duncan's test were performed with level of significant of $p \leq 0.05$.

3. Results and Discussion

3.1. T*exture Analysis*

Texture is a very important aspect in consumers' acceptance of fresh produce. In jackfruit, the crisp texture attests to the freshness of the pulps. The untreated ripe jackfruit pulps were stored at 8^{o}C, and samples taken at intervals for texture analysis. The data is shown in Figure 3.1.

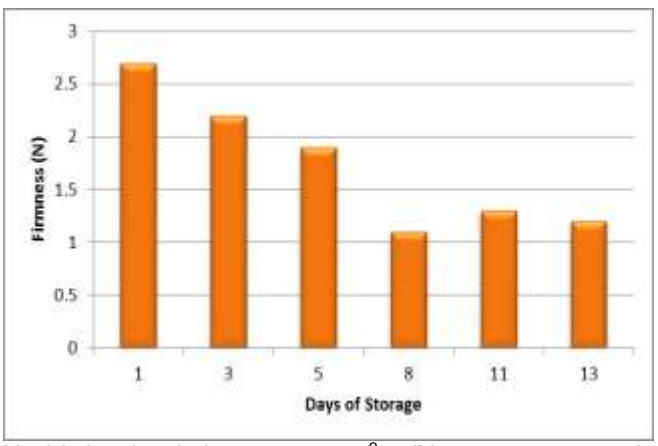


Figure 3.1 **(a)** Texture of control jackfruit pulps during storage at 8^{o}C. (**b**) Data are mean value ± standard deviation of 10 independent samples.

The effects of different calcium infusion methods on the texture of ripe jackfruit pulps were considered. In immersion method where the calcium was introduced into the jackfruit matrix by immersing in the calcium solution, the ripe jackfruit pulps were submerged in calcium solution at $8^{o}C$ for 18 hours. Figure 3.2 summarized the texture data for the ripe pulps immersed in calcium solution at two different concentrations during storage.

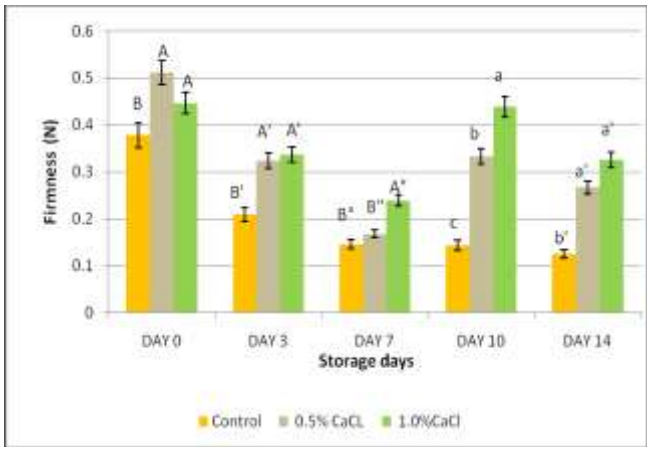


Figure 3.2 **(a)** Texture of stored jackfruit pulps after immersion for 18 hours in different concentrations of calcium. (**b**) Superscript with the same alphabet signifies no significant difference for treatment during same storage day ($p \leq 0.05$). (**c**) Data are mean value ± standard deviation of 10 independent samples.

The control samples were ripe jackfruit pulps immersed in distilled water for 18 hours. During immersion, the immersion liquid diffused into the jackfruit matrix. The diffusion of immersion liquid into the ripe jackfruit pulps matrix caused an increase in the turgor pressure. The increased turgidity and the effect of Ca^{2+} in the ripe jackfruit pulps were correlated by the apparent firmness at day 0. The concentrations of calcium appear not to affect the firmness of the ripe jackfruit pulps significantly.

However, the firmness of the immersed ripe jackfruit pulps decreased at day 7 of storage, followed by increased texture data recorded on day 10 and 14. The increase in texture data observed on day 10 and 14 were not due to increase in firmness of the pulps but rather due to stringiness of the pulps. On both days (day 10 and 14), the blade was not able to execute a clean cut on the pulps. The stringiness brought about by the disintegration of the firmness caused the pulps to be fibrous. Therefore, the infusion of calcium into the ripe jackfruit pulps matrix through immersion failed to maintain the firmness of the pulps during storage. The immersed ripe jackfruit lost its firmness by day 7.

Lamikanra and Watson (2004) found at low temperature penetration of calcium into fruit cells were improved as a result of inward temperature-induced pressure gradient as opposed to outwardly-directed movement at high temperature. Lower temperature also improved covalent intermolecular interactions as a result of the intermolecular proximity and reduced mobility of the molecules (Lamikanra and Watson, 2004).

In vacuum-treatment, the ripe jackfruit pulps were vacuum-infused in calcium solutions for 15 minutes. Initially, vacuum infusion at 5 minutes was also considered. This was because though longer vacuuming time may achieve better calcium infusion, a shorter vacuum time means quicker processing. This translates to lower processing cost. Preliminary data for texture analysis of ripe jackfruit pulps treated for 5 minutes and 15 minutes of vacuum did not indicate any significant differences at day 0. However, after 3 days, the texture of the ripe jackfruit pulps treated to 15 minutes vacuum was much firmer than those subjected to only 5 minutes of

vacuum (Data not shown). Furthermore, according to Collins and Wiley (1967), vacuum treatment for 15 minutes was necessary to remove gases from the tissues. If these gases were not removed, they might interfere with calcium penetration and may expand during processing, causing the tissues to rupture, resulting in soft and mushy texture. Hence, the vacuum infusion of ripe jackfruit pulps in this study was set at 15 minutes as this gave better result.

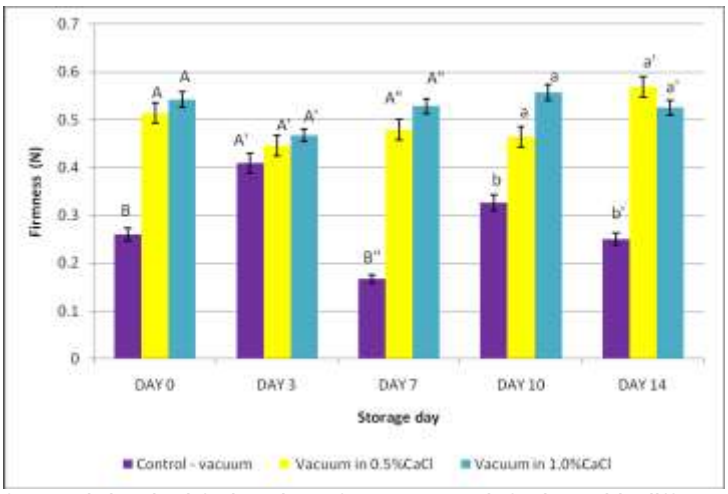


Figure 3.3: (a) Texture of stored ripe jackfruit pulps after vacuum-infusion with different calcium concentrations. **(b)** Data are mean value ± standard deviation of 10 independent samples. **(c)** Superscript with the same alphabet signifies no significant difference between treatments on the same day of storage ($p \leq 0.05$).

Figure 3.3 showed that jackfruits vacuum-infused with calcium displayed better texture retention than control samples. Unlike control ripe jackfruit pulps, jackfruits infused with calcium by vacuum treatment did not lose its texture after 14 days of storage. Similar to immersed jackfruit pulps, the concentration of calcium solution did not have a significant effect on the texture of the ripe jackfruit pulps.

The presence of calcium clearly gave an advantage to the texture of the ripe jackfruit pulps in this experiment. The presence of calcium in the jackfruit matrix reacted with the membranes and cell walls protein increasing its mechanical strength. The concentration of calcium at 0.5% and 1.0% in the infusion solution did not affect the firmness appreciably.

In ripe jackfruit pulps vacuum-infused in calcium solution, the texture was retained even after 14 days. In both methods, the calcium was relocated into the porous structures of the jackfruit pulps matrix; through diffusion of solute in immersion and intercellular-spaces gases displacement in vacuum-infusion.

The experiment proved that although calcium improved the preservation of the texture of the ripe jackfruit pulps, the intactness of the cell walls structures are important as it is the integral part of the outcome. This confirmed that the interaction of calcium ions were with the cell walls of the jackfruit pulps. In the calcium-treated ripe jackfruit pulps, the cell wall structures were still undamaged as infusions were done at low and room temperatures. The thin nature of the pulps enables the calcium ions to permeate through the fruits. Thus the texture is enhanced through the interaction of the calcium and the pectin in the cell walls. According to Lefever *et al.* (2004), the cell wall is responsible for the texture of the tissue and structure integrity of processed fruits.

3.2. *Microscopy Analysis*

The characteristics of the fruits' structures influence the final impregnation level. In densely packed tissue, the exchange of intercellular fluid with external fluid mainly takes place in the central vascular tissue, since no intercellular volume is available in the parenchyma (Gras *et al.,* 2003). Peleg *et al.* (1985) found that samples vacuum-treated with isotonic solution exhibited the response of a viscoelastic sponge, which disappears when calcium is added into the isotonic

solution. This suggests the formation of bonds in the middle lamella and cell walls promoted by calcium.

Microscopic analyses done on the ripe jackfruit pulps cells were carried out with the light microscope and transmission electron microscope. These experiments will give better understanding in terms of the cellular aspects of the jackfruit pulps and how these characteristics influence the textural quality of the minimally processed ripe jackfruit pulps.

3.1.1. Light Microscopy

In light microscopy, the outline of the jackfruit pulps cells structures were distinguished, showing the shapes and structures of the cells and intercellular spaces. Matior Rahman *et al.* (1995) using light microscope and scanning electron microscope found that in immature jackfruit, the perianth was predominantly thin-walled cells packed with starch granules. The starch granules reduced in number and in size as the fruit ripen.

Figure 3.4 showed the light microscopy plate of untreated ripe jackfruit pulps. No starch granules were visible in Figure 3.4, indicating the fruit had attained full ripeness. The cells of the untreated ripe jackfruit pulps were round and intact with the plasmalemma following the shape of the cell wall. All organelles are within the cell wall and plasmalemma boundary.

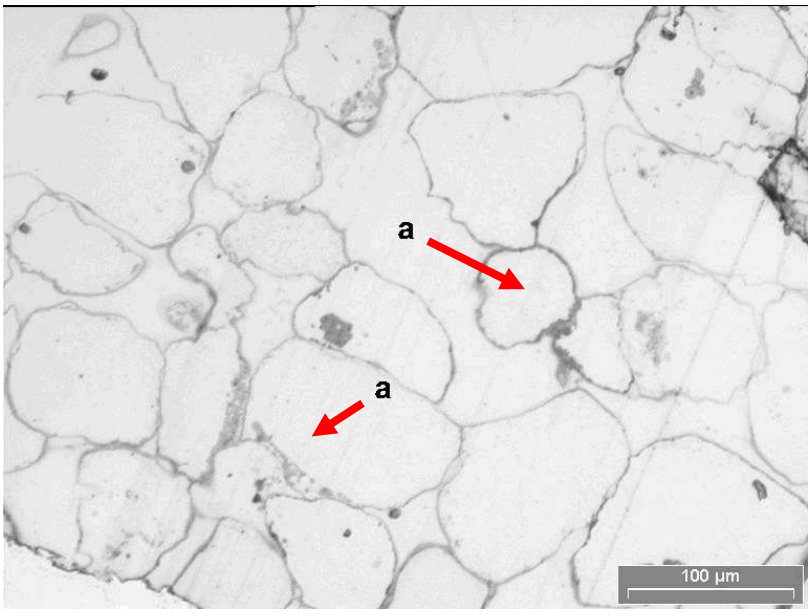


Figure 3.4: Light microscopy images of untreated ripe jackfruit pulps at day 0; "a" indicates intact cell walls. (Magnification: 300x).

The cells of the ripe jackfruit pulps shown in Figure 3.4 were closely packed, with some intercellular spaces, indicating dissolution has not commenced and the middle lamella are still connected. This accounts for the firm texture of ripe jackfruit pulps. Pesis *et al.* (1978) found that the middle lamella of unripe avocado was intact, and the fibrils on both sides of the middle lamella were tightly packed. Anino *et al.* (2005) found that in fresh apple tissue, the cells and intercellular spaces were loosely arranged and the middle lamella well defined.

The cell wall structure after 14 days of storage is shown in Figure 3.5. The cell wall structure of control samples displayed advanced disintegration. This is corroborated by the texture data in Figure 3.1, where after 13 days the jackfruit pulp became soft and limp.

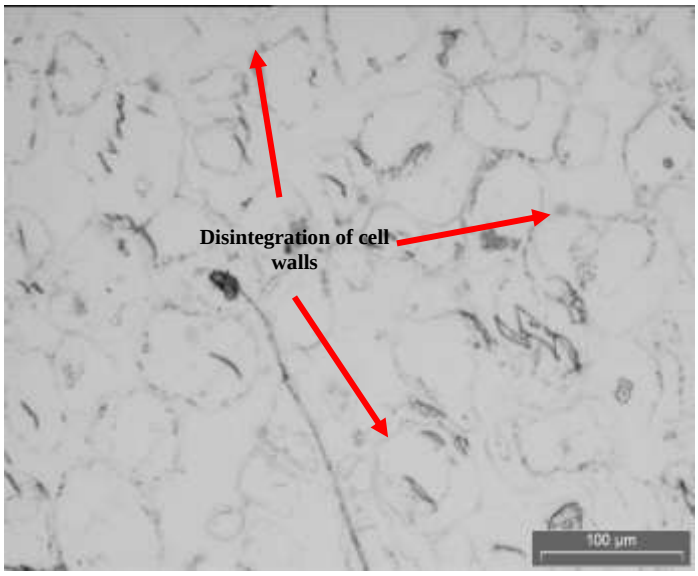


Figure 3.5: Light microscopy images of untreated ripe jackfruit pulps at day 14. Arrows showed dissolution of the ripe jackfruit cell walls. (Magnification: 300 x).

The cells also appeared separated from one another, indicating weakening middle lamella and indistinct cell walls structures. The sizes of the cell, at the same magnification, were smaller than at day 0 (Figure 3.4) and in some cells the plasmalemma seemed to have shrunk from the cell walls. Empty regions appeared in the cell walls and the middle lamella had undergone dissolution in some areas. The fibrils in the cell walls appeared as long, dissociated thread, while the inner parts of the walls were somewhat intact.

In study on avocado by Pesis *et al.* (1978), the cell walls lose their structure as the fruit softened. Platt-Aloia *et al.* (1980) found that in avocado cells as fruit approached climacteric peak, a decrease in the electron density was observed and the structure of middle lamella weakened. Wall striations were evidently separated. Sterling (1954) found that the parenchymatous cells of pears became thin and weak in overripe fruits. Ben-Arie *et al.* (1979) study on pears and apples found that the cell walls appeared to have undergone dissolution simultaneously with softening.

Apple fruits immersed for 2 hours in calcium solution revealed calcium crystals between the cell walls and the plasmalemma (Alzamora *et al.*,1997), detaching the cytoplasma further from the cell walls. Fruit cells that was immersed for 6 hours in calcium chloride solution showed cytoplasma separated from the cell walls and the membrane were broken with vesicle formation. After 22 hours immersion, cell membranes were completely disrupted. After immersion in isotonic solutions, tissue softening occurred due to pectin solubilization and hydrolysis (Alzamora *et al.*, 2005).

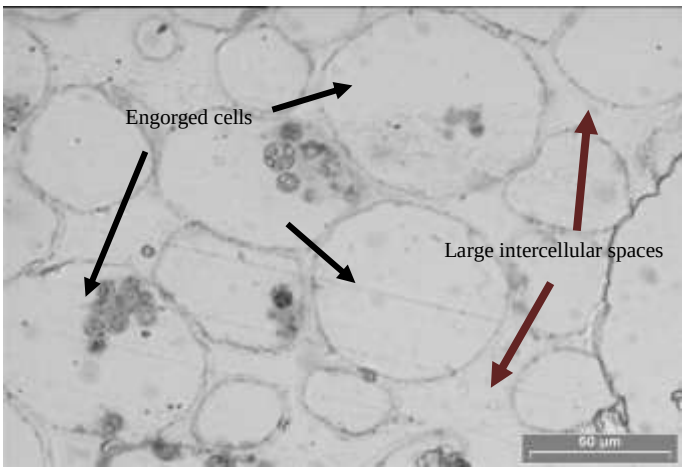


Figure 3.6: Light microscopy images of ripe jackfruit pulps immersed in 0.5% $CaCl_2$ solution for 18 hours at day 0. (Magnification: 300 x).

Figure 3.6 illustrated the light microscopy image of ripe jackfruit pulps that were immersed in 0.5% calcium solution for 18 hours at day 0. The fruit cells appeared intact, with some organelles visible. The diffusion of calcium solution into the fruit matrix caused some cells to engorge, while some remained the same as those seen in untreated ripe jackfruit pulps at day 0 (Figure 3.4). The replacement of gases by calcium solution by immersion method may have caused the increase in intercellular spaces observed in immersed jackfruit pulps sample.

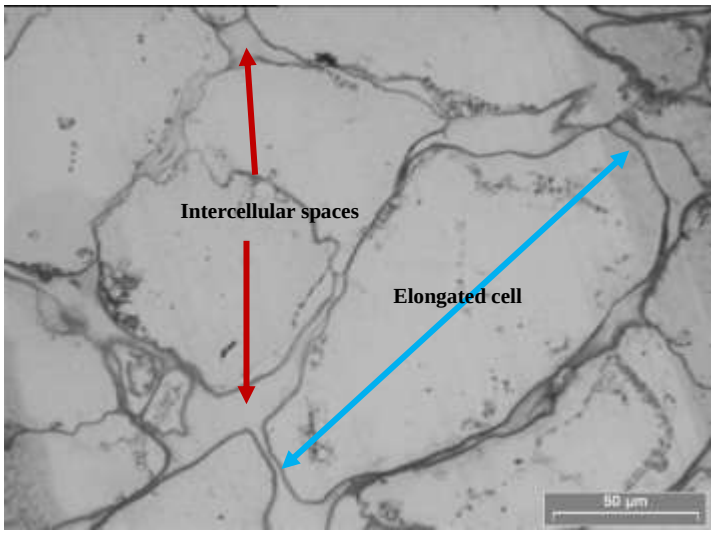


Figure 3.7: Light microscopy images of ripe jackfruit pulps immersed in 0.5% $CaCl_2$ solution for 18 hours at day 14 showing large intercellular spaces (red arrows) and elongated cells (blue arrow) . (Magnification: 300x).

Figure 3.7 exhibited well integrated cell walls of immersed ripe jackfruits pulps after 14 days of storage at 10^oC. The cell walls were still intact and closely adjacent to each other. The cells appeared elongated and no sign of organelles seen in immersed ripe jackfruit pulps after 14 days. According to Knee and Miller (2002) the parenchyma which are the edible parts of the fruit, are more or less isodiametric. However, they are elongated along the radial axis of the fruit. The plasmalemma of the ripe jackfruit cells shown in Figure 3.7 had also contracted. All the black particles observed at day 0 were enclosed in the shrunken plasmalemma.

The diffusion of calcium into the ripe jackfruit cells also brought about an increase in the amount of liquid in the cell system. Some of the gases found in the intercellular spaces are trapped and tend to expand during storage. This is demonstrated by the large intercellular spaces.

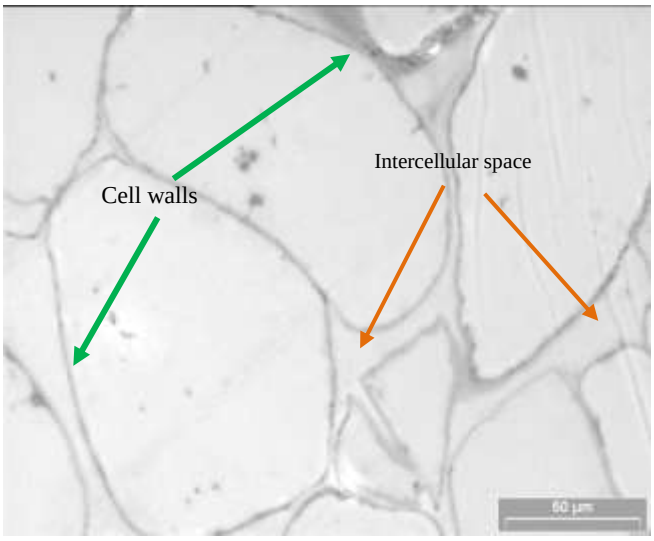


Figure 3.8: Light microscopy images of ripe jackfruit pulps vacuum-infused with 1.0% $CaCl_2$ at day 0. (Magnification: 300x).

Figure 3.8 showed the light microscope image of ripe jackfruit pulps infused with 1.0% calcium through vacuum-infusion. The cell walls were closely attached, indicating intact cell walls. The intercellular spaces were relatively small as shown by the brown arrows. The use of vacuum to introduce the calcium solution into the fruit matrix in this study is sufficient to replace the gases in the intercellular spaces with the calcium solution, and at the same time was gentle enough to not cause the cell walls to collapse.

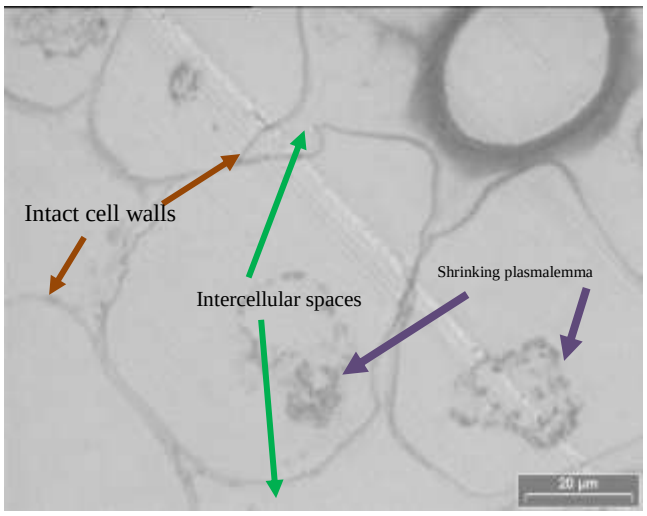


Figure 3.9: Light microscopy images of ripe jackfruit pulps vacuum-infused with 1.0% $CaCl_2$ at day 14. (Magnification: 300x).

Figure 3.9 showed the cells of ripe jackfruit pulps treated with 1.0% calcium after 14 days of storage. The cell walls structures appeared to be intact, with large intercellular spaces. The plasmalemma has shrunk away from the cell walls. The shrinkage of the plasmalemma can be explained due to the flow of the liquid phase from the intercellular spaces into the cell cavity through the cell walls. The same was also observed by Salvatori *et al.* (1998) in apples treated with pulsed vacuum for osmotic dehydration.

Martinez-Monzo *et al.* (1998) elucidated throughout storage, the cell walls of apples started to relax and the cell wall-plasmalemma layers began to separate. Suction effect also promoted

cells to expand and intercellular spaces were filled with external liquid, ensuing to an increase in volume and cells rearrangement (Fito *et al.*, 2000). They also reported that the vacuum treated apples with high osmotic solution concentration exhibited better cell wall roundness, showing reduced plasmalemma volume in the middle of the recovered cell volume.

3.2.2. Transmission Electron Microscopy (TEM)

The transmission electron microscopy was done on stored ripe jackfruit pulps to demonstrate the change in the cell wall structures, especially in relation to the middle lamella. Images of ripe jackfruit pulps at day 0 showed the initial cell wall structure, while the images at day 14 illustrated the changes occurring in the middle lamella and cell wall of the ripe jackfruit pulps after being stored for 14 days at 8^{o}C.

The transmission electron microscope will demonstrate how the treatment method influenced the cell walls structures. Degradation of middle lamella has been claimed to be responsible for cell wall separation and an increase in intercellular spaces as fruit matures (Reeve, 1970; Ben-Arie *et al.*, 1979a and b; Glenn and Pooviah, 1990 and Marangoni *et al.*, 1995).

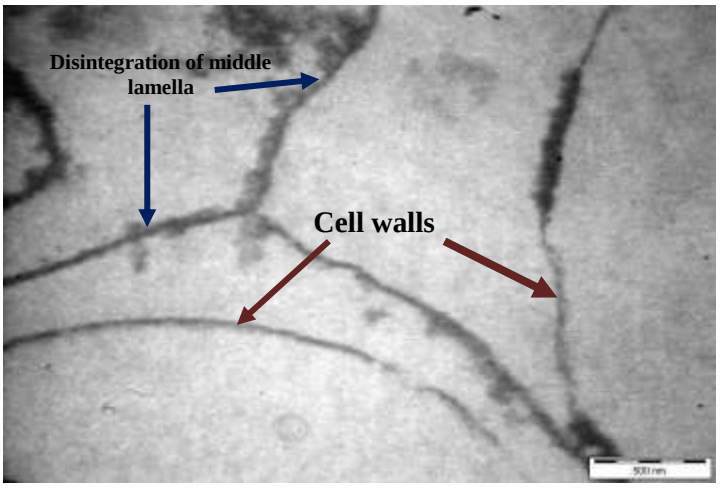


Figure 3.10: Transmission electron microscopy showing the disintegration of middle lamella in untreated ripe jackfruit pulps at day 14.

Figure 3.10 showed the transmission electron microscope (TEM) pictures of untreated ripe jackfruit pulps after 14 days of storage at 8_{o}C. Although the cell walls shown in Figure 3.10 appeared intact, severe degradation of the middle lamella can be observed. In some areas, part of the middle lamella has disintegrated and fragments of the cell were evident. Figure 3.10 further illustrated the disintegration of middle lamella and the cell walls appeared to be well separated. Some parts of the cell walls also seem to have receded.

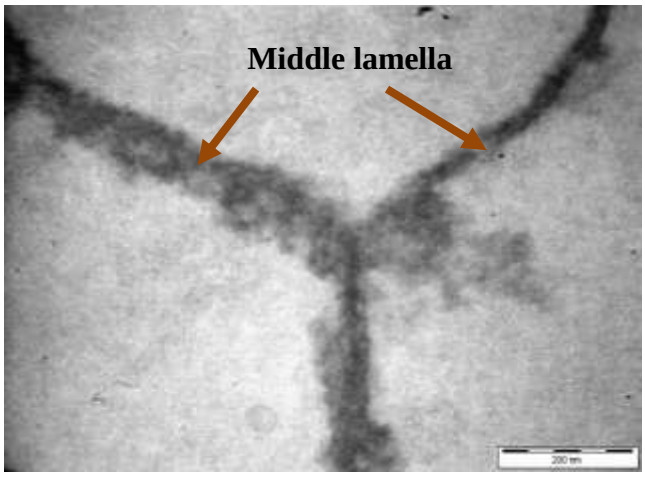


Figure 3.11: Transmission electron microscopy showing the dissolution of the middle lamella of untreated ripe jackfruit pulps at day 14.

The dissolution of middle lamella is also illustrated in Figure 3.11. The dissolution of the middle lamella in Figure 3.11 caused the cells to lose the "cementing" effect that keep the adjacent cells bonded. Crookes and Grierson (1973) explained that as ripening progressed, extensive dissolution of cell walls was observed, especially on the primary cell walls. This coincides with the buildup of PG activity, which caused dissolution of the middle lamella and the release of fibrous components from the primary cell walls (data not shown).

Disruption of middle lamella brought about the separation of the adjacent primary cell walls. This is demonstrated in Figure 1 by the loss of firmness in untreated ripe jackfruit pulps after storage for 13 days. The increase in the PE activity during storage may have trigger off the breakdown of middle lamella structures observed in Figure 3.11.

Figures 3.12 and 3.13 showed the TEM of ripe jackfruit pulps immersed in 1.0% calcium solution for 18 hours after 14 days storage at 8^{o}C. The cells exhibited progressive deterioration of the middle lamella and cell wall structures. In Figure 3.12, the deformation of the cells' edges can be clearly observed, suggesting the disintegration of the cell components. Figure 3.13 showed the separation of adjacent cell walls, bringing about cells detachment.

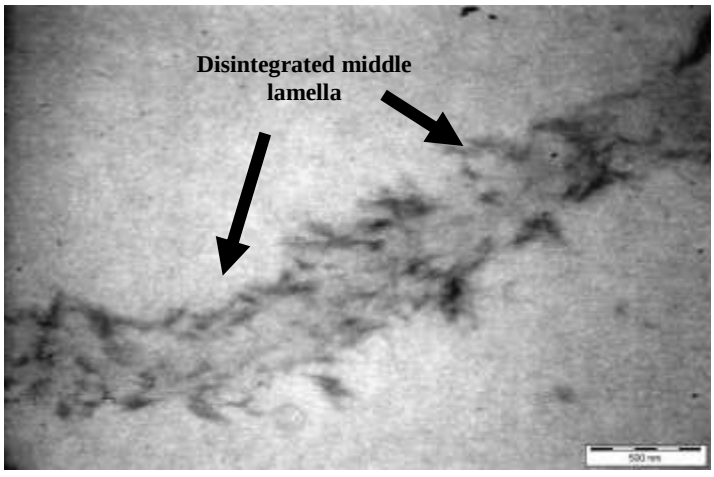


Figure 3.12: TEM of ripe jackfruit pulps treated in 1.0% calcium solution by 18 hours immersion at day 14 showing disintegrated middle lamella.

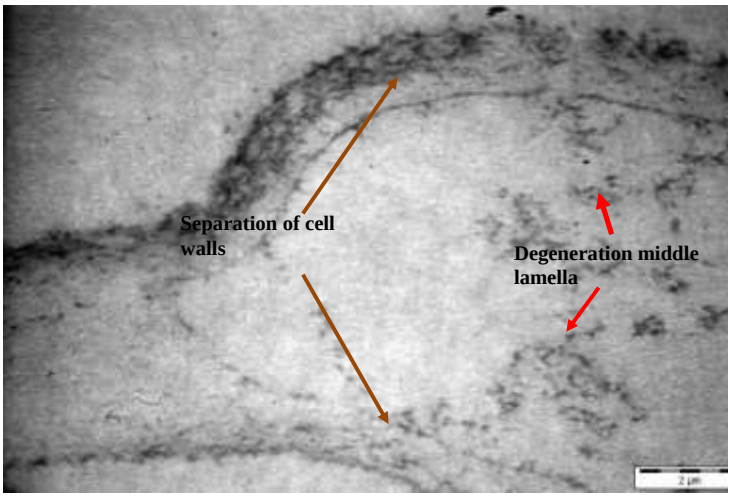


Figure 3.13: TEM of ripe jackfruit pulps treated in 1.0% calcium solution by 18 hours immersion at day 14 showing cell wall separation.

Transmission electron miscopy study of immersed ripe jackfruit cells in this study exhibited degradation of middle lamella and cell-to-cell separation. According to Mauro et al. (2002), extended exposure of potatoes to osmotic solutions in equilibrium led to degradation of cell structure. As the cell wall structure is an important factor in maintaining texture, the loss in firmness is escalated.

Calcium in immersed treatment also provoked severe internal disruption in the cell, and considerable softening of the tissue occurred in spite of the clearly reinforced cell walls. This is corroborated by Alzamora et al. (2005) where the cell membranes of apples immersed in calcium solution showed complete disruption after 22 hours. Study by Quiles *et al.* (2004) on Granny Smith apple rings immersed in 40% calcium for 30 minutes found denser middle lamella and the cell-to cell contact which were intact compared to untreated fruits. These researchers used sodium dodecyl sulphate (SDS) to facilitate penetration of the calcium ions; however this resulted in alteration and perforation of the cellular walls, which ultimately caused the turgor loss.

However, that is not the case in the ripe jackfruit pulps in this study. Although the texture of immersed ripe jackfruits pulps were firm up to day 3, the dissolution of the middle lamella and cells degradation ultimately caused the fruit to lose its firmness on day 7. The disintegration of middle lamella further caused the pulps to become fibrous, which translated to the stringiness that instigated the deceptive increase in the texture data perceived on days 10 and 14.

Figures 3.14 and 3.15 exhibited the TEM images of the middle lamella of ripe jackfruit pulps at 14 days of storage after vacuum-infused with 1.0% calcium. The ripe jackfruit pulps demonstrated a remarkable preservation of middle lamella even after 14 days of storage. Figure 3.14 showed a thickening of middle lamella and cell walls. The integrity of the cells was maintained and cellular detachment was minimized.

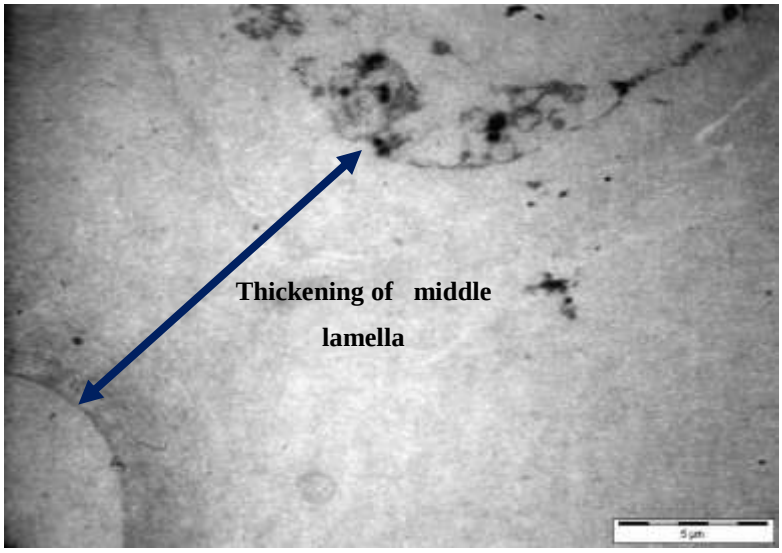


Figure 3.14: TEM of jackfruit vacuum-infused in 1.0% calcium at day 14, showing thickening of middle lamella.

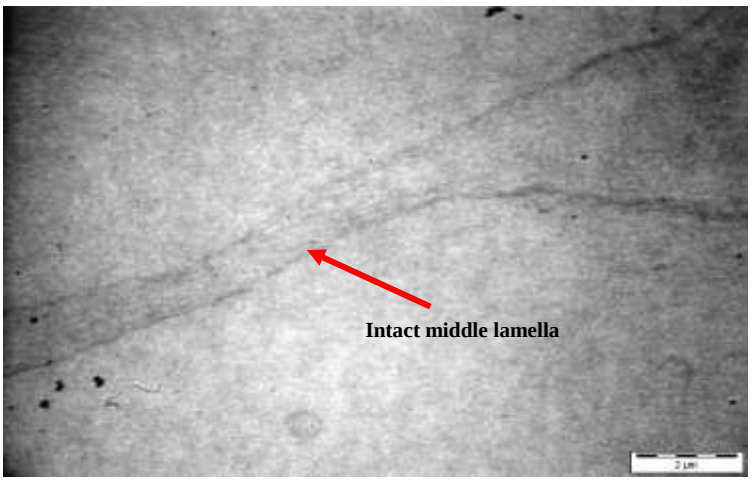


Figure 3.15: TEM of ripe jackfruit pulps vacuum-infused in 1.0% calcium solution at day 14.

Figure 3.15 also showed intact middle lamella of the ripe jackfruit pulps. No sign of dissolution was observed. This correlate to the texture data of ripe jackfruit pulps vacuum-infused with 1.0% calcium after 14 days storage (Figure 3.3).

Data gathered from the microscopy study indicated that the cell walls of ripe jackfruit pulps stored at 8^{o}C underwent complete dissolution at day 14. Treatment with calcium initially helped maintained the integrity of the middle lamella of the cell walls. In ripe jackfruit pulps immersed in calcium for 18 hours, the middle lamella exhibited dissolution by day 14. The intercellular spaces were large, and cells appeared engorged. The infusion of calcium into the jackfruit pulps matrix also brought about an increase in the fluid content of the cells. Increased fluid caused an increase in cells volume which instigated the plasmalemma to rupture. This was demonstrated by the separation of plasmalemma from the cell walls in Figure 3.13.

In vacuum-infused ripe jackfruit pulps, the cell walls exhibited that was close to one another indicating the cementing effect of an intact middle lamella. The TEM images of vacuum-infused ripe jackfruit pulps (Figures 3.14 and 3.15) showed middle lamella that was still undamaged. Hence, although the addition of calcium helps in maintaining the firmness of the stored ripe

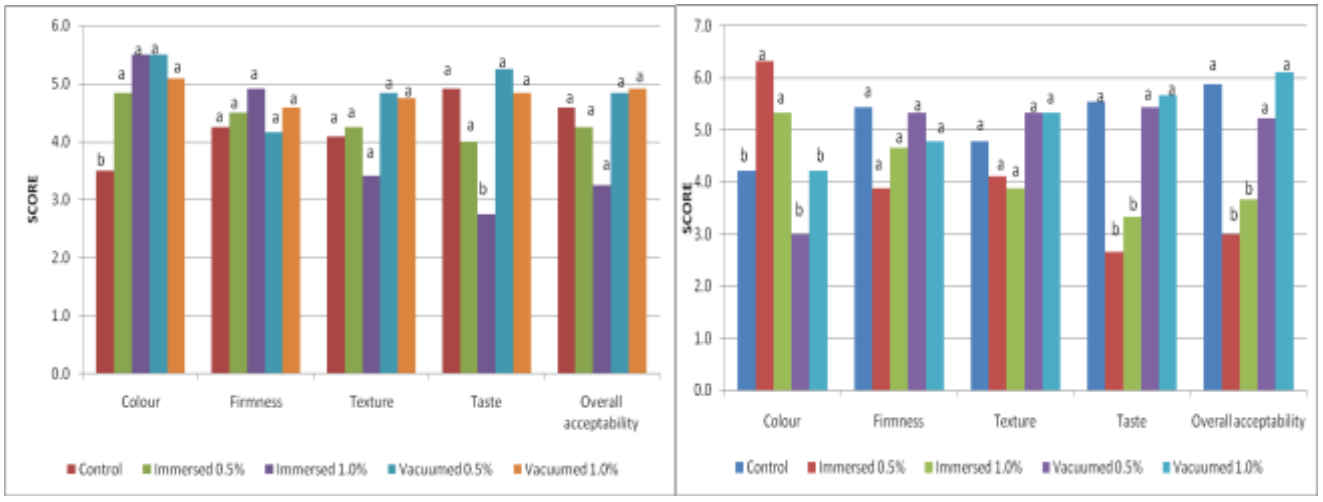
jackfruit pulps, the method of infusion played an important factor in the effectiveness of the calcium in maintaining the texture of the jackfruit pulps. In vacuum-infusion, the pulps were exposed to the calcium solution for only 15 minutes. The duration of exposure was much shorter as compared to in immersion method where exposure time was for 18 hours. The vacuum also expelled the gases in the intercellular spaces. The intercellular cells were filled with calcium solution, which assimilated into the middle lamella, thus strengthening its structure. This ensures the firmness was maintained during storage. Thus, although the concentration of calcium solution did not affect the preservation of firmness in stored ripe jackfruit pulps, the method of infusion did. The ripe jackfruit pulps vacuum-infused with calcium exhibited the best preservation of firmness.

3.2.3 Sensory Evaluation

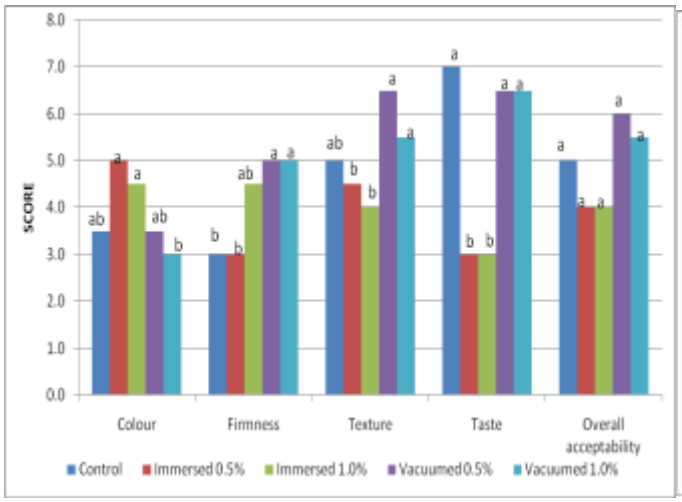
The ripe jackfruit pulps were evaluated for the sensory attributes during storage at 8^{o}C. The attributes investigated were colour, firmness, texture, taste and overall acceptability. Figure 3.16 showed the score obtained for all the samples tested on day 0, immediately after calcium treatment to day 14.

The panelists found most all the samples to be acceptable on day 0 and day 3. However, ripe jackfruit pulps infused with calcium through immersion method showed lower score for taste as compared to the other samples. The panelists commented on bitter taste in the immersed samples. In immersion method, the ripe jackfruit pulps were immersed in the calcium solution for 18 hours. The long immersion time involved may have caused the bitterness in taste. In vacuum-infusion, the shorter contact time of 15 minutes resulted in no detection of bitterness by the panelists.

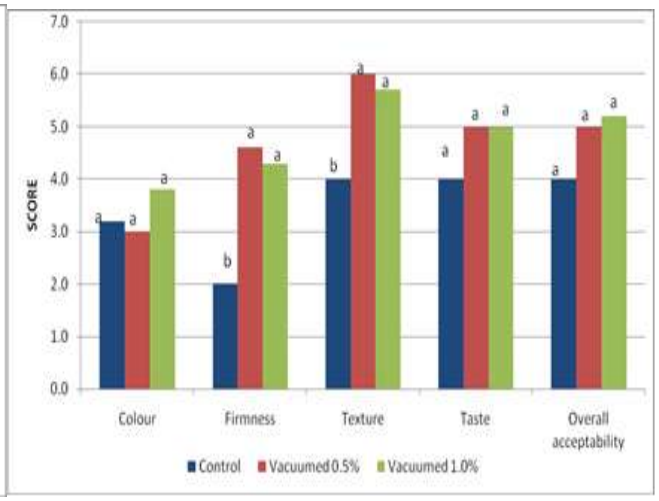
The panelists consistently rated the colour of the immersed jackfruit higher than the other samples. Osmotic process was involved in pulps treated with calcium by immersion. This resulted in changes of the products in term of the optical and mechanical properties (Torreggiani, 1995). These changes are also directly related to the colour, appearance and texture of the products (Chiralt *et al.*, 1999; Talens *et al.*, 2002). This may explain why immersed pulps have higher marks for colour as it was exposed to osmotic process the longest.



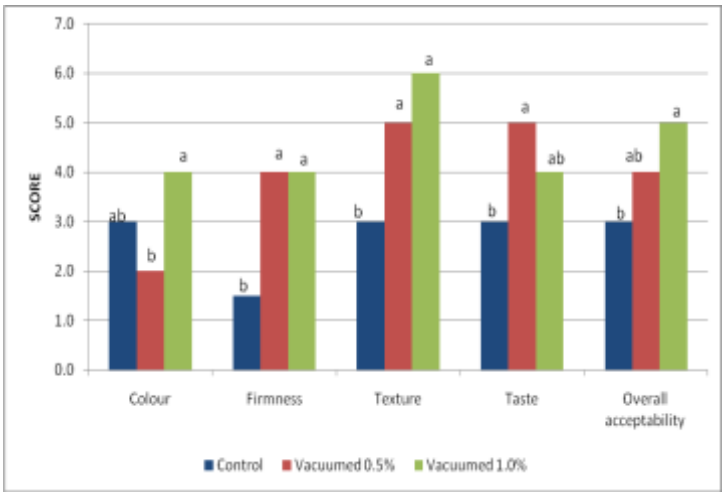
Day 0 Day 3



Day 7



Day 10



Day 14

Figure 3.16: (a) Score for the untreated (control) and calcium-treated ripe jackfruit pulps during storage **(b)** Data are mean value ± standard deviation (n=25). **(c)** Superscript with the same alphabet signifies no significant difference between treatments on the same day of storage ($p \leq 0.05$).

The ripe jackfruit pulps vacuum-infused with 0.5% and 1.0% calcium scored significantly higher for texture and taste attributes on day 7, than the immersed ripe jackfruit pulps. The immersed jackfruit pulps were consistently rated low for taste. However, for overall acceptability, ripe pulps immersed in calcium solution were rated as “neither like nor dislike”.

The firmness and taste quality of ripe pulps infiltrated with calcium by immersion were considered unacceptable by the panelists. The immersed pulps were lower in score than the untreated ripe pulps in term of texture. Based on the texture data, the immersed ripe pulps showed an increased in texture on day 7. However, this increase was not due to the increase in firmness. The immersed samples indicated signs of textural disintegration. An unpleasant smell was detected on day 7 and some panelists found that the firmness/texture was slightly “mushy”. The samples were not evaluated for sensory after 7 days of storage.

On day 10, the untreated pulps scored lower than the vacuum-infused samples on all attributes except colour. The reduction in firmness, texture, taste and overall acceptability of the untreated ripe jackfruit pulps were significant ($p \leq 0.05$) from day 7 to day 10. This showed that the

disintegration in quality of the untreated ripe jackfruit pulps were severe after 7 days of storage at 8^{o}C.

On day 14, the untreated ripe pulps were considered inferior by the panelists, except for colour. The vacuum-infused pulps scored high for texture, indicating that the texture of the products were still acceptable to the panelists even after 14 days of storage.

The sensory evaluation of the ripe jackfruit pulps treated with calcium by different methods of infiltration suggested that both the methods contributed differently to the finished products acceptability. The immersed ripe jackfruit pulps showed good rating for colour during storage as compared to the other samples. The osmotic process which occurred during immersion may help to improve the optical characteristics of the pulps. However, long duration of immersion resulted in the bitter taste as observed by the panelists in immersed ripe jackfruit pulps. This bitter taste was not as assertive in ripe jackfruit pulps infused with calcium by vacuum-infusion.

The shorter contact time during infusion of calcium in the vacuum-infused pulps did not hinder the effect of calcium in preserving the firmness and texture of the ripe jackfruit pulps. This was shown by the high rating received by vacuum-infused pulps for texture and firmness even after 14 days of storage. Unlike immersed pulps, the long infusion time failed to maintain the firmness and texture of the pulps after 7 days. Untreated ripe jackfruit pulps were unacceptable by 10 days of storage.

4. Conclusions

In this study, it was proven that infusion of calcium into the ripe jackfruit pulps helps preserve the textural characteristics of the fruit during storage. Employing higher concentration of calcium solution, 1.0%, did not necessarily suggest better texture retention. However, calcium chloride solution at 1.5% and 2.0% imparted bitter taste to the fruits.

Comparison of infusion methods indicated the vacuum-infusion as the best method of calcium infusion. Texture of the fruits was retained for up to 14 days. The low temperature during vacuum-infusion ensures the structures at cellular level remain intact. Although contact time in calcium solution was shorter than immersion technique, the application of vacuum facilitates calcium solution to replace gases present in the intercellular spaces. This is an advantage over immersion method as intercellular gases may cause quality deterioration. Study under the transmission electron microscope showed evidence of intact cell walls and middle lamella. Incorporating calcium into the jackfruit by vacuum-infusion helps preserve the texture of jackfruit pulps, while maintaining other sensorial characteristics for at least 14 days. Hence, it can be concluded that jackfruit pulps vacuum-infused with 0.5% calcium solution for 15 minutes is better than immersion in calcium solution at 8 ^{o}C for 18 hours.

Acknowledgments

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Conflict of Interest

The authors declare no conflict of interest.

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