Quality of fresh-cut strawberry

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ABSTRACT

The aim of this work was to study the physical, chemical, physiological and microbiological changes during the flow chart of fresh-cut strawberry. Strawberry cvs. Camarosa, Dover and Tudla, derived from experimental area of the Universidade Federal de Viçosa, were selected by color (red ¾) and absence of wound. Afterwards, the minimal processing was evaluated, as follows: fast cooling, water and ice, removal of the calyx followed by conservation at 5±0.5°C and 90-95% RH for 13 days, sanitation, drainage, cut in halves. Removal of the calyx did not result significant difference for fresh weight, total soluble solids, total titratable acidity, pH and ratio TSS/TAA. The rapid cooling resulted in lower electrolyte leakage and respiratory rate, especially sanitized fruits. Sanitization by immersion in chlorine solution slowed the growth of fungi and yeast. Drainage for 20 minutes eliminated practically all water on the surface of the fruits. The fresh-cutting did not affect the visual and nutritional quality of strawberries like appearance, microbriot, vitamin C, anthocyanins and phenolic compounds, consisting of alternative market potential economically viable.

Keywords: Fragaria x ananassa Duch., sanitation, drainage, anthocyanins.

RESUMO

Qualidade de morango minimamente processado

O objetivo deste trabalho foi estudar mudanças físicas, químicas, fisiológicas e microbiológicas durante o fluxograma de processamento mínimo de morango. Morangos das cultivares Camarosa, Dover e Tudla oriundos de área experimental da Universidade Federal de Viçosa, foram selecionados por cor (¾ vermelho) e ausência de danos. Posteriormente, foi avaliado o fluxograma de processamento mínimo: resfriamento rápido, água e gelo; remoção do cálice acompanhado de conservação a 5±0,5°C e 90-95% UR, por 13 dias; sanitização; drenagem; corte, em metades. A remoção do cálice não resultou diferença significativa para massa fresca, sólidos solúveis totais, acidez total titulável, pH e razão SST/ATT. O resfriamento rápido ocasionou menores extravasamentos de eletrólitos e taxa respiratória, sobretudo nos frutos sanitizados. A sanitização por imersão em solução de cloreto retardou o crescimento de fungos e levedura. A drenagem por 20 minutos eliminou praticamente toda a água na superfície dos frutos. O processamento mínimo não afetou a qualidade visual e nutricional dos morangos como aparência, microbriot, vitamina C, antocianinas e compostos fenólicos, consistindo em alternativa de mercado potencial economicamente viável.

Palavras-chave: Fragaria x ananassa Duch., sanitização, drenagem, antociáninas.

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The strawberry is appreciated for its characteristic color, flavor and aroma. The non-climacteric fruit has a high respiratory rate with high moisture content (90%) and sugar content (glucose 4%, fructose 5% and sucrose 0.9%), an ideal substrate for proliferation of microorganisms, fungi and yeasts such as Botrytis cinerea (Prasanna et al., 2007).

Microorganism growth in minimally processed products can be controlled using quality raw material, sanitization, proper drainage and low temperature (Del Aguila et al., 2006). Sanitization reduces the number of microorganisms in the food and will depend on the disinfectant concentration and exposure time (Sant’ana et al., 2002). Drainage removes excess water or water and cell juice from the fruit surface preventing liquid accumulation inside the packages and the later microorganism development.

Generally, immersion in aqueous solution is not recommended in minimal processing of perishable fruit. However, Costa et al. (2006) recorded up to eight days of shelf life in minimally processed strawberries, sanitized with chlorine solution.

The adaptation of the plant tissue after harvest, preparation and obtaining of the end product can be defined by quality indicators such as appearance and nutritional value. Thus the objective of the present study was to study the physical, chemical, physiological and microbiological changes during the minimal processing steps for strawberries.
MATERIAL AND METHODS

Obtaining the raw material
Strawberries of the Camarosa, Dover and Tudla cultivars produced in a greenhouse in an experimental area at the Crop Science Department of the Federal University of Viçosa were collected in plastic trays (before 8 a.m.) and taken to a fruit and vegetable minimal processing unit of the Plant Biology Department, for sampling for color (¼ red) and absence of damage.

Assessing the minimal processing flow gram
1. Calyx and penduncle removal - the calyx was removed by hand and the penduncle using a sharp blade, 3 mm from the base. The fruits were divided into two lots, with and without calyx, placed on PET trays (177x122x40 mm) with 150 g fruits, without a lid, wrapped in poly vinyl chloride film (PVC, 15 μm) and kept in a vertical display with forced air circulation (Metalfrio), at 5±0.5°C, under 90-95 % RH. The green matter, soluble solids, titratable acidity, strawberry pH and the SST/ATT ratio were analyzed every three days, starting on day one, for 13 days (Costa, 2009).

2. Quick cooling - Recently collected fruits were cooled quickly (water and ice, 5±1°C), for 15 minutes and compared with uncooled strawberries (environment, 22±2°C, for the controlled experiment.

3. Sanitization - strawberries were immersed in chlorine aqueous solution (Sumaveg®) at 5±1°C, at the concentrations of 0.5 (tap water); 200 and 400 mg L⁻¹, then rinsed with 5 mg L⁻¹ chlorine at a 5±1°C. The following sanitization times were tested: 30 seconds, 5 and 10 minutes for a fixed sanitization concentration.

4. Drainage - after the sanitization and rinsing steps, the strawberries were submitted to drainage times of 20, 40 and 120 minutes, on perforated trays at the processing environment temperature, 18±2°C under 80±5 % RH.

5. Cutting - after the drainage step, one part of the strawberries was cut (in half, longitudinally) using sharp blades and the other part of the fruits was kept whole to form the control experiment.

Physical, chemical, physiological and microbiological analysis
1. Electrolyte leakage - described by Simon (1977) and adopted by Costa (2009) was estimated using the leakage curve of the cell juice from epidermis tissue and pulp (5 mm² per 2 mm of thickness) removed from the equatorial region of the strawberries using blades up to 2 g sample. The sample was placed on a glass recipient (117 mL) with a lid containing 40 mL deionized water and the leakage readings were taken in CD-850 conductivity every hour for four hours. The samples were then submitted to eight sessions of 30 seconds in a microwave oven (Electrolux ME 28s) with a 30 second interval with the doo open between each session to prevent overflowing. The leakage was obtained according to Stuart (1939):

\[ E_\% = \left( \frac{E_{4h}}{E_t} \times 100 \right) \]

where: \( E_\% \) = electrolyte leakage, %; \( E_{4h} \) = leakage in 4 h and, \( E_t \) = total leakage.

2. Standard filamentous fungi and yeast count - the surface spreading technique was used, inoculating 0.1 mL of the 10⁻¹, 10⁻² and 10⁻³ dilutions in triplicate containing Agar, potato and dextrose – BDA (Oxoid®), acidified with 10% tartaric acid, pH 3.5 and incubated at 25°C, for 5 to 7 days (Beuchat & Cousin, 2001).

3. Quantity of water absorbed - estimated by gravimetry, on 0.01 g precision scales from the differences in fresh matter before strawberry sanitization and after sanitization, rinsing and drainage.

4. Carbon dioxide quantification - about 50 g strawberries were kept in 170 mL hermetically sealed flasks for 0, 0.5, 1, 2, 4, 8 and 12 hours to make the accumulation curve and identify the ideal sampling time. The quantification was estimated according to Costa (2005), with a 1.0 cm³ aliquot of the atmosphere of the flasks in ultrafine syringes (29 G1/2” needles) and injected in a GC-14B gas chromatographer (Shimadzu, Kyoto), with a heat conductivity detector and column filled with Porapak-Q. The drag gas was nitrogen with 30 cm³/min flow and 85 mA electric current. The temperature of the column, injector and detector were adjusted to 40, 100 and 180°C, respectively.

5. Sample extraction for vitamin C, anthocyanin and phenol compounds - the cell juice was extracted in a Comfort CE55 - Walita® domestic centrifuge and filtered through four layers of gauze to quantify the vitamin C, anthocyanins and phenol compounds.

5.1. Vitamin C - according to Strohecker & Henning (1967), using the colorimetric method of 2,4-dinitrophenylhydrazine (DNPH) with adaptations. About 2 g sample were diluted in 100 mL 0.5% oxalic acid, 20 mL was filtered and 300 µg active carbon were added and left to rest for 15 minutes. From the clarified sample 400 µL were reacted with 3.6 mL 0.5% oxalic acid, 60 µL 2.6-0.2% dichlorophenolindophenol (DCPIP), 1 mL 2.4-DNPH 2% and 20 µL 10% thiourea. The reaction took place in a water bath, 100°C for 10 minutes and was interrupted in a water bath with ice for 10 minutes. Then, 5 mL 85% sulfuric acid were added slowly and left to rest for 10 minutes. The standard curve was prepared with ascorbic acid and the readings made on a Hitachi U-2000 spectrophotometer at 520 nm.

5.2. Anthocyanins - described by Giusti & Wroslstad (2001) by pH difference, with adaptations. The method is based on two buffer systems: potassium chloride 0.025 M, pH 1.0 sodium acetate, 0.4 M pH 4.5. For each system, 5 mL buffer plus 200 µL sample were used to obtain absorbency readings between 0.100 and 1.200, at the maximum wave lengths of the visible (εmax.vis. = 497 nm) and at 700 nm. The difference in absorbency between the buffer systems was calculated according to the following equation:

\[ A_1 = (A_{\text{εmax.vis.}} - A_{700m})_{pH1.0} \]
\[ A_2 = (A_{\text{εmax.vis.}} - A_{700m})_{pH4.5} \]
\[ \Delta A = A_1 - A_2 \]

The pigment content was estimated from the most abundant anthocyanin in the strawberry (pelargonidin-
3-glucoside), by the equation:

\[ At = \frac{(\Delta A \times PM \times f \times 100)}{(e \times 1)} \]

where: \( At = \) anthocyanin, mg/100 g fresh matter; \( \Delta A = \) difference in absorbancy, \( (A_{INIT} - A_{FINAL}) \); \( PM = \) molecular weight of the pelargonidina-3-glucoside, 451.2; \( f = \) dilution factor for the strawberry, 25x; \( e = \) coefficient of molar absorptivity, 15600.

5.3. Total phenol compounds - according to Waterhouse (2006) by Folin-Ciocalteu, reacting 25 µL of the sample in 1.575 µL distilled water and 100 µL Folin-Ciocalteu, then shaken and left to rest for five minutes. After adding 30 µL 20% sodium carbonate, the solution was kept in a water bath at 40°C for 30 minutes. In basic culture the phosphowolframic and phosphomolybdic acids reduce when oxidizing phenol compounds giving rise to blue wolframic oxides \((W_8O_{23})\) and molybdenum \((MoO_{23})\). The standard curve was prepared with galic acid and absorbency reading at 765 nm.

6. Statistical analysis - Analysis of variance was carried out using the SAEWG 9 program. The F test was applied to assess the significance and the Tukey test was used at the level of 5% probability whenever necessary. The cultivar and the replication for each experimental arrangement varied in function of the quantity of raw material available.

RESULTS AND DISCUSSION

Strawberry storage after removing the calyx

Removing the calyx had no significant effect for green matter, SST, ATT, pH and the SST/ATT ratio compared to the fruits with the calyx (Table 1A). There was, however, significant effect of storage time for green matter, SST, pH and the SST/ATT ratio.

At the end of the 13 days, the loss of accumulated green matter was 0.8% in the fruit with and without calyx (Table 1A). This value may have been due to the type of packaging used, a PET tray with PVC film (15) that functioned as a physical barrier to water vapor loss. Furthermore, using low temperature, 5±0.5°C and high relative humidity, 90-95%, contributed to reducing green matter loss from the strawberries.

It is believed that the presence of the calyx accelerates water loss from the calyx + pseudofruit set, because it raises the fruit transpiration rate (personal communication). However, the green matter loss observed in the Dover strawberry was minimal (Table 1A). This performance cannot be adopted as a general rule because in addition to temperature, relative humidity and packaging type, the cultivar may present calyx with variable morphological pattern, as observed visually in the Camarosa and Oso Grande fruits, with larger bracts in expansion compared to Dover (data not shown).

Generally, strawberries are used for fresh consumption with the calyx free from dirt and contaminations (Cantillano, 2004). In industry, the strawberries can be used with or without the calyx.

In the fresh strawberry, a turgid and clean calyx indicates freshness and attractiveness and can be used by consumers as a desirable characteristic. In eggplant, for example, the calyx is used as a quality indicator by producers and consumers (Moretti & Pineli, 2005).

The SST content remained practically constant throughout the storage period, ranging from 7.3 to 6.4%, with greatest decrease on the second day of analysis (Table 1A). This characteristic is of commercial interest, especially for fresh strawberries, because the consumer prefers sweeter fruit (Fumis et al., 2003).

The pH increased in the fruit with and without calyx from 3.17 to 3.94 during storage (Table 1A). The Dover strawberry was considered acid, not very sweet or even sour. The main acids present in the fruit, citric and malic, can directly affect the flavor, cell pH and coloring by anthocyanins (Fumis et al., 2003; Cantillano, 2004).

The SST/ATT ratio decreased after the second day, ranging from 10.6 to 9.3 and remained practically constant until the 13th day (Table 1A) with values above the commercially required (8.75) characterized by sweet fruits with low acidity.

**Strawberry under cooling, sanitization, drainage and cutting**

a) Electrolyte leakage

Electrolyte leakage up to 4 h four was the most indicated, with

Table 1. Fresh weight loss. total soluble solids. total titratable acidity. pH and ratio TSS/TTA of Dover strawberries. with and without calyx. kept at 5±0.5°C under 90-95% RH; (perda de massa fresca. sólidos solúveis totais. acidez total titulável. pH e razão SST/ATT de morango Dover. com e sem cálice. mantidos a 5±0.5°C sob 90-95% UR). Viçosa. UFV. 2009.

<table>
<thead>
<tr>
<th>Days</th>
<th>PMF (%)</th>
<th>SST (%)</th>
<th>ATT (%)</th>
<th>pH</th>
<th>SST/ATT</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>with</td>
<td>without</td>
<td>with</td>
<td>without</td>
<td>with</td>
</tr>
<tr>
<td>1</td>
<td>0.00 e</td>
<td>0.00 e</td>
<td>7.3 a</td>
<td>7.3 a</td>
<td>0.69 a</td>
</tr>
<tr>
<td>4</td>
<td>0.27 d</td>
<td>0.25 d</td>
<td>6.6 b</td>
<td>6.5 b</td>
<td>0.71 a</td>
</tr>
<tr>
<td>7</td>
<td>0.42 c</td>
<td>0.39 c</td>
<td>6.8 b</td>
<td>6.7 b</td>
<td>0.68 a</td>
</tr>
<tr>
<td>10</td>
<td>0.59 b</td>
<td>0.57 b</td>
<td>6.6 b</td>
<td>6.6 b</td>
<td>0.65 a</td>
</tr>
<tr>
<td>13</td>
<td>0.78 a</td>
<td>0.73 a</td>
<td>6.3 b</td>
<td>6.3 b</td>
<td>0.64 a</td>
</tr>
<tr>
<td>CV (%)</td>
<td>9.5</td>
<td>9.3</td>
<td>4.9</td>
<td>4.8</td>
<td>4.8</td>
</tr>
</tbody>
</table>

In the line, no significant difference and in the column means followed by same letter did not differ. Tukey at the level of 5% probability (na linha. não houve diferença significativa e na coluna médias seguidas de mesma letra. não diferem entre si. Tukey ao nível de 5% de probabilidade).
Table 2. Levels of vitamin C, anthocyanins and phenolic compounds after minimal processing Camarosa, Dover and Tudla strawberries (têores de vitamina C, antocianinas e compostos fenólicos após o processamento mínimo de morangos Camarosa, Dover e Tudla). Viçosa, UFV, 2009.

<table>
<thead>
<tr>
<th>Cultivar</th>
<th>Vitamin C (mg/100 g of fresh mass)</th>
<th>Anthocyanins</th>
<th>Phenolic compounds</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Whole fruit</td>
<td>Cut fruit</td>
<td>Whole fruit</td>
</tr>
<tr>
<td>Camarosa</td>
<td>-</td>
<td>-</td>
<td>43 bA</td>
</tr>
<tr>
<td>Dover</td>
<td>59 aB</td>
<td>54 bA</td>
<td>25 aB</td>
</tr>
<tr>
<td>Tudla</td>
<td>66 aA</td>
<td>58 bA</td>
<td>-</td>
</tr>
</tbody>
</table>

Means followed by the same small letter in the line and capital in the column did not differ, Tukey at the level of 5% probability (médias seguidas de letra minúscula na linha e maiúscula na coluna, não diferem entre si, Tukey ao nível de 5% de probabilidade).

discrete sensitivity among the cultivars Camarosa (9%), Tudla (11%) and Dover (13%) (Figure 1A).

The relative growth rate in volume of the strawberry is a result of cell division and filling or of their joint action, and the contribution of each factor is relative to the species, variety or cultivar. Furthermore, the genetic difference among cultivars influences the structural and mechanical development of the tissue (Havis, 1943) and strawberries consisting structurally of small cells are more resistant (Havis, 1943) to post-harvest handling and minimal processing.

Electrolyte leakage in non-cooled strawberries (22±2°C) and without sanitization was greater compared to the sanitized fruit (Figure 1b) and this difference was smaller in the strawberries submitted to cooling (5±1°C). The difference was around 5% among the non-sanitized fruits at 22±2°C and the cooled fruits, showing that cooling (water plus ice) reduced the field heat in the fruits.

Strawberries cooled quickly after harvest, in a forced air chamber at 1°C for one hour, and stored for one week at 1°C plus one day at 20°C, had attractive coloring, less green matter loss, greater firmness, minimal vitamin C and sugar content loss compared to strawberries when there was a six hour delay in cooling and exposure to 30°C (Nunes et al., 1995).

Soft and succulent fruit consist of cells with high solute content, a condition that can generate turgor pressure above the limit of cell wall resistance (Simon, 1977) making them sensitive and vulnerable to leakage. In

Figure 1. A) curve for electrolyte leakage in Camarosa, Dover and Tudla strawberries; B) leakage in Camarosa strawberry with and without cooling, not sanitized (NS) and sanitized (S); C) leakage of whole and cut fruits, with and without cooling (A) curva para extravasamento de eletrólitos em morangos Camarosa, Dover e Tudla; B) extravasamento em morango Camarosa com e sem resfriamento, não sanitizado (NS) e sanitizado (S); C) extravasamento de frutos inteiros e cortados, com e sem resfriamento; barras verticais indicam desvio padrão da média). Viçosa, UFV, 2009.
the case of the sanitized strawberries, the epidermis and adjacent cells may have increased the turgor pressure, and thus acquired less mechanical resistance. In this sense, sanitization may be harmful but not sufficiently to prevent its use in minimal processing of strawberries or fruits with similar sensitivity.

Electrolyte leakage ranged from 38 to 43% in the cut strawberries with and without cooling, respectively, while in the whole fruit, this variation was 34 and 36% in the same cooling order (Figure 1C). Thus using cooling reduced the stress induced by cutting especially the electrolyte leakage resulting from cell breakdown.

The cells of the fruits did not break easily in situ, because they were under pressure from the neighboring cell layers or protected by resistant epidermis, cuticle or skin cells. Once the skin was removed, the pulp cells become vulnerable to any hypertonic condition (Simon, 1977). The strawberry has a thin and fragile epidermis that may make it more sensitive to cell leakage when submitted to minimal processing conditions such as washing, sanitization, rinsing and cutting.

b) Microorganism growth

The sanitization concentrations and sanitization times did not differ on the strawberry microbiota (Figure 2). Regarding the sanitization concentrations tested, the decrease in the number of log UFC was 0.3 for 200 and 400 mg L$^{-1}$ chlorine compared to the non-sanitized and sanitized fruits with 0.5 mg L$^{-1}$ (Figure 2). At the 5 and 10 minute sanitization times, the reduction was 0.5 log UFC g$^{-1}$ compared to the non-sanitized and strawberries sanitized for 30 seconds, with 200 mg L$^{-1}$ chlorine (Figure 2). This showed that the microbiota present on the strawberry during the processing was insufficient to damage the fruit safety.

Microorganism incidence on strawberries and other low pH fruit (3.5 and 4) is due to filamentous fungi and yeast. Furthermore, this pH is low enough to inhibit the growth of other microorganisms such as bacteria (Park et al., 2005).

The microbiological changes that occur at the start and during fruit and other vegetable storage vary with the microbiota of each product and are directly linked to factors such as hygiene.
during handling and processing location, temperature, relative humidity and water quality (Porte & Maia, 2001).

C) Drainage

Drainage was essential in the strawberry minimal processing and removed the excess water from the fruit surface by 2.5%, regardless of the sanitization time (Figure 2). The water available in the Dover strawberry, sanitized and rinsed for 30 seconds, fell to 0.7 and 0.3% in drainage for 20 and 40 minutes, respectively (Figure 2A). In the fruits sanitized and rinsed for five minutes, drainage for 20 and 40 minutes resulted in water absorbed with 1.3 and 1%, respectively (Figure 2B). In the fruits sanitized and rinsed for 30 seconds and drained for 20 minutes permitted the minimum of water adhering to the fruit surface, besides making the process quick compared to the other drainage times studied.

Figure 4. Production of carbon dioxide after processing Camarosa, Dover and Tudla whole strawberries, not sanitized and sanitized; fruits, whole and cut (produção de dióxido de carbono após o processamento de morangos Camarosa, Dover e Tudla inteiros, não sanitizado e sanitizado; frutos, inteiro e cortado). Viçosa, UFV, 2009.

Cutting in minimal processing is a very intense stress to the tissue because some physiological manifestations are due to the rupture of the cell structures such as increase in carbon dioxide production (Porte & Maia, 2001). In some minimally processed products, the respiratory rates can increase three to seven-fold compared to the intact tissue, with accelerated oxygen intake.
Thus respiration in fruits and vegetables can serve as an indicator of catabolic alteration, especially when the tissue is peeled or cut (Maistro, 2001).

e) Vitamin C, anthocyanins and phenol compounds

The vitamin C contents in the whole Dover and Tudla strawberries were greater, about 8 to 12%, respectively, compared to the cut strawberries (Table 1B).

Vitamin C predominates in the strawberry in the form of ascorbic acid with an average content of 60 mg/100 g MF and can vary with the cultivar and ripening stage (Pazinato, 1999; Cordenunsi et al., 2005).

In minimal processing, vitamin C can serve as an excellent quality indicator because it is labile and susceptible to degradation in the presence of light and oxygen so that its content can be affected at the cutting stage. Ascorbate oxidation by ascorbic oxidase increases under adverse conditions such as high temperatures, metallic ions, physical and chemical damage and with exposure to products that contain halogens in the molecule such as hydrochloride salts used in sanitization (Lee & Kader, 2000).

The cut Camarosa strawberry was outstanding compared to the whole fruit with anthocyanin and phenol compound contents of around 15 and 5% more, respectively (Table 1B). The Dover strawberry had lower anthocyanin contents compared to Camarosa, with practically equal contents in the whole and cut fruit (Table 1B). The anthocyanin contents found in the whole and cut Seascape strawberry was around 45 mg/100 g MF (Gil et al., 2006), close to that estimated for Camarosa (40-50 mg 100/g MF) and greater than the contents detected in Dover (25 mg 100/g MF), showing that the anthocyanin content is a characteristic intrinsic to the cultivar. The Dover strawberries had a higher anthocyanin content compared to the Campineiro and Oso Grande cultivars in the study by Cordenunsi et al. (2005).

The anthocyanins and the factors that affect their synthesis, content and stability account for the color in the strawberry, and pelargonidin-3-glucoside is the most common anthocyanin in strawberries (Lopes da Silva et al., 2003).

Regarding the phenol compound contents, the Dover strawberry performed similarly to the Camarosa (Table 1B). The phenol compound contents in the strawberry can vary with the cultivar and the extraction and quantification methods, and can function as anti-oxidants through the phenolic acids and the anthocyanins themselves (Kosar et al., 2004). The phenol compound content estimated in the whole and cut Seascape strawberry was 55 mg/100 g (Gil et al., 2006), below that estimated for Camarosa and Dover, close to 95 and 80 mg/100 g MF, respectively.

Strawberry minimal processing did not result in drastic loss in the visual and nutritional quality of the fruit, especially of the vitamin C, anthocyanin and phenol compound contents and was shown to be an economically viable market alternative and promising for strawberry producers.

The strawberry minimal processing flow chart was defined starting with operations of cooling with water and ice, removing the calyx and penduncle, sanitization, rinsing, draining, cutting and wrapping. Future studies are very important on the performance of minimally processed strawberries under cooled storage.

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