

*Full Length Research Paper*

# **Influence of Ca CaCl<sub>2</sub> dipping on postharvest quality and shelf life of bell pepper (*Capsicum annuum* L. cv. California Wonder)**

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**Bell pepper (*Capsicum annuum* L.) experiences significant loss during postharvest handling. This study aimed to provide an alternative postharvest handling technology for the vegetable. The factor studied was Ca CaCl<sub>2</sub> at 4 levels: 0% (distilled water), 3, 6 and 9%. Weight loss and total soluble solids were determined at an interval of 3 days whereas total chlorophyll, Fe, ascorbic acid and Ca content were determined at an interval of 4 days. Shelf life elapsed when fruit lost 25% of their initial weight. Calcium CaCl<sub>2</sub> reduced weight loss by up to 16.7%, increased Ca content by up to 252.7%, maintained total soluble solids by up to 11.8%, total chlorophyll by up to 23.3%, Fe by up to 10.1%, ascorbic acid by up to 13.9% and Ca content by up to 13.7% and extended shelf life by up to 3 days. Calcium CaCl<sub>2</sub> at 6% was the best treatment and therefore can be used by bell pepper growers, retailers and consumers to maintain postharvest quality of bell pepper.**

**Key words:** Chemical treatment, fruit vegetable, fresh weight loss, ascorbic acid.

## **INTRODUCTION**

Postharvest losses in horticultural crops in developing countries is as high as 45% due to poor postharvest handling (Kitinoja and Kader, 2015); and is even higher in Sub-Saharan Africa (SSA) (Kitinoja and Kader, 2015). In bell pepper (*Capsicum annuum* L.), losses of 28.6 and 38.7% have been reported during dry and wet seasons, respectively in Nigeria (Tsegay et al., 2013). A short shelf life, even under the most favourable conditions is a major postharvest limiting factor in bell pepper handling (Ilić et al., 2017). Since bell pepper is a non-climacteric fruit, its

senescence is mainly accelerated by excessive water loss through respiration.

Calcium CaCl<sub>2</sub> has shown promising results in maintaining quality and improving shelf life of fruits and vegetables such as tomato (Demes et al., 2021), pear (Sajid et al., 2014), apple (Shirzadeh et al., 2011), apricot (Liu et al., 2017), African eggplant courgettes (var. *cylindrica*) (Chepngeno et al., 2016), strawberry (Jouki and Khazaei, 2012) and green bell pepper (Bagnazari et al., 2018).

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Low temperature storage and Modified Atmosphere Packaging (MAP) have been successfully used in maintaining quality and extending shelf life of bell pepper. The most effective method has been rapid cooling after harvest followed by storage at low temperature and high relative humidity (Bayogan et al., 2017). Bell pepper being a tropical fruit, suffers chilling injury at temperatures below 7°C, which favors development of fungal diseases (Ilić et al., 2017). The cost of purchasing, installing and running a cold storage facility is also high and unaffordable for most small scale bell pepper growers, retailers and consumers in developing countries; hence, rendering the technology untenable.

Modified Atmosphere Packaging using plastic bags has also been used for a long time for maintenance of quality of bell pepper. However, their use may trigger development of anaerobic microorganisms (Manolopoulou et al., 2012). These together with the restricted use of plastics in several countries due to environmental pollution have made the technology unreliable. Therefore, the objective of this study was to determine the effects of Ca CaCl<sub>2</sub> dipping, an alternative postharvest treatment, on postharvest quality and shelf life of bell pepper.

## MATERIALS AND METHODS

### Experimental site

The study was conducted at Egerton University, Njoro, Kenya. Egerton lies at a latitude 0°23' south, longitude 35°35' East; and is 2,200 m above sea level (Jaetzold and Schmidt, 2006). Average minimum temperature, maximum temperature and relative humidity of the laboratory site was 11 °C, 24.5 °C and 64.7%, respectively (Egerton Meteorological Weather Station, 2020).

### Experimental materials

Bell pepper (cv. California Wonder), fruit was produced at the Horticulture, Research and Teaching Field of Egerton University, Njoro, Kenya under white agronet covers. Fruits were harvested at mature green stage, packed in plastic buckets, and taken to the laboratory, where fruits free from bruises and blemishes were selected and used for the study. Calcium CaCl<sub>2</sub> which was purchased from Pyrex Kenya Limited Company, Nairobi, Kenya was used for the study.

### Preparation of Ca CaCl<sub>2</sub> solutions

A stock solution of CaCl<sub>2</sub> [25% weight per volume (w/v) CaCl<sub>2</sub>] was prepared by weighing 250 (g) of CaCl<sub>2</sub> using an electronic weighing balance (Denver Instrument XL-1810, USA) and dissolving in 1,000 (ml) of distilled water. Working solutions (3%, 6% and 9% CaCl<sub>2</sub>) (Jan et al., 2013) volumes were then obtained from the stock solution using the formula below;

$$C_1V_1=C_2V_2$$

Where, C<sub>1</sub> - Concentration of the working solution. V<sub>1</sub> - Volume of the stock solution; C<sub>2</sub> - Concentration of the stock solution.

V<sub>2</sub> - Volume of the working solution (Osuji et al., 2016)

The volumes obtained were then diluted to 1,000 ml mark using distilled water to obtain 3, 6 and 9% CaCl<sub>2</sub>. Distilled water served as 0% CaCl<sub>2</sub> and was used as a control treatment.

### Treatments application

Before treatment application, all fruits were disinfected by washing for 5 min using 0.5% (v/v) NaClO (Lerdthanangkul and Krochta, 1996). This was followed by air drying of fruit at room temperature (25 ± 2°C) until the disinfecting solution on fruit skin was completely dry. Based on the treatments, the fruits were dipped in CaCl<sub>2</sub> solutions placed in a water bath at 60 °C for 5 min (Shirzadeh et al., 2011). They were then removed and allowed to air-dry at room temperature until CaCl<sub>2</sub> solutions on fruit skin was completely dry. Control fruits were dipped in distilled water for 5 min, removed and allowed to air-dry at room temperature (25 ± 2 °C) until distilled water on fruit skin was completely dry. After treatments application, all fruits were stored on plastic trays under ambient conditions (25 ± 2°C temperature and 65 ± 2% relative humidity) until they senesced.

### Experimental design

The experiment was a single factor experiment arranged in a randomized complete block design, with 3 replications. Blocking was done against different harvesting times; harvesting of the 3 blocks was done at 1 month interval. In total, there were 12 experimental units with each experimental unit represented by a plastic tray containing 30 fruits.

### Data collection

Data collection commenced immediately after treatments application and continued until fruit lost 25% of their initial weight on average (Sibomana et al., 2015). Data collection was done on fresh weight loss and total soluble solids (TSS) at 3 days intervals; and total chlorophyll, Fe ascorbic acid and Ca content at 4 days intervals. 3 fruits per experimental unit were selected at random at the onset of the study, marked and used for data collection throughout the study for non-destructive variables which were fresh weight loss and shelf life. On the other hand, 3 fruits per experimental unit were also randomly selected from the remaining fruits and used to collect data for the destructive variables (TSS, total chlorophyll, Fe, ascorbic acid and Ca content). Data for each destructive variable was collected from the 3 fruits with a new set of fruits used on each sampling date. The variables were determined as described below.

### Percentage (%) fresh weight loss

The fresh weight (g) of the 3 selected fruits per experimental unit was measured using an electronic weighing balance (Denver Instrument XL-1810) immediately after treatment application (before storage). The same fruits were thereafter weighed at 3 days intervals until they lost 25% of their initial total weight. Progressive % fresh weight loss was determined using the formula by Moneruzzaman et al. (2008). Average % fresh weight loss of the 3 fruits was calculated and recorded as average % weight loss per fruit for the time period (Moneruzzaman et al., 2008). The shelf life of the fruits on the other hand was determined by counting the number of days the fruits took from harvesting to lose 25% of their initial weight (Sibomana et al., 2015).

### Total soluble solids content

Total Soluble Solids (%TSS) content was determined using a portable hand-held refractometer RHB-32/ATC (YHEQUIPMENT CO., LIMITED, Shenzhen City, China) as described by Opiyo and Ying (2005). A small piece of pepper fruit was cut and squeezed and the juice obtained dropped onto a refractometer and readings taken. Average %TSS of the 3 fruits was calculated and recorded as average %TSS per experimental unit for the time period.

### Iron content

Iron (Fe) content was determined using an Atomic Absorption Spectrophotometer (model 210 VGP, Buck Scientific, Norwalk, CT) following Jones and Case (1990). Dried ground sample (1 g) was weighed into crucibles and ashed in a furnace at a temperature of 550 °C for 2 h. The ash was cooled to room temperature ( $25 \pm 2$  °C), transferred into a 100 mL beaker and 10 mL of the digestion mixture added. Distilled water (50 mL) was added. Activated charcoal (1 g) was added to obtain a clear sample and stirred. The contents were gravity filtered through Whatman No.5 filter paper into a 100 mL volumetric flask. The filtrate was filled to the mark with distilled water. Into a cuvette, 10 mL of filtrate was pipetted and absorbance read at 248 nm. Iron standard solutions of 0, 5, 10, 15, 20 and 25 µg/g were prepared from FeSO<sub>4</sub>. Into a cuvette 10 mL of each standard solution was pipetted and absorbance read at 248 nm, and a standard curve developed. The amount of Fe was calculated against the standards, converted to µg/g and expressed using the formula of Okalebo et al. (2002).

### Calcium content

Calcium (Ca) content was determined using an Atomic Absorption Spectrophotometer (model 210 VGP, Buck Scientific, Norwalk, CT) following Jones and Case (1990). Dried ground sample (1 g) was weighed into crucibles and ashed in a furnace at a temperature of 550 °C for 2 h. The ash was cooled to room temperature ( $25 \pm 2$  °C), transferred into a 100 mL beaker and 10 mL of the digestion mix added. Distilled water (50 mL) was added. Activated charcoal (1 g) was added to obtain a clear sample and stirred. The contents were gravity filtered through Whatman No.5 filter paper into a 100 mL volumetric flask. The filtrate was filled to the mark with distilled water. Into a cuvette, 10 mL of filtrate was pipetted and absorbance read at 422 nm. Calcium standard solutions of 0, 5, 10, 15, 20 and 25 µg/g were prepared from CaCl<sub>2</sub>. Into a cuvette 10 mL of each standard was pipetted and absorbance read at 422 nm, and a standard curve developed. The amount of Ca was calculated against the standards, converted to mg/100g and expressed using the formula of Okalebo et al. (2002).

### Total chlorophyll content

Total chlorophyll content (µg·g<sup>-1</sup>) in bell pepper fruits was determined following Goodwin and Britton (1988). A solution of acetone and hexane was prepared in a ratio of 4:5. Fresh fruits sample of 0.5 g was weighed and ground in 10 mL of acetone-hexane mixture with a mortar and pestle. The homogenate was transferred into 50 mL Eppendorf tubes and volume made to 25 mL using acetone: hexane mixture. It was centrifuged at  $2,683 \times g$  for 10 min. A cuvette was filled with distilled water, placed inside a spectrophotometer (Shanghai 712, China), set at a wavelength of 645 nm and then at 663 nm and absorbance of 0.00 read. The same procedure was repeated with the supernatant, absorbance

read, and the average values were recorded. Readings were taken from each of 3 fruits per experimental unit. Total chlorophyll concentration in the supernatant was calculated from absorbance at 645 nm and 663 nm using the formula of Goodwin and Britton (1988).

### Ascorbic acid content

Ascorbic acid (Vitamin C) was determined by titration with 2, 6-dichloro-phenol-indophenol dye following a standard procedure (AOAC, 1990). Using an electronic weighing balance (Denver Instrument XL-1810, USA); 10 g of fruit sample was weighed. The weighed fruit sample was extracted in 20 mL 5% oxalic acid using a mortar and pestle, and then gravity filtered through cotton wool. Ascorbic acid standard solution was prepared by dissolving 0.05 g of pure ascorbic acid in a small volume of 5% oxalic acid solution and then diluting to 250 mL with the same oxalic acid solution. Ascorbic acid standard solution (10 mL) was titrated with 0.005% indophenol solution to a persistent slight pink colour end point and 10 mL of oxalic acid as a blank. The amount of ascorbic acid corresponding to 1 mL of indophenol solution was calculated. Into a 50 mL flask, 10 mL of the gravity filtered sample extract was pipetted and made to the mark with the 5% oxalic acid solution. The standard indophenol solution was used to titrate 10 mL of the filtrate to a slight pink end point. Vitamin C content was calculated following Obel et al. (2019).

### Data analysis

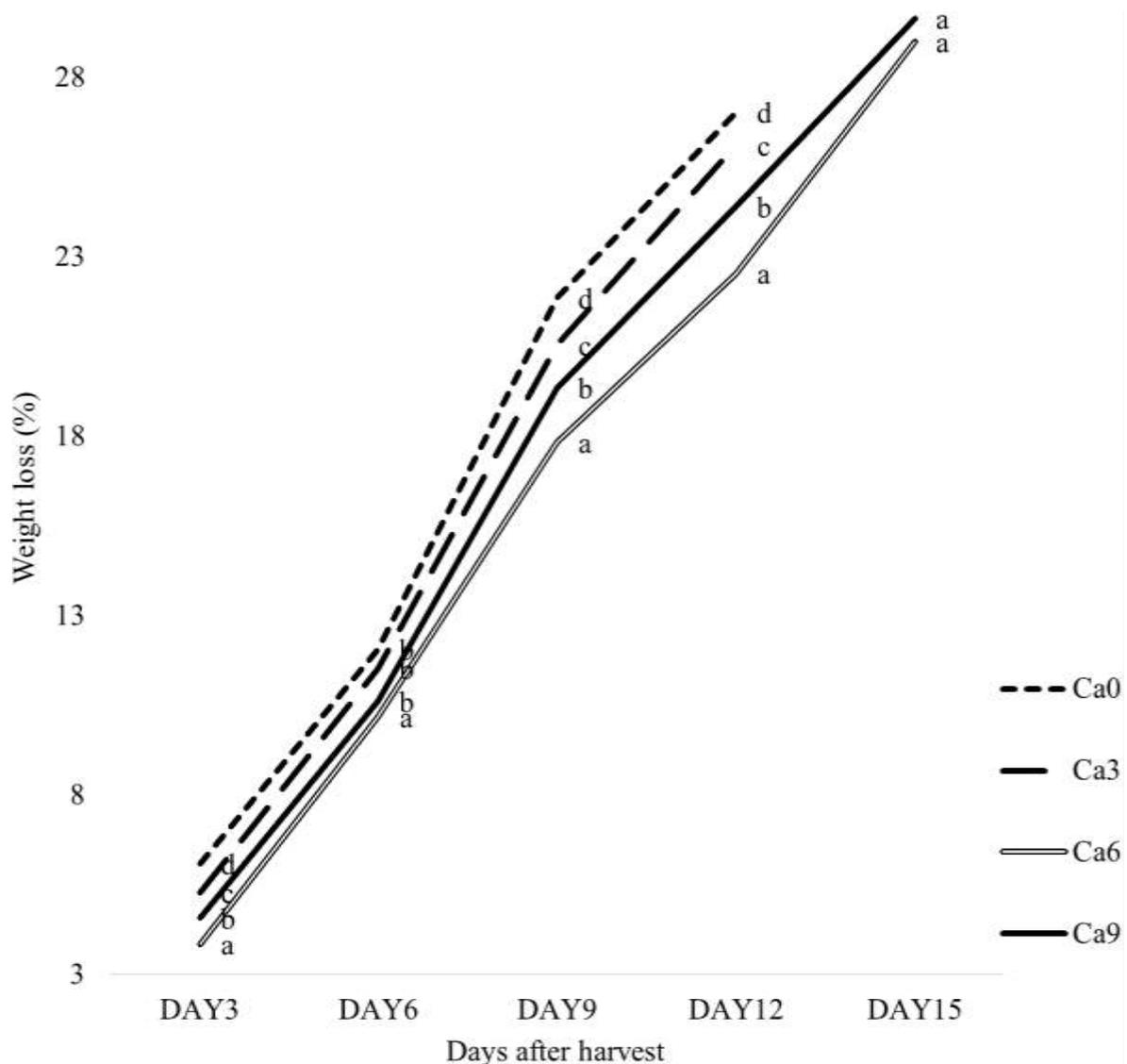
All the data were subjected to analysis of variance (ANOVA) in SAS (ver. 9.0, SAS Institute Inc., Cary, NC). Significant means at F-Test were separated using Tukey's Honestly Significant Difference (SAS Institute Inc., Cary, NC, 2010).

## RESULTS AND DISCUSSION

### Effect of Ca CaCl<sub>2</sub> dipping on fresh weight loss and shelf life of bell pepper fruit

#### Fresh weight loss

Ca CaCl<sub>2</sub> dipping had a significant effect on fresh weight loss of bell pepper from 3 to 12 DAH (days after harvest) of storage (Figure 1). At 3 DAH, the lowest weight loss of 3.9% was recorded under Ca6 treatment (fruits treated with 6% Ca CaCl<sub>2</sub> dipping), followed by weight loss of 4.6% under Ca9 treatment (fruits treated with 9% Ca CaCl<sub>2</sub> dipping) then 5.3% under Ca3 treatment (fruits treated with 3% Ca CaCl<sub>2</sub> dipping) with the highest weight loss of 6.1% recorded for fruits under Ca0 treatment/control treatment (fruits treated with 0% Ca CaCl<sub>2</sub> dipping/ distilled water) (Figure 1). A similar trend was observed at 9 and 12 DAH (Figure 1). At 6 DAH, a significantly lower weight loss of 10.2% was recorded under Ca6 treatment as compared to a higher weight loss of 12.1%, 11.5% and 10.6% recorded under Ca0, Ca3 and Ca9 treatments, respectively (Figure 1). At 15 DAH, fruits under Ca6 and Ca9 treatments recorded a weight loss of 29 and 29.6%, respectively and were not



**Figure 1.** Effect of Ca  $\text{CaCl}_2$  dipping on weight loss of bell pepper. Ca0 is fruit treated with 0% Ca  $\text{CaCl}_2$  dipping, Ca3 is fruit treated with 3% Ca  $\text{CaCl}_2$  dipping, Ca6 is fruit treated with 6% Ca  $\text{CaCl}_2$  dipping and Ca9 is fruit treated with 9% Ca  $\text{CaCl}_2$  dipping. <sup>a</sup> Means followed by the same letter at any days after harvest are not significantly different using Tukey's test at  $p \leq 0.05$ .

Source: Authors

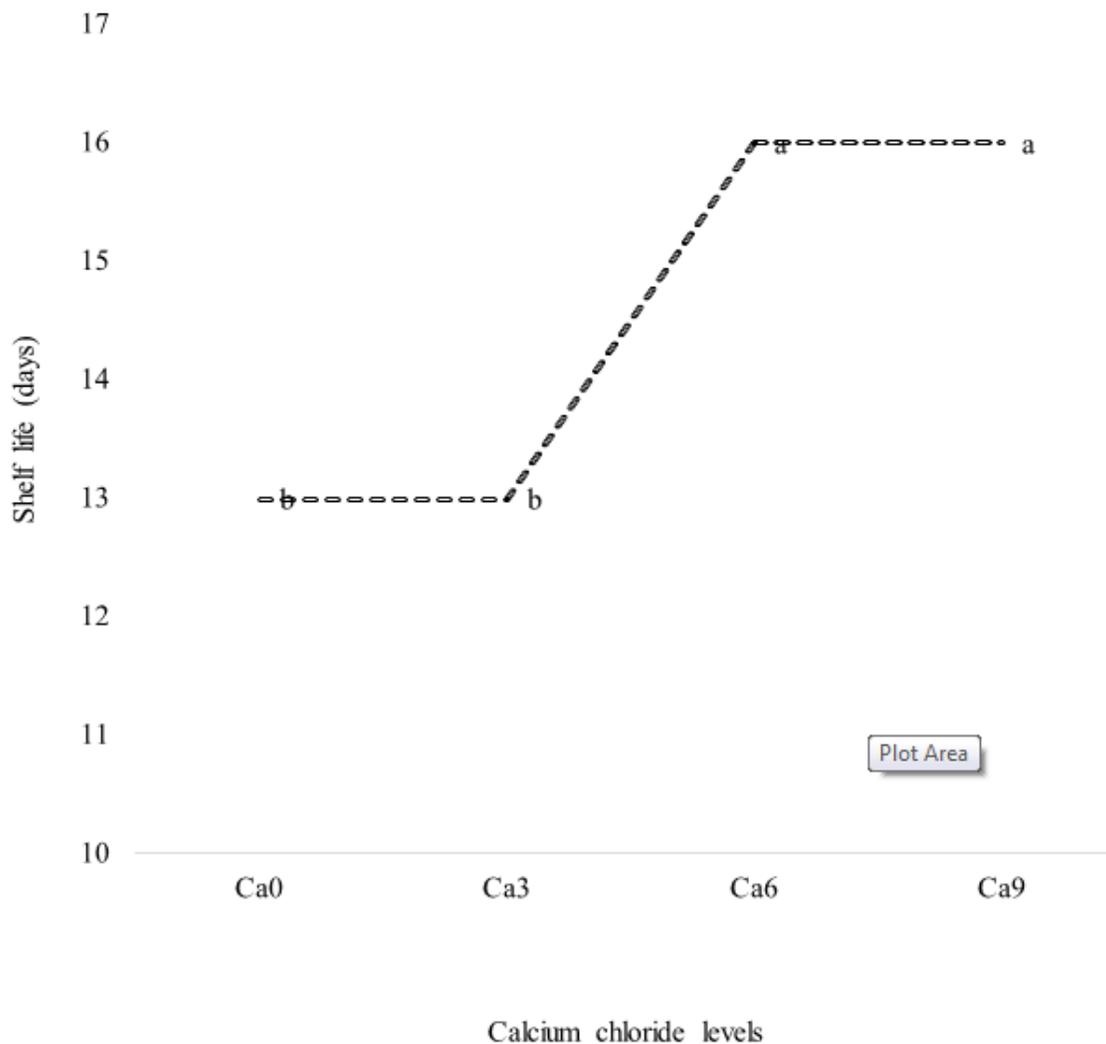
significantly different from each other (Figure 1). A general increase in weight loss was also observed as storage duration progressed regardless of the treatment applied (Figure 1)

### Shelf life

Shelf life of bell pepper fruits was significantly influenced by Ca  $\text{CaCl}_2$  dipping (Figure 2). Fruits treated with 6 and 9 % Ca  $\text{CaCl}_2$  dipping recorded a significantly longer shelf life of 16 days as compared to a shorter shelf life of 13 days recorded for fruits treated with 0 and 3% Ca

$\text{CaCl}_2$  dipping (Figure 2).

Dipping bell pepper fruits in Ca ( $\text{CaCl}_2$ ) solution reduced weight loss during storage as compared to when fruits were not dipped in  $\text{CaCl}_2$ . Fruits dipped in 6%  $\text{CaCl}_2$  recorded the lowest weight loss. Among the different  $\text{CaCl}_2$  concentrations, reduction in weight loss was observed to increase with an increase in  $\text{CaCl}_2$  concentrations. As expected, weight loss in all fruits increased as storage duration progressed regardless of the treatment applied. A fruit harvested from a plant is still alive and respiring; therefore, the stored carbohydrates are continuously used and water lost during this process leading to increased weight loss with an increase in

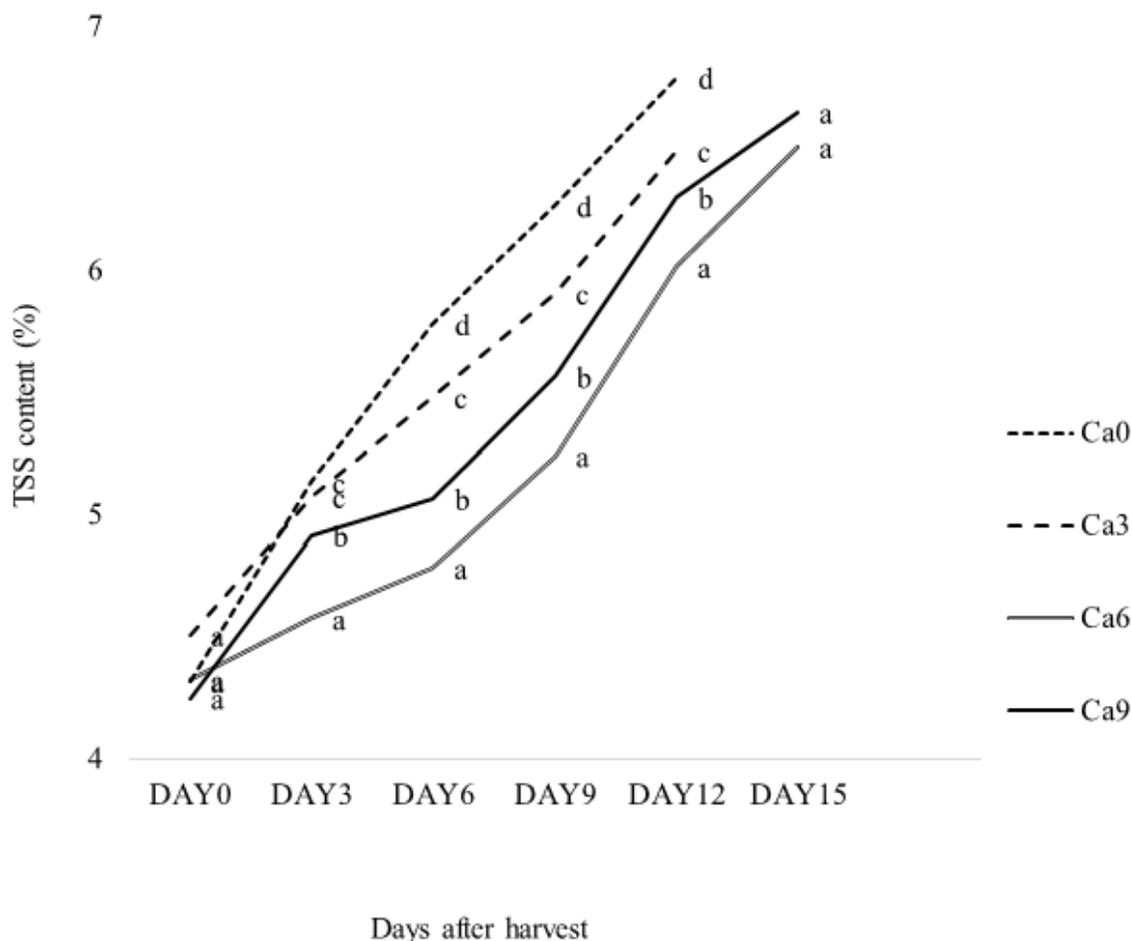


**Figure 2.** Effect of Ca  $\text{CaCl}_2$  dipping on shelf life of bell pepper. Ca0 is fruit treated with 0% Ca  $\text{CaCl}_2$  dipping, Ca3 is fruit treated with 3% Ca  $\text{CaCl}_2$  dipping, Ca6 is fruit treated with 6% Ca  $\text{CaCl}_2$  dipping and Ca9 is fruit treated with 9% Ca  $\text{CaCl}_2$  dipping. <sup>a</sup> Means followed by the same letter along the x- axis are not significantly different using Tukey's test at  $p \leq 0.05$ .

Source: Authors

storage duration. Ca  $\text{CaCl}_2$  plays a vital role in maintaining and modulating various cell functions by increasing membrane stability and cell strength (Bhatla and Lal, 2018). It also protects the membrane from degrading enzymes which enhances better linkages between pectic substances within the cell wall; thereby, increasing the cohesion of cell walls and improving the integrity and turgidity of cell wall leading to reduced water loss from the fruit (Bhatla and Lal, 2018). Ca maintains membrane functionality and integrity by lowering losses of phospholipids and proteins, reducing ion leakage, regulating enzymatic activities, thereby retarding moisture loss due to high rate of respiration and senescence (Vandana et al., 2015). This could offer explanation for the reduced weight loss in fruits treated with  $\text{CaCl}_2$

solution over the storage period observed in the current study. Akhtar et al. (2010) also reported a lower decrease in weight of loquat fruits treated with 3%  $\text{CaCl}_2$  as compared to 1%  $\text{CaCl}_2$  treated and control fruits which recorded a higher decrease in weight. A lower decrease in weight of apple fruits treated with 9%  $\text{CaCl}_2$  as compared to control fruits has also been reported (Jan et al., 2013). Jouki and Khazaei (2012) also reported a lower decrease in weight of strawberry fruits treated with 0.5%  $\text{CaCl}_2$  as compared to control fruits. Sajid et al. (2014) also observed a decrease in weight loss with increasing  $\text{CaCl}_2$  concentrations where pear fruits treated with 9%  $\text{CaCl}_2$  recorded the lowest mean weight loss while those dipped in distilled water recorded the highest mean weight loss during storage and contrary to this



**Figure 3.** Effect of Ca  $\text{CaCl}_2$  dipping on total soluble solids content of bell pepper. Ca0 is fruit treated with 0% Ca  $\text{CaCl}_2$  dipping, Ca3 is fruit treated with 3% Ca  $\text{CaCl}_2$  dipping, Ca6 is fruit treated with 6% Ca  $\text{CaCl}_2$  dipping and Ca9 is fruit treated with 9% Ca  $\text{CaCl}_2$  dipping. <sup>a</sup> Means followed by the same letter at any days after harvest are not significantly different using Tukey's test at  $p \leq 0.05$ . Source: Authors

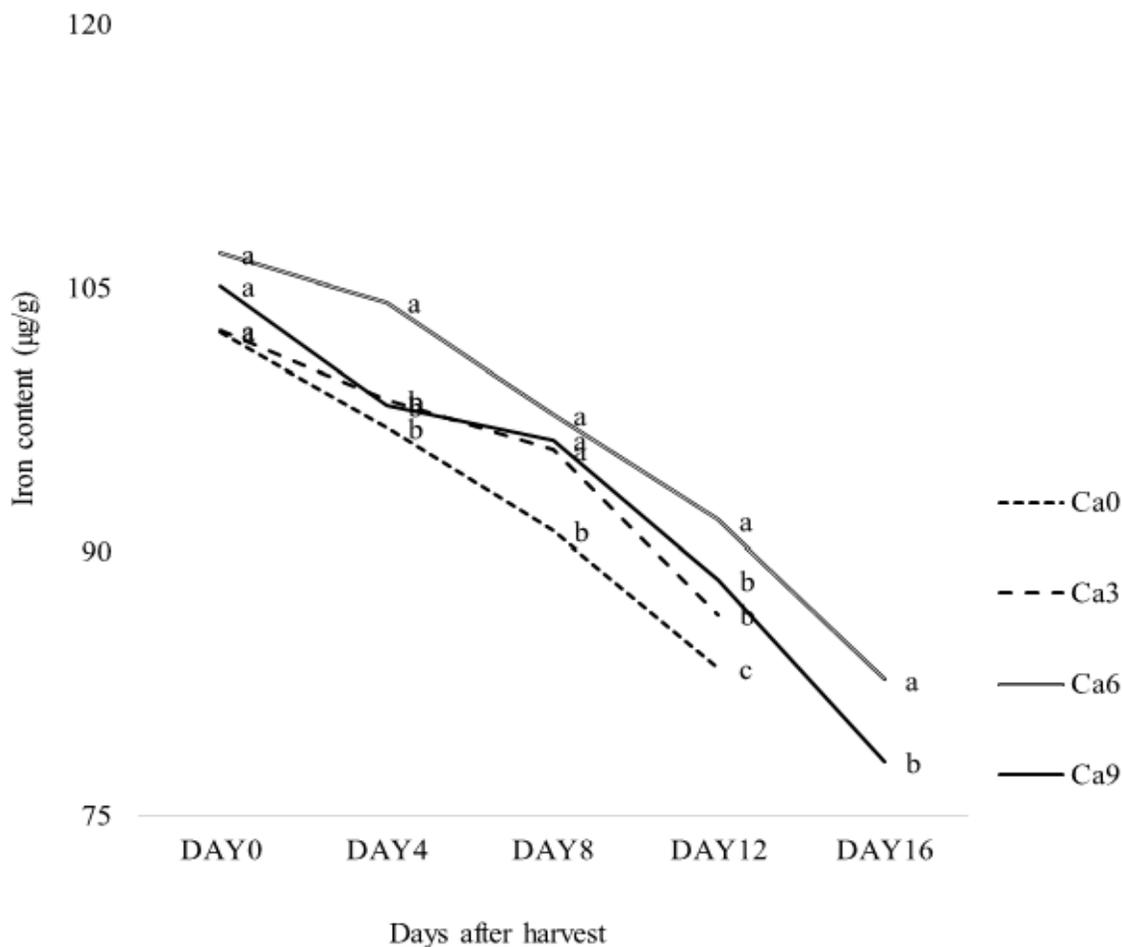
observation, fruits dipped in 9%  $\text{CaCl}_2$  (Ca9) did not record the lowest weight loss as expected in the current study. This was possibly due to formation of insoluble Ca salts (phosphates, oxalates, carbonates and sulfates) which rendered Ca unavailable for strengthening the cell wall and cytoplasmic membrane (Maathuis and Diatloff, 2013) which usually occurs when fruits are exposed to high concentration of salt for a certain duration of time (Maathuis and Diatloff, 2013).

$\text{CaCl}_2$  dipping (6%  $\text{CaCl}_2$ ) also extended the shelf life of bell pepper fruits. Control fruits and those dipped in 3%  $\text{CaCl}_2$  recorded a shorter shelf life. This could be attributed to reduced weight loss caused by  $\text{CaCl}_2$  dipping in the fruits. These results may be associated with the contribution of Ca to maintain the cellular organization and regulate enzymatic activities by protecting the membrane from degrading enzymes which enhance better linkages between pectic substances within the cell wall (Bhatla and Lal, 2018). As a result of

this, the cohesion of cell walls is increased leading to improved integrity and turgidity of cell wall thereby reducing water loss and senescence and extending shelf life (Bhatla and Lal, 2018).

#### Effect of Ca $\text{CaCl}_2$ on total soluble solids of bell pepper fruit

Ca  $\text{CaCl}_2$  dipping also had a significant effect on TSS content of bell pepper from 3 to 12 DAH of storage (Figure 3). At 3 DAH, fruits under Ca6 treatment recorded the lowest TSS content of 4.6%, followed by the TSS content of 4.9% under Ca9 treatment with the highest TSS content of 5.1% recorded under Ca0 and Ca3 treatments (Figure 3). The lowest TSS content of 4.8% was recorded under Ca6 treatment, followed by TSS content of 5.1% under Ca9 treatment, then 5.5% under Ca3 with the highest TSS content of 5.8% recorded under



**Figure 4.** Effect of Ca  $\text{CaCl}_2$  dipping on Fe content of bell pepper. Ca0 is fruit treated with 0%  $\text{CaCl}_2$  dipping, Ca3 is fruit treated with 3%  $\text{CaCl}_2$  dipping, Ca6 is fruit treated with 6% Ca  $\text{CaCl}_2$  dipping and Ca9 is fruit treated with 9%  $\text{CaCl}_2$  dipping. <sup>a</sup> Means followed by the same letter at any days after harvest are not significantly different using Tukey's test at  $p \leq 0.05$ .

Source: Authors

the control treatment at 6 DAH (Figure 3). A similar trend was observed at 9 and 12 DAH of storage (Figure 3). The TSS content of 6.5 and 6.7% under Ca6 and Ca9 treatments, respectively were not significantly different at 15 DAH (Figure 3). TSS content of all treated fruits was also observed to increase during storage (Figure 3).

Fruits dipped in 6%  $\text{CaCl}_2$  solutions recorded a lower TSS content compared to control fruits which had significantly higher TSS content during 16 days storage.  $\text{CaCl}_2$  slows alteration in cell wall structure which breakdown into simple sugars leading to reduced ripening process (Sajid et al., 2014). As a result, delayed degradation of starch by amylase to produce sugar is achieved.  $\text{O}_2$  normally accelerates the ripening process. A significantly high TSS content in control fruits can thus be attributed to increased  $\text{O}_2$  in fruits' tissues which could have increased the rate of bell pepper fruit ripening. Similarly, a decrease in fruits' TSS content with an increase in  $\text{CaCl}_2$  concentrations and an increase in TSS

content as storage duration progressed have been reported in  $\text{CaCl}_2$  treated fruits such as eggplant and courgette (Chepngeno et al., 2016) and pear (Sajid et al., 2014). According to Sajid et al. (2014), the higher TSS content observed in untreated fruits and those treated with low concentration of  $\text{CaCl}_2$  could have been due to rapid conversion of starch present in the fruits during picking into sugar during storage period.

#### Effect of Ca $\text{CaCl}_2$ on Fe content of bell pepper fruit

Fe content of bell pepper fruits was significantly influenced by  $\text{CaCl}_2$  dipping from 4 DAH through the end of storage (Figure 4). At 4 DAH, a significantly higher Fe content of 104.3  $\mu\text{g/g}$  was recorded under Ca6 treatment as compared to a lower Fe content of 97.1  $\mu\text{g/g}$ , 98.7  $\mu\text{g/g}$  and 98.3  $\mu\text{g/g}$  recorded under Ca0, Ca3 and Ca9 treatments, respectively (Figure 4). At 8 DAH, fruits under

Ca3, Ca6 and Ca9 treatments recorded a significantly higher Fe content of 91.3 µg/g, 95.8 and 96.4 µg/g, respectively compared to a lower Fe content of 97.8 µg/g recorded under the control treatment (Figure 4). At 12 DAH, the highest Fe content of 91.8 µg/g was recorded under Ca6 treatment, followed by Fe content of 86.5 µg/g and 88.4 µg/g under Ca3 and Ca9 treatments, respectively with the lowest Fe content of 83.4 µg/g recorded under Ca0 treatment (Figure 4). Fruits treated with 6% CaCl<sub>2</sub> dipping recorded a significantly higher Fe content of 82.8 µg/g as compared to a lower Fe content of 78.1 µg/g, recorded for fruits treated with distilled water at 16 DAH (Figure 4). A general decrease in Fe content with an increase in storage duration was also recorded (Figure 4).

Dipping bell pepper in 6% CaCl<sub>2</sub> also slowed the rate of decline in Fe content of fruits. A higher rate of decline was observed in control fruits that were dipped in distilled water only. Ca alters intracellular and extracellular processes that retard respiration by reducing the activities of oxidizing enzymes within the tissues of a fruit (Xu et al., 2014). Ca reduces the level of reactive oxygen species (ROS) which include; the superoxide anion radical (O<sub>2</sub><sup>-</sup>), H<sub>2</sub>O<sub>2</sub>, HOCl, (OH<sup>-</sup>), and (O<sub>2</sub>). Reactive oxygen species (ROS) are formed through endogenous enzymatic and non-enzymatic reactions within the cell and within the mitochondria. The activities of antioxidant enzymes such as glutathione, glutathione peroxidase, glutathione transferases, catalase and superoxide dismutase are therefore increased in the mitochondria to reduce ROS (Sheu et al., 2006). These antioxidant enzymes keep the concentrations on ROS in check. Fe ions are responsible for the synthesis of many proteins (ferredoxin and cytochromes) that carry electrons during respiration (Bhatla and Lal, 2018). The low Fe ions concentration observed in fruits dipped in distilled water could thus have been as a result of increased respiration rates in control fruits in which most of the ions were used to synthesize the proteins to carry electrons. Higher Fe content in Ca treated fruits on the other hand could have been as a result of increased activities of antioxidant enzymes favoured by increased levels of Ca ion in cells which could have led to reduced ROS thereby reducing the rate of respiration and ultimately the amount of Fe ions used in the synthesis of proteins responsible for electron transport during respiration (Bhatla and Lal, 2018).

#### **Effect of Ca CaCl<sub>2</sub> on total chlorophyll content of bell pepper fruit**

There were significant differences on total chlorophyll content of bell pepper fruits that were treated with CaCl<sub>2</sub> dipping from 4 DAH through the end of storage (Figure 5). Fruits under Ca6 and Ca9 treatments recorded significantly higher total chlorophyll content of 427µg/g

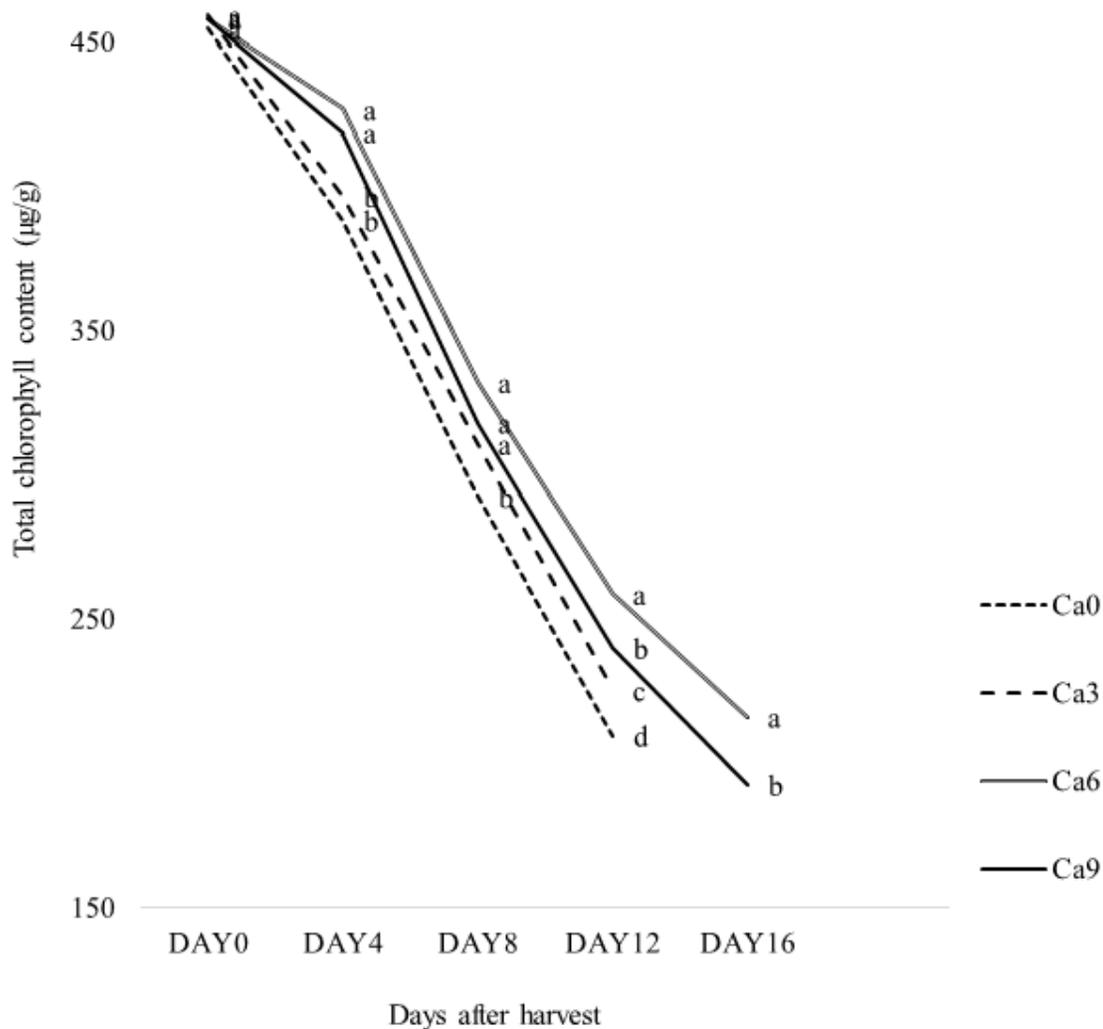
and 418.9 µg/g, respectively as compared to lower total chlorophyll content of 388.6 µg/g and 396.8 µg/g, respectively recorded under Ca0 and Ca3 treatments at 4 DAH (Figure 5). At 8 DAH, fruits treated with 3, 6 and 9% CaCl<sub>2</sub> dipping recorded a significantly higher total chlorophyll content of 311.1 µg/g, 332.4 µg/g and 318.4 µg/g, respectively as compared to a lower total chlorophyll content of 292.7 µg/g recorded for fruits treated with pure distilled water (Figure 5). At 12 DAH, the highest total chlorophyll content of 259 µg/g was recorded under Ca6 treatment, followed by 240.3 µg/g under Ca9 treatment, then 225.7 µg/g under Ca3 treatment with the lowest total chlorophyll content of 210 µg/g, recorded under the control treatment (Figure 5). Fruit treated with Ca6 treatment recorded a significantly higher total chlorophyll content of 216.5 µg/g as compared to a lower total chlorophyll content of 192.9 µg/g recorded under Ca9 treatment at 16 DAH (Figure 5). A decline in total chlorophyll content in bell pepper fruits as storage progressed in all treated fruits was also recorded (Figure 5).

CaCl<sub>2</sub> dipping was also found to delay total chlorophyll breakdown in bell pepper fruits. At the end of storage, fruits treated with 6% CaCl<sub>2</sub> (Ca6) had the highest total chlorophyll content. In fruits that were dipped in distilled water, a rapid loss of total chlorophyll was observed. Ca inhibits the activity of both 1-aminoacyclopropane 1-carboxylate synthase (ACC synthase) and ACC oxidase (ACO) leading to reduced respiration, ripening and total chlorophyll degradation (Zhang et al., 2019). The rapid degradation of total chlorophyll observed in fruits dipped in distilled water (control) observed in this study could have been due to increased rate of respiration and ripening caused by increased O<sub>2</sub> concentrations in fruits' tissues.

#### **Effect of Ca CaCl<sub>2</sub> dipping on ascorbic acid content of bell pepper fruit**

Ascorbic acid content in bell pepper fruit was influenced by CaCl<sub>2</sub> dipping during storage from 4 DAH through 12 DAH (Figure 6). At 4 DAH, fruits treated with 6% CaCl<sub>2</sub> dipping recorded a significantly higher ascorbic acid content of 24mg/100g, followed by ascorbic acid content of 22.5mg/100g recorded under Ca3 and Ca9 treatments with the lowest ascorbic acid content of 21.7mg/100g recorded under the control treatment (Figure 6). A similar trend was observed at 8 and 12 DAH (Figure 6). At 16 DAH, ascorbic acid content of 20.4mg/100g recorded under Ca6 was not significantly different from 20.2mg/100g ascorbic acid content recorded under Ca9 treatment (Figure 6). Ascorbic acid content of fruits decreased with an increase in storage duration in all treated fruits (Figure 6).

The highest content of ascorbic acid content in fruits was achieved with 6% CaCl<sub>2</sub> dipping. Control fruits on the



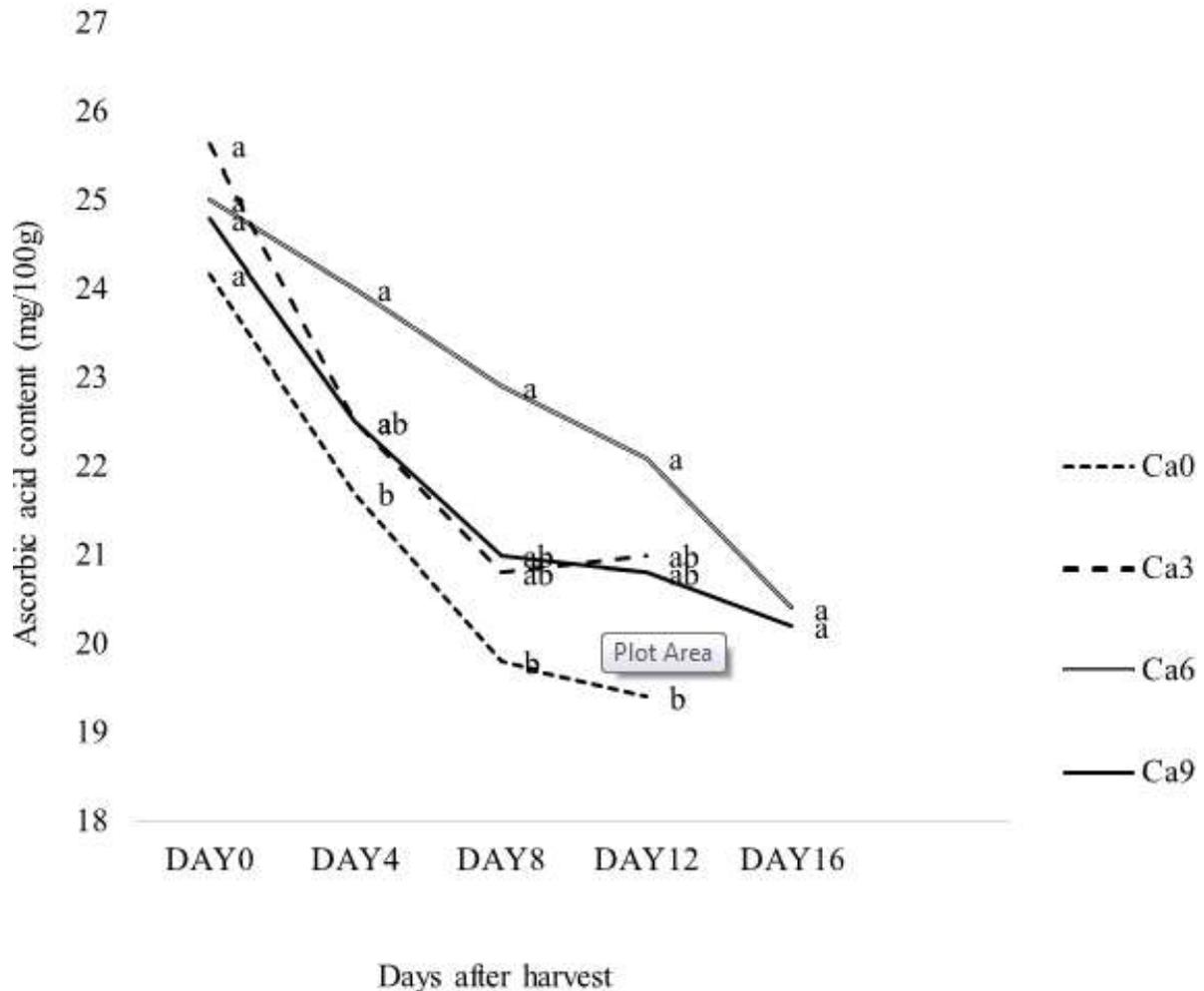
**Figure 5.** Effect of Ca  $\text{CaCl}_2$  dipping on total chlorophyll content of bell pepper. Ca0 is fruit treated with 0%  $\text{CaCl}_2$  dipping, Ca3 is fruit treated with 3%  $\text{CaCl}_2$  dipping, Ca6 is fruit treated with 6%  $\text{CaCl}_2$  dipping and Ca9 is fruit treated with 9%  $\text{CaCl}_2$  dipping. <sup>a</sup> Means followed by the same letter at any days after harvest are not significantly different using Tukey's test at  $p \leq 0.05$ .  
Source: Authors

other hand exhibited the lowest ascorbic acid content during 16 days storage. A general trend of decrease in ascorbic acid content with increasing storage duration in fruits subjected to all  $\text{CaCl}_2$  treatments was also observed in this study. During storage, oxidizing enzymes like ascorbic acid oxidase, peroxidase, catalase and polyphenol oxidase might be causing decrease in ascorbic acid content of fruits. Ca ions are responsible for the reduction in activities of oxidizing enzymes (Goutam et al., 2010) which reduces the rate of oxidation of ascorbic acid in fruit tissues. This could possibly explain the gradual decrease of ascorbic acid in fruits treated with  $\text{CaCl}_2$  as storage progressed as well as the high ascorbic acid content in the same fruits at the end of storage. Control fruits exhibited a rapid decline in ascorbic acid content with increasing storage duration

due to increased activities of oxidizing enzymes which led to rapid degradation of ascorbic acid through increased oxidation. Decrease in ascorbic acid content and a higher ascorbic acid content in fruits with increasing storage duration and  $\text{CaCl}_2$  treatment has also been observed in other vegetables such as tomato, carrot, eggplant and courgette (Chepngeno et al., 2016; Sajid et al., 2014; Akhtar et al., 2010).

#### Effect of Ca $\text{CaCl}_2$ dipping on calcium content of bell pepper fruit

Calcium chloride dipping had a significant effect on the calcium content of bell pepper fruit from 4 DAH to 16 DAH (Figure 7). At 4 DAH, a significantly higher calcium

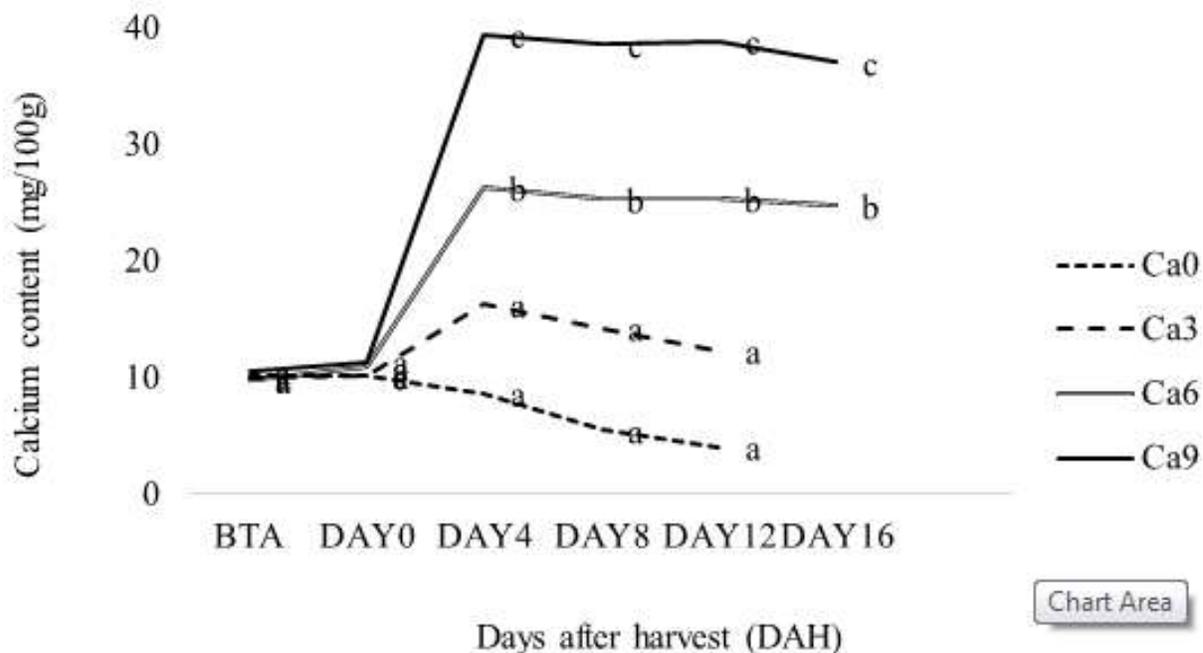


**Figure 6.** Effect of Ca  $\text{CaCl}_2$  dipping on ascorbic acid content of bell pepper. Ca0 is fruit treated with 0%  $\text{CaCl}_2$  dipping, Ca3 is fruit treated with 3%  $\text{CaCl}_2$  dipping, Ca6 is fruit treated with 6%  $\text{CaCl}_2$  dipping and Ca9 is fruit treated with 9%  $\text{CaCl}_2$  dipping. <sup>a</sup> Means followed by the same letter at any days after harvest are not significantly different using Tukey's test at  $p \leq 0.05$ .  
Source: Authors

content of 39.45 mg/100g was recorded under Ca9 treatment followed by calcium content of 26.3mg/100g recorded under Ca6 treatment with the lowest calcium content of 8.7 mg/100g and 14.3 mg/100 g recorded under Ca0 and Ca3 treatments, respectively (Figure 7). A similar trend was observed at 8 and 12 DAH. At 16 DAH, fruits under Ca9 treatment recorded a significantly higher calcium content of 37.03 mg/100g as compared to a lower calcium content of 24.76 mg/100g recorded under Ca6 treatment (Figure 7).

Calcium chloride dipping resulted in a higher Ca content in fruits. Fruits dipped in 9%  $\text{CaCl}_2$  had the highest Ca content during storage. The control treatment (fruits dipped in distilled water) had the lowest Ca content at the end of storage period. Calcium content of bell pepper fruits increased significantly with an increase in

$\text{CaCl}_2$  concentrations; and this was observed 4 days after dipping treatment. Thereafter, Ca content in fruits decreased with increase in storage duration though the decrease was not significant. Calcium treatment increases the amount of  $\text{Ca}^{2+}$  in the central vacuoles, Endoplasmic Reticulum (ER), mitochondria and cell walls where it is stored as calcium pectate (Bhatla and Lal, 2018). This explains the increase in  $\text{Ca}^{2+}$  content in the fruits after the fruits were dipped in calcium chloride. Calcium pectate is an essential constituent of plant cell wall and it helps in joining the cell together. It is also used for transport and retention of other nutrients. It is required for normal functioning of the membrane, where it binds phospholipids and membrane proteins. To carry out all these, tissues require enough calcium pectate and this could explain the decrease in  $\text{Ca}^{2+}$  content in fruits as



**Figure 7.** Effect of Ca  $\text{CaCl}_2$  dipping on calcium content of bell pepper. BTA is before treatment application. Ca0 is fruit treated with 0%  $\text{CaCl}_2$  dipping, Ca3 is fruit treated with 3%  $\text{CaCl}_2$  dipping, Ca6 is fruit treated with 6%  $\text{CaCl}_2$  dipping and Ca9 is fruit treated with 9%  $\text{CaCl}_2$  dipping. <sup>a</sup> Means followed by the same letter at any days after harvest are not significantly different using Tukey's test at  $p \leq 0.05$ .  
Source: Authors

storage duration progressed. Other researchers have also reported higher Ca content in tissues of fruits treated with higher concentration of  $\text{CaCl}_2$  during storage (Jan et al., 2013; Senevirathna and Daundasekera, 2010).

## Conclusion

Calcium  $\text{CaCl}_2$  dipping significantly influenced postharvest quality and shelf life of bell pepper. Six percent  $\text{CaCl}_2$  dipping was the best treatment in terms of fresh weight loss reduction, maintenance of total soluble solids, Fe, total chlorophyll, ascorbic acid and Ca content and extension of shelf life of bell pepper fruit.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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