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Article

# Quality Changes Monitoring of Fresh-Cut Apples during Distribution in Real-Time Cold Chain Systems Application

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**Abstract:** Fresh-cut apple is commonly known as minimally processed agriculture products because of their convenience and ease of consumption. However, during storage, the quality of the apple rapidly changes after the cutting process due to enzymatic and non-enzymatic processes. This study aimed to monitor the quality changes of the fresh-cut apples at various temperatures and different types of products in the application of cold chain system during distribution and storage. The results showed that the total viable count in fresh-cut apples initially differed between apple only and mixed products (apple, orange, and tomato) during the storage period ( $p < 0.05$ ). Moreover, there are no significant differences ( $p > 0.05$ ) in physicochemicals except firmness, titratable acidity, and pH, which are highly correlated to the microbial growth (total viable count and total coliform number). Furthermore, the optimal temperature for extend the shelf-life of fresh-cut apple was at 4°C.

**Keywords:** fresh-cut apple; temperature; quality changes; storage; cold chain

## 1. Introduction

Fresh agriculture products are well known as a high source of essential nutrients for the human body and consist of natural functional properties such as phenols, antioxidants, flavonoids, and vitamins. Apple is one example of nutritious produce and is common in daily consumption. Fresh-cut apples, categorized as ready-to-eat food products, are minimally processed foods that are washed, peeled, and cut into practice shapes to be consumed and packaged in convenient containers for easy handling [1,2]. However, during storage, the quality of fresh-cut apples quickly changes due to cell damage in apple skin by cutting and slicing, which triggers enzymatic browning. This process produces undesirable attributes such as changes in taste, odor, texture, color, nutritional value, and loss of economic importance in the market [3,4].

Many studies have been conducted to develop the preservation method of fresh-cut produce with minimal process. These techniques include chemical treatments, modified atmosphere packaging, plasma-activated water treatment, thermal treatment, edible coating, hurdle principal, pulsed electric field (PEF), ascorbic acid coating, cold chain systems, and supercooled storage [5–7]. Ascorbic acid is used in apples to act as an anti-browning agent with the combination of  $\text{CaCl}_2$  and effectively halts the enzymatic browning and avoids the tissue softening on the fresh-cut apples [8,9]. Cold chain is essential for maintaining quality and safety in short shelf-life perishable products such as fresh-cut apples. Besides, apple is significantly affected by fluctuating temperature [10,11].

By using the combination of ascorbic acid coating and cold chain systems, the preservation of fresh-cut apples can potentially be retarded in the long term. Therefore, this study aimed to identify the quality changes during the storage time of fresh-cut apples at various temperatures and conditions. Besides, evaluate the characteristics of the samples in the contribution to quality degradation.

## 2. Materials and Methods

### 2.1. Sample and Material Preparation

Fresh-cut apples (150g), coated by ascorbic acid 0.01%, were purchased and stored at different temperatures (15°C for 10 days, 8°C for 20 days, and 4°C for 30 days) at Korea Food Research Institute. Before being placed in the storage room, the samples were distributed by refrigerated food truck at 8°C. During sampling, the interval time was 1 day, 2 days, and 3 days according to the treatment condition. Two samples were placed and compared between apple only and mix (apple, orange, tomato) at 4°C. Real-time internal, surface, and environment temperature in the fresh-cut apple and the storage room were recorded using thermo recorder TR-52i with installed temperature sensor TR-5106 and TR-5320 (T&D Corp., Shimadachi, Nagano, Japan).

### 2.2. Qualities Analysis of Fresh-cut Apples

#### 2.2.1. Moisture Content and Total Soluble Solid

Moisture content was determined using oven-drying techniques. Two grams of sample were placed in an aluminum container and dried using the dry oven (HK-D0134F) for 3 hours at 135°C. The sample moisture was calculated using the following equation:

$$\text{Moisture (wb) (\%)} = \frac{\text{initial weight (g)} - \text{dried sample (g)}}{\text{sample weight (g)}} \quad (1)$$

Moreover, total soluble solid (TSS) was performed using a Refractometer (PAL-1, Atago Co., Ltd., Tokyo, Japan) with three replications of the analysis.

#### 2.2.2. Titratable Acidity and pH

A sample of fresh-cut apples was blended using a fruit juicer (H-100-SBFA01, Hurom Corp., Seoul, Korea) and placed in a 50 mL beaker glass. Moreover, the sample's pH value was obtained using a digital pH meter (TA-70, DKK-TOA Corp., Tokyo, Japan) [12]. Titratable acidity (TA) was calculated by getting NaOH 0.1N volume of 10-gram apple juice until it reached pH 8.2. The TA value was determined as malic acid according to the following equation:

$$TA = \frac{\text{Volume NaOH (mL)} \times 0.1 N \times 134.09 \times 100}{10 \text{ gr} \times 1000} \quad (2)$$

#### 2.2.3. Changes in Appearance and Color Value

The appearance of the apple was captured using mobile camera with the specification of 108MP, f/1.8, 24mm (wide), 1/1.33", 0.8µm, PDAF, Laser AF, OIS. Moreover, the color of the fresh-cut apples was measured using an automatic colorimeter (CR-400, Konica Minolta Inc., Japan) with six samples and three repetitions. The color value was set as CIELAB color parameters  $L^*$ ,  $a^*$ , and  $b^*$ . Total color difference ( $\Delta E$ ), Browning index (BI), and whiteness index (WI) were obtained by calculating according to the following formulas [13,14]:

$$\Delta E^* = \sqrt{(L_{fresh}^* - L_{sample}^*)^2 + (a_{fresh}^* - a_{sample}^*)^2 + (b_{fresh}^* - b_{sample}^*)^2} \quad (3)$$

$$BI = \frac{[100(x-0.31)]}{0.172}, \text{ where } x = \frac{a^*+1.75 L^*}{5.645 L^* + a^* - 3.012 b^*} \quad (4)$$

$$WI = 100 - \sqrt{(100 - L^*)^2 + a^{*2} + b^{*2}} \quad (5)$$

#### 2.2.4. Texture Analysis

Texture analysis (Firmness) of the fresh-cut apple during storage was determined using TA.XT Plus texture analyzer (Stable Micro Systems, UK) as force (g). The analysis was conducted through pre-test, test speed, and post-test of 5 mm/sec with 10 replications using a single blade set (HDP/BS) [2].

#### 2.2.5. Total Viable Count and Total Coliform Count

Total viable count (TVC) and total coliform (CC) were determined by 3M™ Petrifilm™ Aerobic Count (AC) plated method 6400/6403/6406/6442 and Coliform Count Plate (CC) 6402/6412. Ten grams of fresh-cut apple were seized aseptically and diluted with 10-fold by 0.85% saline solution in a sterile filter bag. Furthermore, the sample was homogenized for one minute with four strokes per second using a 400 CC stomacher (BagMixer, Interscience Intl., France). Each instance was moved and diluted with 9 mL of 0.85% saline solution. A particular dilution series (1 mL) was placed in petrifilms and incubated at  $35^{\circ}\text{C} \pm 1^{\circ}\text{C}$  for 48 hours  $\pm 3$  hours (AC plate) and  $32^{\circ}\text{C} \pm 1^{\circ}\text{C}$  for 24 hours (CC plate).

#### 2.3. Statistical Analysis

Two-way analysis of variance (ANOVA) was performed to analyze the data of the experiments and followed by Tukey's multiple range test at 0.05 of significant level. The analysis of variables correlation (Pearson correlation) was also performed using GraphPad Prism version 10.0.3 for Windows, GraphPad Software, Boston, Massachusetts USA, [www.graphpad.com](http://www.graphpad.com).

### 3. Results and Discussion

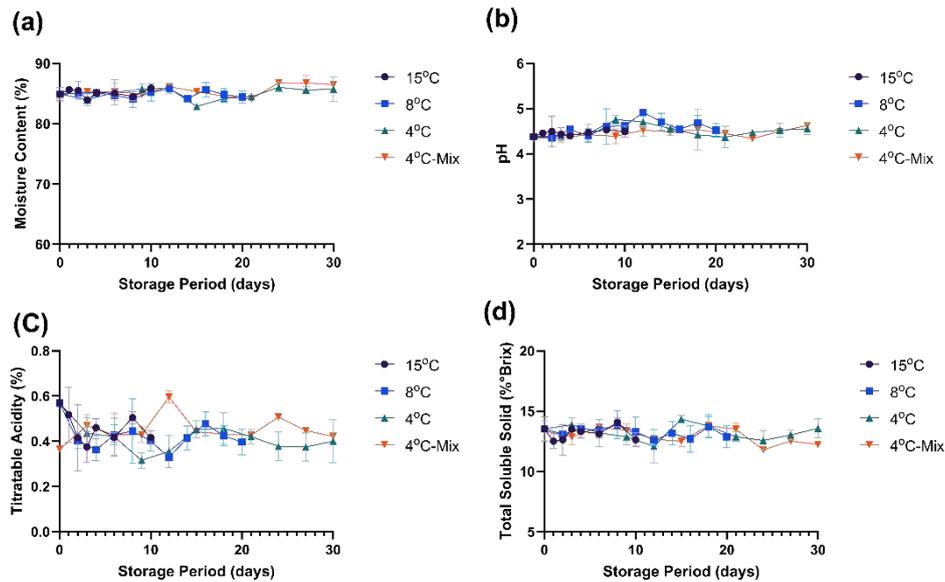
#### 3.1. Physicochemical Properties

##### 3.1.1. Moisture Content, Total Soluble Solid, pH and Titratable Acidity

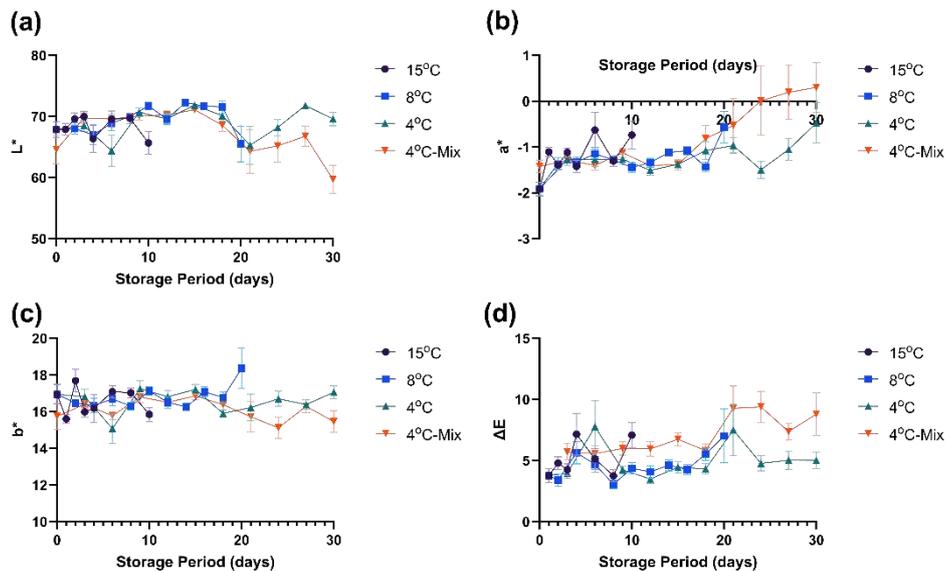
The moisture content in the fresh-cut apples is relatively stable during storage, whether stored at  $15^{\circ}\text{C}$ ,  $8^{\circ}\text{C}$ , or  $4^{\circ}\text{C}$  ( $p > 0.05$ ) (Figure 1 (a)). The plastic packaging of the sample could prevent water loss for over 30 days at  $4^{\circ}\text{C}$ . Moreover, the pH value, titratable acidity, and total soluble solids of all treatments show no remarkable changes from the initial day of storage until the final day of observation ( $p > 0.05$ ) (Figure 1 (b), (c), and (d)). These results align with the previous studies, which show no difference in physicochemical properties changes during storage at  $4^{\circ}\text{C}$  [15–17]. Therefore, the physicochemical properties of fresh-cut apples coated by ascorbic acid in plastic packaging have no significant transformation.

##### 3.1.2. Changes of Color and Appearance

The color variable of fresh-cut apples such as  $L^*$  (whiteness/darkness) and  $b^*$  (yellowness/blueness) in Figure 2 (a) and (c) showed no significant difference among all temperature treatments during storage even though the value from initial day to final day was different ( $p > 0.05$ ). In contrast, the  $a^*$  value, which is represented as (redness/greenness) significantly increased ( $p < 0.05$ ) from day 0 to day 30 of the experiment at  $4^{\circ}\text{C}$ , to day 20 at  $8^{\circ}\text{C}$ , and day 10 at  $15^{\circ}\text{C}$  (Figure 2 (b)). Figure 2 (d) illustrates the fluctuating change of total color difference gradually of fresh-cut apples from day 0 to day 10 at  $15^{\circ}\text{C}$ , from day 0 to day 20 at  $8^{\circ}\text{C}$ , and from day 0 to day 30 for sample apples and mixed.

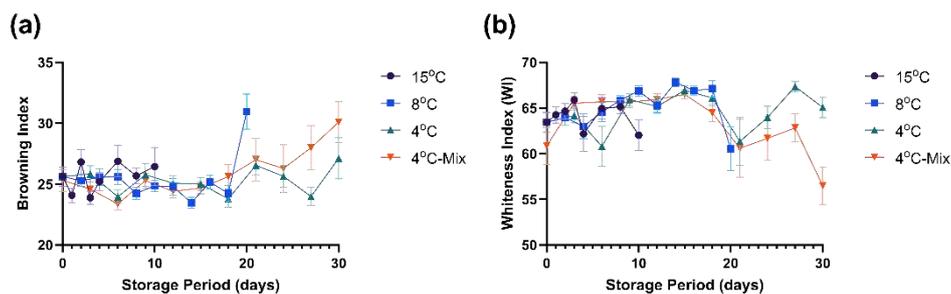


**Figure 1.** (a) Moisture content (%); (b) Changes in pH value; (c) Titratable acidity (TA); and (d) Total soluble solid of fresh-cut apples during storage. The data are expressed in mean  $\pm$  SE,  $n = 3$  with ( $p < 0.05$ ) significantly different.

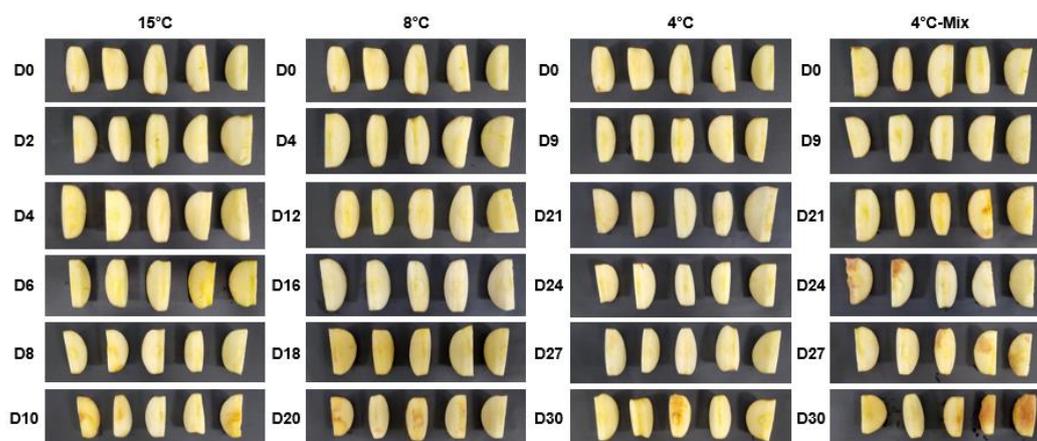


**Figure 2.** Changes in (a)  $L^*$ , (b)  $a^*$ ; (c)  $b^*$ ; and (d)  $\Delta E$  of fresh-cut apple during storage. The data are expressed in mean  $\pm$  SE,  $n = 18$  with ( $p < 0.05$ ) significantly different.

The results indicated that the color parameters, such as the browning index and whiteness index, gradually changed compared to the beginning of observation (Figure 3). Ascorbic acid (AA) could prevent the browning process until 12 days under covered films[18]. The BI tends to increase over 25.61, which is the average value of fresh products. On the other hand, the WI decreased significantly ( $p < 0.05$ ) for all treatments from the initial value of 64.59. In Figure 4, It shows the changes in apple appearance during storage under various temperatures, the apple stored at 15°C slightly change after day 10, the browning process occurred after day 18 at 8°C. Besides, the color of fresh-cut apple significantly change ( $p < 0.05$ ) after 30 days of storage at 4°C. On the other hand, after 21 days of storage at 4°C, the mixed sample deteriorated 9 days earlier than the sample without mixed with orange and tomato in the packaging.



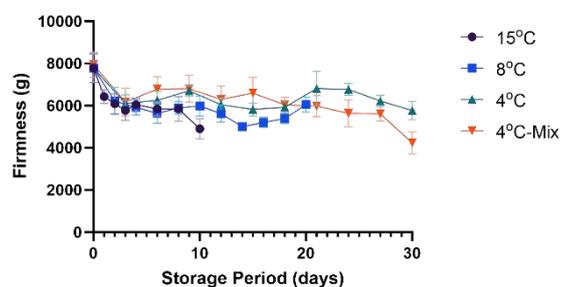
**Figure 3.** Changes in (a) Browning index and (b) Whiteness index of fresh-cut apple during storage. The data are expressed in mean  $\pm$  SE,  $n = 18$  with ( $p < 0.05$ ) significantly different.



**Figure 4.** Changes in appearance of fresh-cut apple during storage under different temperatures.

### 3.1.3. Texture Analysis (Firmness)

The firmness changes of fresh-cut apples with different treatments are shown in Figure 5. It can be seen that the initial force value was 7777.93 g (apples only) and 7944.42 (mixed) and decreased significantly to 4908.24 g at 15°C, 5387.28 g at 8°C, 5769.91 g at 4°C, and 4233.86 g at 4°C (Mixed sample). The texture changes in fruits are related to the transformation of cell wall polymers because of the non-enzymatic, enzymatic reactions and high-pressure processing [15,19].

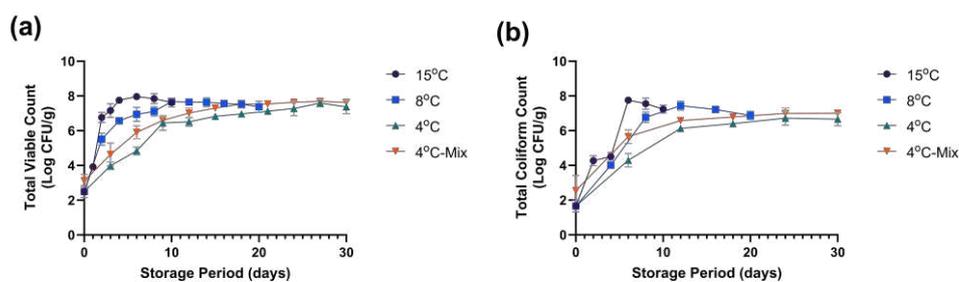


**Figure 5.** Changes in firmness (g) of fresh-cut apples during storage. The data are expressed in mean  $\pm$  SE,  $n = 10$  with ( $p < 0.05$ ) significantly different.

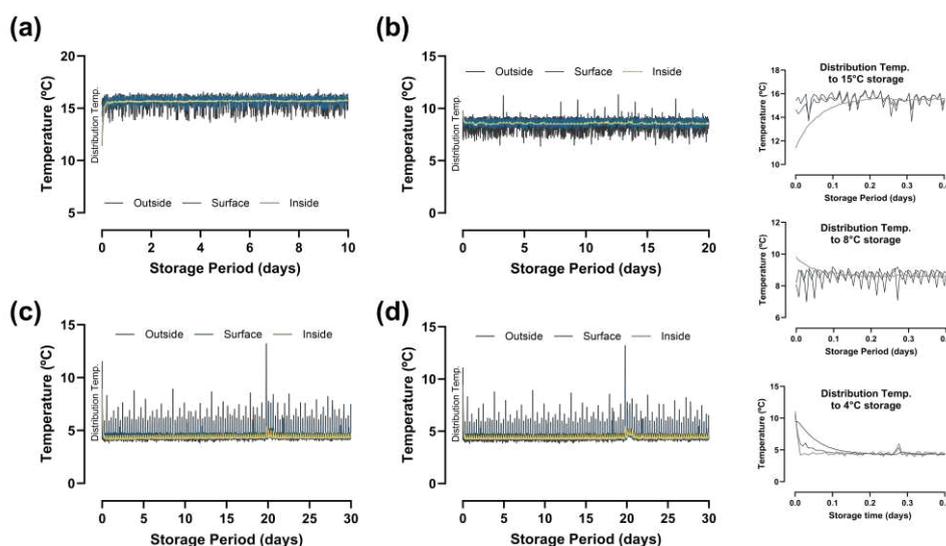
### 3.2. Temperature Changes and Microbial Growth

The microbial growths were evaluated during storage and can be seen in Figure 6. The assessment was conducted to evaluate the food handling and safety in the case of sanitation practices

and equipment from the producer due to the apple, either washed or unwashed, potentially had been contaminated external factors [20,21]. The TVC on the initial day was 2.59 Log CFU/g and 3.2 Log CFU/g (Mix). The number of microbes significantly increased to the maximum number 7.98 Log CFU/g at 15°C, 7.69 Log CFU/g at 8°C, 7.62 Log CFU/g at 4°C, and 7.69 Log CFU/g at 4°C (Mix). Besides, the total coliform of fresh-cut apples during storage increased significantly ( $p < 0.05$ ) from the initial value of 1.74 Log CFU/g and 2.88 Log CFU/g (Mix) in Figure 6 (a). The total coliform incline to 7.77 Log CFU/g at 15 °C, 7.49 Log CFU/g at 8°C, 6.85 Log CFU/g at 4°C, and 7.06 Log CFU/g at 4°C (Mix) in Figure 6 (b). It can be seen that temperature treatment under 4°C can prevent the growth of microbe compared to others. In Figure 7, The internal temperature of apple product was recorded during distribution at  $9.84 \pm 1.03^\circ\text{C}$  which categorized as exotic chill ( $\approx 10^\circ\text{C}$ ) [22]. After the storage, The temperature (inside, surface, and outside) of the fresh-cut apple product gradually changes during the storage and constantly maintained at the controlled temperatures (15°C, 8°C, 4°C, and 4°C-Mixed sample) until the end of monitoring which showed the actual recorded temperature at  $15.65 \pm 0.25^\circ\text{C}$ ,  $8.69 \pm 0.20^\circ\text{C}$ ,  $4.39 \pm 0.23^\circ\text{C}$ , and  $4.34 \pm 0.21^\circ\text{C}$ , respectively. In this condition, the temperature contributed to halt and controlled the growth of microorganisms which initially presented in the products which the storage temperatures (15°C and 8°C) were not proper to extend the shelf-life of the fresh-cut apples products.



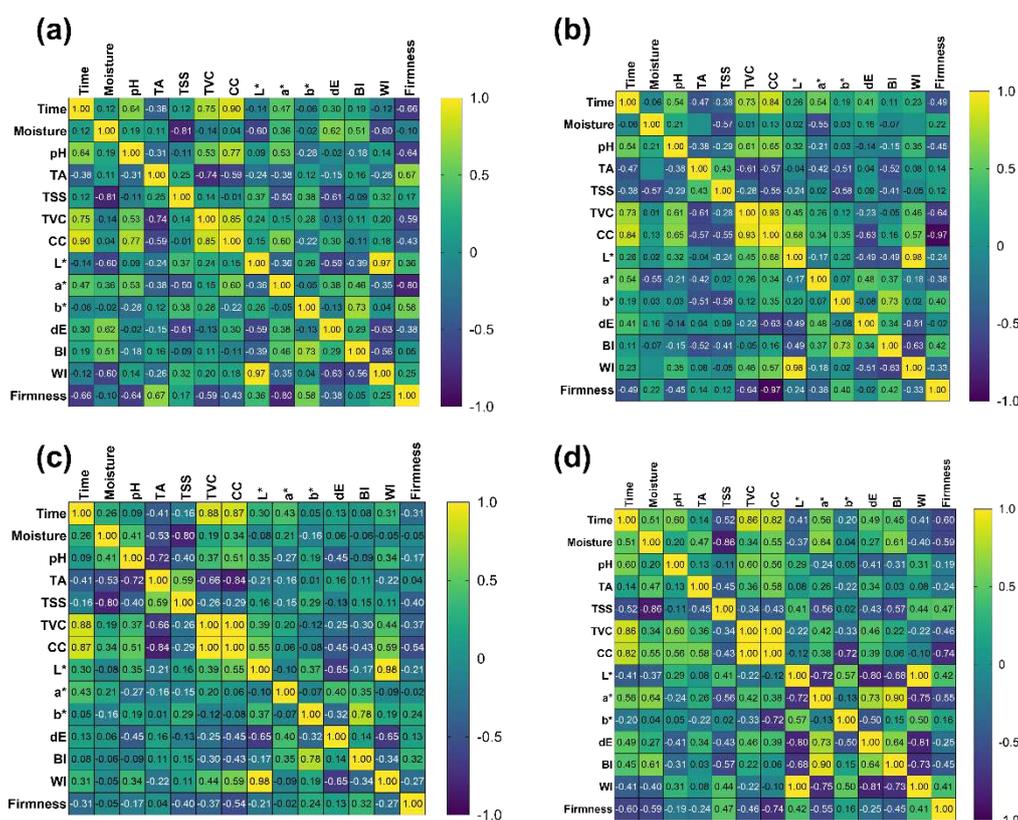
**Figure 6.** Growth of (a) total viable count (TVC) and (b) total coliform count (CC) of fresh-cut apple during storage under different temperature conditions. The data are expressed in mean  $\pm$  SE,  $n = 3$  with ( $p < 0.05$ ) significantly different.



**Figure 7.** Temperature records of fresh-cut apple during distribution ( $10^\circ\text{C}$ ) and storage at (a) 15°C, (b) 8°C, (c) 4°C and (d) 4°C-Mix in outside, surface, and inside the product packaging. The data are expressed in mean with  $n = 2$ .

### 3.3. Statistical and Variables Correlation Analysis during storage

Pearson correlation coefficient ( $r$ ) determine the linear relationship between variables which contain numerical code for grouping the variable from the weakest to the strongest relation (-1 to 1) [23]. Figure 8 shows Pearson correlation among all fresh-cut apple qualities (variables) during storage at different temperatures. At 15°C, the variables that have medium to high correlation to storage time were pH (0.64), firmness (-0.66), TVC (0.75), and CC (0.90). At 8°C, TVC and CC were the most correlated to storage time, 0.73 and 0.84, respectively. The same as at 8°C, the highest correlation in variables of fresh-cut apples to the period of storage at 4°C were TVC (0.88) and CC (0.87). Furthermore, the variables of the sample mix stored at 4°C had a significant correlation to time pH (0.60), firmness (-0.60), TVC (0.86), and CC (0.82). TVC and CC have a high correlation and contribute to the changes of the physicochemical properties of the fresh-cut apple. During storage at 15°C, TVC correlated to the changes of TA and firmness. Besides, at 8°C, TVC contributes to pH, TA, and firmness changes. However, at 4°C, TVC coexists with the decrease of TA (0.66) and pH (0.6) for mixed samples. CC is in line with the increase of pH and  $a^*$  positively and negatively correlated with TA (-0.59) at 15°C. Moreover, CC contributes to increased pH,  $L^*$  and decreased TA, TSS, delta E, and firmness at 8°C treatment. Variable of CC at 4°C influence the decrease of titratable acidity during storage and to  $b^*$  and firmness of mix sample.



**Figure 8.** Pearson correlation among variables of fresh-cut apple stored at (a) 15°C, (b) 8°C, (c) 4°C, and (d) 4°C – Mixed sample.

## 4. Conclusions

This study evaluated the effects of the storage temperature of fresh-cut apples, which are coated by ascorbic acid and placed packed in plastic cups, on changes in quality properties. It can be concluded that fresh-cut apples during storage at 15°C, 8°C, and 4°C have no significant differences in physicochemicals except firmness, titratable acidity, and pH, which are related and correlated to

microbial growth such as total viable count and total coliform number. These results also were confirmed by the statistical analysis of the Pearson correlation.

**Author Contributions:** Conceptualization, J.-H.K. and J.-Y.K.; methodology, J.-H.A. , J.-Y.K. and A.J.L.; validation, J.-H.K., J.-H.A. and J.-Y.K.; formal analysis, A.J.L.; resources, J.-H.A. and J.-Y.K.; data curation, J.-Y.K. and A.J.L.; writing—original draft preparation, A.J.L.; writing—review and editing, J.-Y.K. and A.J.L.; visualization, A.J.L.; supervision, J.-Y.K.; project administration, J.-Y.K.; funding acquisition, J.-Y.K. All authors have read and agreed to the published version of the manuscript.

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**Conflicts of Interest:** The authors declare no conflict of interest.

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