Current Applied Science and Technology Vol. 21 No. 3 (July-September 2021)

Efficient Comparison of Calcium Chloride and Calcium Gluconate Immersions on Quality Maintenance and Bioactive Compounds of Ready-to-cook Baby Corns

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Received: 7 October 2020, Revised: 16 December 2020, Accepted: 2 February 2021

Abstract

The efficiency of calcium chloride ($CaCl_2$) or calcium gluconate (Ca-Glu) immersion on physicochemical quality and bioactive compounds of ready-to-cook baby corns during cold storage at 4 ± 1ºC for 7 d was investigated. Baby corns were immersed in 1% (w/v) $CaCl_2$, 1% (w/v) Ca-Glu or distilled water (control) for 1 min. The biological parameters such as superficial colour attributes, weight loss, texture, pectin fractions, antioxidant activities and bioactive compounds of baby corns were determined. Both calcium immersions did not affect superficial the colour attributes of the baby corn compared to the control sample during storage. Baby corn texture was maintained by calcium immersions, with $CaCl_2$ evidently better maintaining texture compared to Ca-Glu. The texture maintenance by calcium immersions was associated with the retardation of increased EDTA-soluble pectin content and decreased Na_2CO_3-soluble pectin content. Ca-Glu immersion exhibited the enhancement of antioxidant activity and the concentrations of total phenols and ascorbic acid as well as the retention of free radical scavenging activity and flavonoid content during storage. In conclusion, 1% Ca-Glu immersion is a feasible alternative for maintaining texture and enhanced nutritional value of ready-to-cook baby corns during storage.

Keywords: baby corn; calcium immersion; texture; bioactive compound
DOI 10.14456/cast.2021.40

1. Introduction

Baby corn, typically recognised as immature ear corn, (*Zea mays* L.), is a well-known and important commercial crop. Typically, baby corn is harvested after silk become visible for 1 to 3 days and the cob size is approximately 4.5 cm to 10 cm in length and 7 mm to 17 mm in diameter [1]. Baby corn contains high contents of soluble and reducing sugars and is of high nutritional values as it contains proteins, vitamins, dietary fibre and minerals, especially iron and phosphorous [2]. Baby corn is utilized commercially in both fresh and processed forms. For fresh consumption, most baby corns

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are produced from sweet corn seeds because these offer better flavour and taste than field corn seeds [1]. In Thailand, baby corn in the form of a fresh-cut vegetable, is an important vegetable product produced for export as well as domestic markets [3]. It is commonly acknowledged that baby corn is very perishable due to its high metabolic rate and moisture loss [4, 5]. Three main problems that affect the marketable quality of fresh baby corn are the loss of crispness and sweetness and the incidence of tip-browning [3, 6]. These problems are caused by high respiration rate, ear desiccation and enzymatic browning activity [3, 4]. Previous work reported that shrink wrapping and cold storage delayed senescence and browning incidence and maintained desirable appearance, texture and flavour due to the suppression of respiration rate and the prevention of moisture loss [7]. Attia *et al*. [4] reported that 1% $CaCl_2$ dip followed by wrapping with polypropylene film could extend the pleasant fresh-liked quality of baby corns stored at 5ºC due to the inhibition of browning and moisture loss and maintenance of total sugars content. Exogenous calcium application is accepted as an effective approach for maintaining physicochemical quality and prolonging shelf-life, and for delaying disease attack and senescence of postharvest fruit and vegetables [8, 9]. Calcium can also preserve texture by maintaining cell wall structure and osmotic tonicity and by reducing membrane lipid dysfunction [10]. In commercial, $CaCl_2$ has been widely used for postharvest commodities; however, it might impart undesirable bitterness to products, affecting organoleptic quality. The application of other calcium salts had been suggested by Labin-Goldscher and Edenstein [11] that calcium lactate (Ca-Lac), calcium citrate and calcium gluconate (Ca-Glu) do not provide bitter taste and enhance nutritional value. Varela *et al*. [12] reported that 1% $CaCl_2$ immersion provided a slight bitter taste in fresh-cut apple but there was no effect on the organoleptic evaluation by trained panels. Youryon *et al*. [13] reported that exogenous postharvest application of Ca-Glu could induce the formation of antioxidants and retard membrane lipid peroxidation in 'Queen' pineapple better than $CaCl_2$ application during storage at 13ºC. However, the comparison of the effects of the application of various calcium salts on physicochemical quality of ready-to-cook baby corns has not been recently studied. Thus, the aim of this study was to compare the efficiency of $CaCl_2$ and Ca-Glu immersions on physicochemical quality of ready-to-cook baby corns during short-term storage at 4 ± 1°C.

2. Materials and Methods

2.1 Raw materials and treatments

Baby corns cv. 'Pacific' were derived from a commercial corn plot at Kampangsan District, Nakhon Pathom Province, Thailand. The baby corns were harvested at 47-49 days after planting (DAP) when the length of silk was approximately 3-5 cm. The baby corns were delivered to the laboratory at Department of Agricultural Education, King Mongkut's Institute of Technology Ladkrabang (KMITL), Bangkok, within 2 h., with control of the inside temperature of container below 10°C. The baby corns were peeled and cleaned by rinsing with potable water and dipped in 100 µl l^{-1} sodium hypochlorite for 2 min. After that, the baby corns were immersed into water (control), 1% (w/v) calcium chloride ($CaCl_2$) or 1% (w/v) calcium gluconate (Ca-Glu) for 1 min. Five ears of baby corns were packed in a foam tray (83 x 135 x 15 mm in dimension) and wrapped with Linear Low-Density Polyethylene (LLDPE) film (12.7 µm thickness). The ready-to-cook baby corns were stored at 4 ± 1°C for 7 days. Physicochemical quality attributes such as visual appearance, superficial colours, weight loss, texture, pectin substances, antioxidant activities and bioactive compounds were determined during storage.

2.2 Colour, weight loss and texture measurements

Colour, weight loss and texture of the baby corns were determined at the beginning of storage (day 0) until day 7. Colour attributes were measured using a Minolta colorimeter, CR-400 (Minolta, Japan). Lightness (L^*), yellowness (b^*), hue and chroma values were recorded every day during storage. The weight of ready-to-cook baby corns during storage period was recorded daily. The percentage of fresh weight loss was calculated by comparison with the weight on initial day. The texture of the baby corns was measured using a Texture Analyser, EZ-SX (Stable Micro Systems, USA). A cutting blade was used for texture measurement (hardness) and the compression force as Newton (N) was presented. The blade was driven at speed of 0.5 mm s^{-1} to a depth of 5 mm.

2.3 Pectin substances assays

Acetone insoluble solid (AIS) of the baby corns was prepared following the procedure described by Supapvanich and Tucker [14]. The AIS was extracted with 50 mM ethylenediamine tetraacetic acid (EDTA) in 50 mM sodium acetate, pH 6.5 solution at room temperature for 12 h and then filtered through a GF/A filter paper. The filtrate was collected and the EDTA-soluble pectin fraction (soluble pectin) was precipitated using absolute ethanol. The pallet was again extracted with 50 mM sodium carbonate (Na_2CO_3) in 50 mM sodium acetate, pH 6.5, solution at 4°C for 24 h followed by ambient temperature for 2 h. The suspension was then filtered through a GF/A filter paper and the Na_2CO_3- soluble pectin fraction (insoluble pectin) in the filtrate was precipitated using absolute ethanol. Both the EDTA-soluble and Na_2CO_3- soluble pectin fractions were hydrolysed with 1 M H_2SO_4 at 95°C. Galacturonic acid content of both pectin fractions was determined using the procedure of Ahmed and Labavitch [15]. Data were expressed as mg galacturonic acid per kg (mg kg^{-1}).

2.4 Antioxidant activities

A 10 g of baby corn was extracted with 60% (v/v) ethanol solution. The extract was used to determine antioxidant activities and the concentrations of total phenols and flavonoids. Ferric reducing antioxidant potential (FRAP value) and DPPH radical scavenging activity were determined according to the procedures of Benzie and Strain [16]. FRAP value was calculated using a linear equation derived from a Trolox standard curve and presented as mmole Trolox equivalent per kg sample (mmol TE kg^{-1}). DPPH radical scavenging activity was assayed using the method of Brand-Williams *et al*. [17]. The percentage of decreased optical density (OD) at 517 nm wavelength was calculated and presented as the percentage of free radical scavenging activity (%).

2.5 Total phenols, flavonoids and total ascorbic acid assays

Total phenols concentration was determined using the method of Slinkard and Singleton [18]. The concentration of total phenols was calculated using a linear equation of gallic acid standard curve and presented as mg gallic acid equivalents per kg (mg kg^{-1}). The concentration of flavonoids was determined using the procedure according to Jia *et al*. [19]. Flavonoids concentration was reckoned using a linear equation derived from catechin standard curve. The data were expressed as mg catechin equivalents per kg fresh weight of fruit (mg kg^{-1}). Ascorbic acid in the samples was extracted using 5% cold metaphosphoric acid. The concentration of total ascorbic acid (AsA) was assayed using the procedure of Hashimoto and Yamafuji [20]. The data were expressed as mg ascorbic acid per kg fresh weight (mg kg^{-1}).

2.6 Statistical Analysis

The experiments were performed using a completely randomized design (CRD). The data were analysed using analysis of variance (ANOVA) and the Duncan's Multiple Range Test (DMRT) at $P \leq 0.05$. All data were presented as the mean of four replications (n = 4) ± standard deviation (SD).

3. Results and Discussions

3.1 Superficial colour attributes

Superficial colour including *L**, *b**, hue and chroma value of the ready-to-cook baby corns are presented in Figure 1. Superficial colours of all treatments seemed to remain constant over the storage. The averages of *L**, *b**, hue and chroma values were approximately 76.5, 32, 92.5 and 31.9, respectively. Moreover, we did not find undesirable visual appearance of the baby corns over the storage (data not shown). These results suggest that 1% $CaCl_2$ and 1% Ca-Glu immersions did not affect superficial colour of the baby corns during refrigerated storage for 7 days. It is commonly recognised that undesirable visual appearance of ready-to-cook baby corns involving discoloration is mainly caused by moisture loss during storage [21]. In this study, the change in superficial colour of the baby corns was protected by the LLDPE film package, which is recommended for fresh commodities because its good water vapour barrier properties reduce moisture loss [22].

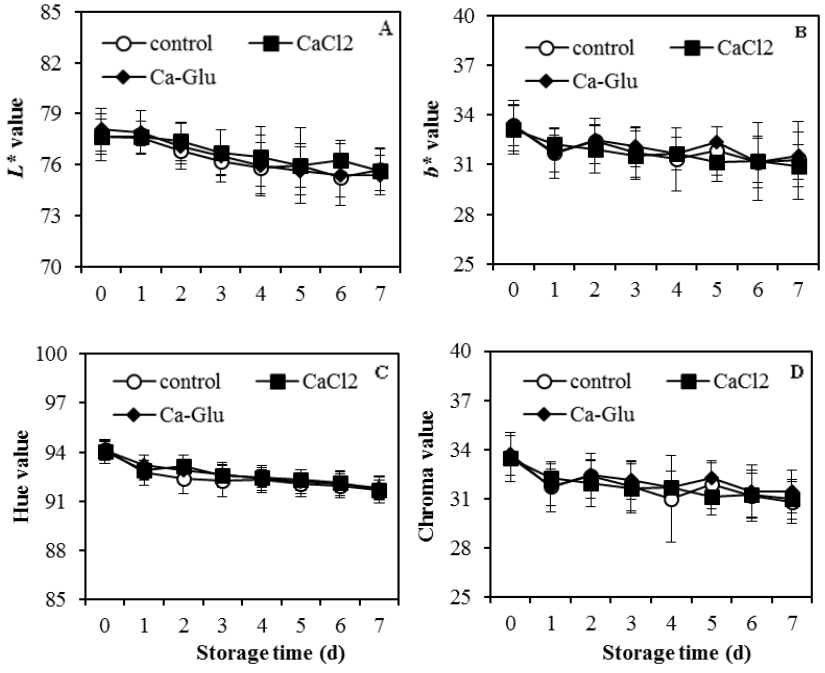

Figure 1. Colour attributes including *L** (A), *b** (B), hue (C) and chroma (D) values of baby corns treated with $CaCl_2$ and Ca-Glu compared with control samples during storage. Data are presented as mean (n = 4) with SD bar.

3.2 Weight loss and texture

Weight loss and texture change of the baby corns during storage are presented in Figure 2. It was found that an increase in weight loss was observed in all treatments during storage. The weight loss of control samples was significantly higher than that of all calcium treated samples within the first 3 days of storage period ($P < 0.05$). This might be associated with the maintenance of cell membrane function by Ca^{+} as described by Lester and Grusak [8] and Youryon *et al*. [13]. After 3 d of storage, the increased weight loss of all treatments was not significantly different over the storage ($P < 0.05$) (Figure 2A). At the end of storage, the moisture loss of all treatments was approximately 7.5%. The texture of the baby corns was measured using cutting force. Both of the calcium salts immersions could maintain texture being evidently greater than the control samples (Figure 2B). The cutting force value of control samples continuously decreased and was significantly lower than that of $CaCl_2$ and Ca-Glu treated baby corns ($P < 0.05$). $CaCl_2$ immersion maintained the texture of the baby corns better than Ca-Glu immersion. The average cutting force value of control, $CaCl_2$ and Ca-Glu treated baby corns for 7 days storage was 15.60, 16.98 and 16.29 N, respectively. The recent results revealed that both $CaCl_2$ and Ca-Glu immersions might not obviously affect the loss of fresh weight but they maintained the texture of the baby corns during storage, and especially in the case of $CaCl_2$ immersion. The loss of moisture is recognised as a main factor affecting texture change of fresh commodities [23]. The result showed that the increased weight loss during storage might not be the main factor affecting the loss of texture when the baby corns were packed in a package protecting the loss of moisture. It is commonly acknowledged that exogenous calcium application prevents the softening process of postharvest commodities due to the formation of calcium pectate structure. Calcium pectate enhances cell wall strengthening and furthermore it is not a substrate of polygalacturonase (PG), a cell wall hydrolase [23]. The recent results, the decreased weight loss of control baby corns during the first 3 days of storage was concomitant with the reduction of cutting force. Meanwhile, the weight loss of both $CaCl_2$ and Ca-Glu treated baby corns was evidently lower than that of control samples and their cutting force values were higher than control samples. However, after the third day of storage, the weight loss of all treatments was similar, which might be due to the moisture equilibrium in the package. The different effects of $CaCl_2$ and Ca-Glu immersion on texture maintenance of the baby corns might be associated with their soluble properties; $CaCl_2$ provides a higher elemental calcium content in solution than does Ca-Glu [24, 25].

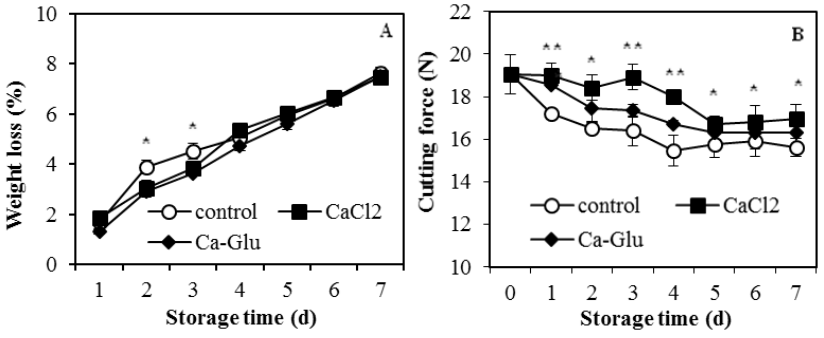

Figure 2. Weight loss (A) and texture (cutting force) (B) of ready-to-cook baby corn treated with $CaCl_2$ and Ca-Glu compared with control samples during cold storage. Data are presented as mean (n = 4) with SD bar. Significant differences between treatments are indicated with asterisks [** ($P < 0.01$); * ($P < 0.05$)].

3.3 Pectin substances

Table 1 shows the concentration of EDTA-soluble and Na_2CO_3- soluble pectin fractions of the baby corns after storage for 7 days. The EDTA-soluble pectin fraction of control samples was markedly increased whilst Na_2CO_3-soluble pectin fraction was obviously decreased when compared to those on day 0 of storage. Both calcium salt immersions delayed an increase of the EDTA-soluble and a decrease of the Na_2CO_3-soluble pectin fractions of the baby corns. $CaCl_2$ immersion showed the best result in preventing the increase of the EDTA-soluble pectin fraction as well as the decrease of the Na_2CO_3-soluble pectin fraction of the baby corns compared to Ca-Glu immersion. Changes in both the EDTA-soluble and Na_2CO_3-soluble pectin fractions were concomitant with texture changes of ready-to-cook baby corns during storage (Figure 2B). Decreased cutting force during storage was accompanied by increased EDTA-soluble pectin fraction and decreased Na_2CO_3-soluble pectin fraction. Theoretically, exogenous calcium application inhibits cell wall polymerization by the binding of Ca^{2+} with demethylated polygalacturonic backbone of pectin to form calcium pectate [23, 25]. We found that the cutting force value of $CaCl_2$ treated baby corns was evidently higher than that of Ca-Glu treated samples (Figure 2B), which was concomitant with the amounts of EDTA-soluble and Na_2CO_3-soluble pectin fractions. As described above, solubility of calcium salts is related to the elemental calcium concentration in the solution, which $CaCl_2$ provides a higher elemental calcium concentration than Ca-Glu [24, 26]. This might influence the absorption of Ca^{2+} during immersion period. Although the Ca^{2+} content in baby corn tissue was not determined, the results of texture and pectin fractions could confirm that the solubility of calcium salt influenced the texture retention of the baby corns during storage.

Table 1. The concentration of pectin substances of ready-to-cook baby corn treated with $CaCl_2$ and Ca-Glu compared with control samples during cold storage

Treatment	Storage time (d)	EDTA-soluble pectin (mg kg^{-1})	Na_2CO_3-soluble pectin (mg kg^{-1})
Control	0	190.05 ± 29.77 ^{c}	100.89 ± 8.09 ^{c}
$CaCl_2$	0	170.50 ± 24.17 ^{c}	140.84 ± 15.81^{a}
Ca-Glu	0	173.30 ± 15.03 ^{c}	118.41 ± 10.50 ^{b}
Control	7	252.84 ± 20.92 ^{a}	52.84 ± 2.40 ^{e}
$CaCl_2$	7	172.84 ± 22.56 ^{c}	110.28 ± 18.33^{bc}
Ca-Glu	7	224.50 ± 19.99 ^{b}	71.21 ± 10.64 ^{d}

Data are means (n) ± SD. Different letters in each pectin substance represent a significant difference among treatments at $P < 0.05$.

3.4 Antioxidant activities

The effects of calcium salt treatments on antioxidant activities including FRAP and DPPH radical scavenging activity are shown in Figure 3. During 3 days of storage, the decrease in FRAP was

observed for all treatments. After that, an increase in the FRAP of all treatments was observed and reached its highest value on the fifth day of storage. Ca-Glu treatments obviously enhanced FRAP more than $CaCl_2$ and control treatments. At the end of storage (day 7), the FRAP of all treatments markedly declined. However, we found that the FRAP of Ca-Glu treated baby corns was significantly higher than that of control samples ($P < 0.05$) whilst the lowest FRAP value was found in control samples. The increase in FRAP on day 5 of storage might have been related to the simulation of defence mechanism caused by chilling temperature and calcium treatment, whereas the decrease in FRAP on day 7 of storage might have been associated with deterioration process of the baby corn. The DPPH radical scavenging activity of control samples markedly declined during the storage period (Fig 3B). $CaCl_2$ immersion slightly induced DPPH radical scavenging activity during 3 days of storage and markedly declined afterwards. Meanwhile, Ca-Glu immersion could maintain DPPH radical scavenging activity during 5 days of the storage and then declined. At the end of storage, DPPH radical scavenging activity of Ca-Glu treated baby corns was significantly higher than that of other samples ($P < 0.05$). These results suggested that Ca-Glu immersion could enhance antioxidant activities in the baby corns rather than $CaCl_2$ immersion. Previous works reported that calcium application could induce antioxidant activity in plants [27]. Aghdam *et al*. [28] suggested that $CaCl_2$ stimulated antioxidant system in cornelian cherry fruits due to the inducement of phenylpropanoid-flavonoids pathways. Youryon *et al.* [13] reported that Ca-Glu treatment enhanced antioxidant system including antioxidant enzyme activities in 'Queen' pineapples more than $CaCl_2$ treatment. Among the calcium salt treatments in the present study, Ca-Glu immersion was more likely to enhance as well as maintain antioxidant activities of the baby corns than $CaCl_2$ immersion. However, the mechanism of Ca-Glu enhance DPPH scavenging activity is more superior than $CaCl_2$ treatment which is ambiguous and further studies are needed.

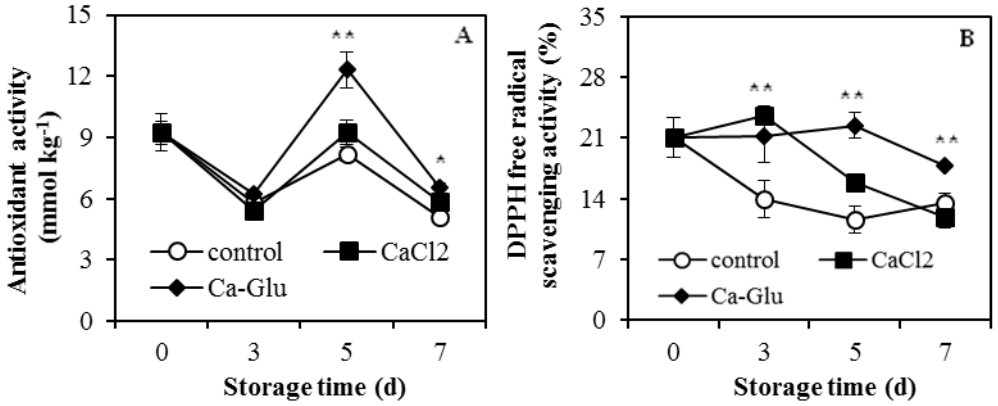

Figure 3. Antioxidant activity (FRAP) (A) and DPPH radical scavenging activity (B) of ready-to-cook baby corn treated with $CaCl_2$ and Ca-Glu compared with control samples during cold storage. Data are presented as mean (n = 4) with SD bar. Significant differences between treatments are indicated with asterisks [** ($P < 0.01$); * ($P < 0.05$)].

3.5 Bioactive compounds

Bioactive compounds such as total phenols, flavonoids and ascorbic acid concentrations of the baby corns are shown in Figure 4. The total phenols concentration of all treatments obviously increased

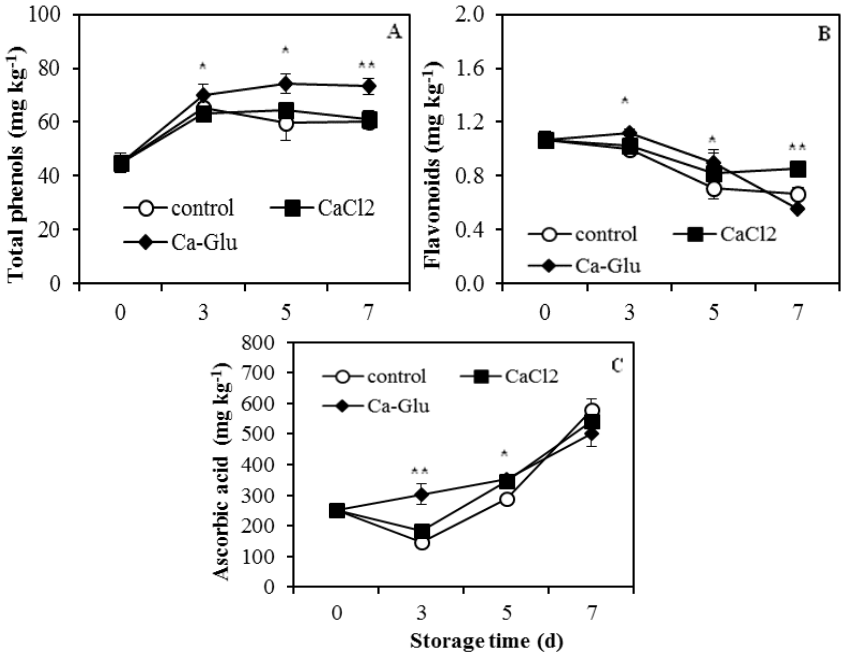

Figure 4. Total phenols (A), flavonoids (B) and ascorbic acid (C) concentrations of ready-to-cook baby corn treated with $CaCl_2$ and Ca-Glu compared with control samples during cold storage. Data are presented as mean (n = 4) with SD bar. Significant differences between treatments are indicated with asterisks [** ($P < 0.01$); * ($P < 0.05$)].

during 3 days of storage, and that of Ca-Glu treated baby corns was significantly higher than those of control and $CaCl_2$ treated samples ($P < 0.05$). After that, total phenols content of Ca-Glu and $CaCl_2$ treated samples remained constant over the storage. The total phenols concentration of control treated samples declined after 3 days of storage. At the end of storage, total phenols concentration of Ca-Glu treated baby corns was significantly higher than those of other samples ($P < 0.05$), whereas no significant difference between control and $CaCl_2$ treated samples was observed. The flavonoids content of Ca-Glu treated baby corns increased during 3 days of storage whilst that of control and $CaCl_2$ treated samples remained constant. After that, the flavonoids content of all treatments decreased. During 5 days of storage, flavonoids content of Ca-Glu treated baby corns was significantly higher than that of control samples ($P < 0.05$). At the end of storage, we found that the flavonoids concentration of $CaCl_2$ treated samples was significantly higher than other samples ($P < 0.05$). The efficiency of calcium treatment enhancing phenolic compounds has been described by Aghdam *et al.* [28] in which calcium stimulated PAL activity and then triggered the phenylpropanoid-flavonoids pathways in cornelian cherries. It is commonly acknowledged that phenolic compounds and flavonoids are synthesized through phenylpropanoid-flavonoids pathways. Many previous studies have reported the enhancement of total phenols and flavonoids contents by various calcium salt applications such as in sweet peppers [27], plums [29], cherries [28] and guavas [30]. We also found that the ascorbic acid concentration of control and $CaCl_2$ treated baby corns decreased during the first 3 d of storage, whilst increased ascorbic acid concentration was found in Ca-Glu treated samples (Figure 4C). After the third day of storage, the increment of ascorbic acid concentration was found in all treatments. On the fifth day of storage, the ascorbic acid concentration of both calcium treated baby corns was significantly higher than that of control samples ($P < 0.05$).

However, at the end of storage (day 7), no significant difference in ascorbic acid concentration of all treatments was observed. Durrani *et al*. [29] reported that the treatments with $CaCl_2$ and Ca-Glu exhibited higher retention of ascorbic acid concentration in plums when compared to untreated fruits during storage. Moreover, foliar application of calcium induced ascorbic acid content in strawberries [31]. However, the results showed that Ca-Glu immersion enhanced ascorbic acid content of ready-to-cook baby corns rather than $CaCl_2$ immersion during 3 days of storage. This might be associated with released gluconic acid which is recognised as a precursor of L-ascorbic acid biosynthesis in plants [32]. The present result also found that the higher total phenols content of Ca-Glu treated baby corn was concomitant with the higher antioxidant activities of FRAP and DPPH radical scavenging activity compared to control and $CaCl_2$ treated samples (Figure 3).

4. Conclusions

Here, our research indicated that the immersion of different calcium salts solutions showed different effects on quality maintenance and improvement of ready-to-cook baby corns during cold storage. Both the $CaCl_2$ and Ca-Glu immersions had no influence on the changes in colour attributes of ready-to-cook baby corns during 7 days storage. The texture of the ready-to-cook baby corns was preserved by immersion in both calcium salts, but $CaCl_2$ evidently maintained better texture when compared to Ca-Glu. The loss of texture was accompanied by an increased EDTA-soluble pectin fraction and decreased Na_2CO_3-soluble pectin fraction during cold storage. Based on our findings, calcium treatments could enhance as well as maintain antioxidant activities and bioactive compounds during refrigerated storage. Ca-Glu at the concentration of 1% (w/v) was likely to enhance antioxidant activity and bioactive compounds to a greater extent than was 1% $CaCl_2$ immersion.

5. Acknowledgements

This work was funded by Kasetsart University Research and Development Institute, Kasetsart University, Thailand. The grant code was KURDI29.61. We gratefully acknowledge the assistance of Dr. J. Anuchai, V. Chimsonthon, C. Kijka and A. Boonlila.

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