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UV-C in Washing Treatment for Preserving Quality and Functional Content of Minimally Processed *Fragaria Vesca* Strawberry

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I. INTRODUCTION

Minimally processed products are quite popular among consumers, who are demanding ready-to-eat fruits and vegetables, presumably due, in part, to their characteristics of freshness, low caloric content, commodity to be used and an active promotion of fruits and vegetables as basic components of a healthy diet (5). Nevertheless, it is well known that minimal processing alters the integrity of the fruit and induces surface damages increasing lightly the tissue respiration and leading biochemical deteriorations such as browning, off-flavour development and texture breakdown decreasing the fresh-cut fruit quality (14, 16).

Strawberries that are one of the most popular fruits worldwide, are rich in nutrients but also highly perishable, being susceptible to mechanical injury, desiccation, decay and physiological disorders during storage. Among fruits, they have one of the highest antioxidant activities (6). The shelf life of fresh strawberries at cold temperature is usually less than 5 days. This storage time is reduced when the product is minimally processed. Improvements in shelf-life can be achieved by using good quality raw products, special care during processing and along the trade chain, control of temperature and relative humidity, and use of modified atmosphere packaging (19).

In order to decrease microbial contamination, fresh-cut industry commonly uses sodium hypochlorite as disinfection agent but by-products such as trihalometanes and chloramines are potentially harmful for healthy human and the production of high amounts of wastewater producing environmental risk making necessary the search of alternative disinfectant agents.

Organic acids, mainly citric, lactic and acetic acid, which are in GRAS (Generally Recognized As Safe) status, have been investigated because of their bactericidal activity (21, 24). Physical methods including ultraviolet radiation (UV) have been shown to be capable of killing or inhibiting bacterial growth (27). Its application to a re-circulating water stream maintains the water at a reasonable bacteriological quality, but has no effect at all on surfaces either of the process machinery or on the product itself (8). As pathogens can survive for relatively long times in water, they can subsequently contaminate the product that passes through it before microbial inactivation with UV occurs. The maintenance of the quality of the process water is very important as it might serve as a source of cross-contamination. In fact, the main effect of sanitizing treatments for washing fresh-cut produce is to reduce the microbial load and keep process water free from contamination rather than having a preservative effect (9).

In the present study, samples were directly processed in a fresh-cut processing line and cleaning room. The main purpose of this work was to evaluate if the use of UV radiation in the washing water affects the microbial growth, sensory and functional quality of fresh processed *Fragaria vesca* strawberry.

II. MATERIALS AND METHODS

a) Materials

Strawberries (*Fragaria vesca* L.) were provided by Asociación para el Desarrollo del Sistema Productivo Vinculado a la Agricultura Onubense (ADESVA) (Huelva, Spain).

b) Sample preparation

The temperature in the processing clean room was maintained at 8 °C. The steps of processing were: (a) *reception*: strawberries were carefully selected for uniform size and colour as well as the absence of

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damage and defects; (b) *cutting*: the calyx was cut off using special knives designed for fruit; (c) *washing*: strawberries were washed during 90 seconds using one patented industrial washing equipment, model CAMEL (Turatti, Italy) coupled with a ultraviolet system of six lamps (Montagna, Italy). The following treatment were applied: water and UV-C radiation (30 mW cm⁻²); (d) *draining*: strawberries were drained using an industrial drying tunnel system by hot/cold air, Mod. "Domino Junior Laboratorio" (Turatti, Italy); (e) *packaging*: 150.0 g of strawberries were placed in a polypropylene (PP) tray and thermally sealed with a PP film (permeability to O₂ was 114 cm³m⁻² 24h at 25 °C) in order to generate a passive modified atmosphere (MAP). An industrial packaging, Mod. "Verpackungs-Systeme" (Western, Germany) was used; (f) *storage*: packaged samples were stored at 4 °C in refrigeration for up to 10 days.

c) Total soluble solids, titratable acidity and pH

Total soluble solids (TSS), titratable acidity (TA) and pH were measured for each independent homogenate. TSS were measured by refractometry using a RE40 refractometer (Mettler Toledo, S.A.E., Coslada, Spain); results are expressed as °Brix. TA and pH were determined using a DL50 Graphix automatic titrator (Mettler Toledo, S.A.E., Coslada, Spain). Results were expressed as g malic acid 100 g⁻¹ fresh weight (fw).

d) Colour

Flesh colour was measured using a CR-200 tristimulus colorimeter (Minolta, Tokyo, Japan), with an 8 mm diameter viewing area and using illuminant D65. Chromatic analyses were conducted in accordance with the CIE (Commission Internationale de l'Eclairage) system of 1976. Values of L*, a* and b* were used to define a three-dimensional colour space.

e) Head space gas composition

O₂ and CO₂ content of all strawberries trays were measured using an O₂ and CO₂ meter PAK 12P (Control y Suministros S.A, Barcelona, Spain). Each tray was used only for a single determination.

f) Total phenolic content, anthocyanins content and total antioxidant activity and

Total phenolic content was obtained from 5 g of homogenate. The colorimetric reaction was developed by using Folin-Ciocalteu reactive. After one-hour reaction, absorbance was measured at 760 nm with a UV-2401 PC spectrophotometer (Shimadzu Scientific Instruments, USA). TPC was quantified by an external standard method using gallic acid and expressed as mg gallic acid 100 g⁻¹ fresh weight (15).

Anthocyanins were extracted from 10 g of homogenate in 50 ml acidic methanol solution (0.2 % HCl), evaluated following the González-Gómez method (10), absorbance was measured at 520 nm with a UV-2401 PC spectrophotometer (Shimadzu Scientific

Instruments, USA). The quantification was carried out by the external standard calibration method, using cyaniding 3-O-rutinoside as standard and expressed as mg cyaniding 3-O-rutinoside 100 g⁻¹ fresh weight (n=4). Total antioxidant activity was determined using a Thermo Scientific Appliskan spectrophotometer (Thermo Fisher Scientific, Waltham, Massachussets, USA), equipped with a 24-well plate. For the spectrophotometric reaction, 20 µl sample juice was added to 1 ml ABTS (2,2'-azinobis(3-ethylbenzoithiazolone 6-sulphonate) (n=4). Absorbance loss was measured at 750 nm for 20 min. Trolox was used as external standard to quantify TAA, being expressed as mg Trolox 100 g⁻¹ fresh weight (Cano et al., 1998).

g) Microbiological analyses

For microbiological assessments of mesophiles, psychrotrops, moulds and yeast, the following methods according to ISO 4833 (11) and ISO 7954 (12) were used and the results were expressed as log CFU g⁻¹.

h) Firmness determination

Firmness was measured using a Stable Micro Systems Texture Analyzer TA-XT2i (Aname, Spain) through a compression assay on the whole strawberry. The force was applied to produce a 2 % deformation by a 100 mm aluminium plate. The maximum force (N) was calculated.

i) Sensory evaluation

Sensory quality was evaluated by a semi-trained panel consisting of eight members. Samples were scored for overall visual quality using an interval hedonic scale, where the extremes and center of the interval were represented as follows: 0 'very bad', 5 'limit of acceptance from the consumers point of view', and 10 'very good'. The remaining attributes, flavor and firmness, were evaluated in a 5 point scale, where 5 = fully characteristic of the product, 2.5 = moderate and 0 = not characteristic. Defects of the product, off-odors, browning and dehydration were evaluated as follows: 5 = severe, 2.5 = moderate and 0 = absence. The samples were coded with random three-digit numbers to mask the treatment identity in order to minimize subjectivity and to ensure test accuracy. All quality evaluations were performed in a sensory room.

j) Statistical analysis

For statistical studies, SPSS 18.0 software was used (SPSS Inc., Chicago, IL, USA). Correlations were estimated with the Pearson test at p<0.05 significance level. Data are expressed as means ± S.D. and were analyzed using a one-way analysis of variance (ANOVA). When ANOVA detected significant differences between mean values, means were compared using LSD Tukey's test.

III. RESULTS AND DISCUSSION

a) Total soluble solids (TSS), titratable acidity (TA) and Ph

The results are presented in Figure 1. There were not some significant differences in TSS during storage days. The values for TSS and TA found in this study are similar to others given for this fruit (2, 13). The pH of studied strawberries was very low (about 3.5) and during the storage did not change significantly ($p > 0.05$); the found values of pH were within the limits pH 3.0 to 5.0 that promote the processes of copigmentation (1).

b) Colour

The value of a^* parameter offers better the evidence of deterioration or browning of the product, so a positive a^* value indicates redness on the hue circle and $-a^*$ indicates greenness. The results of the evolution of the parameter a^* are shown in Figure 2. In our study, in general, a^* showed not any significant differences during storage days so, we can conclude that when UV radiation is used in water as washing treatment, the pigments conservation is good. The evolution for a^* found in this study has a similar behaviour to other given for minimally processed strawberries treated with chitosan as preservative (2).

c) Head space gas composition

As one would expect a decrease in the headspace oxygen concentration along with an increase in the headspace carbon dioxide concentration was detected. The package headspace was monitored over 10 days of storage (data not shown). Starting from an atmospheric gas concentration, levels of 18-19 % O_2 and 3-4 % CO_2 within packages were established after 5 days and until the end of the storage time at 4 °C. In this study, the values found for O_2 and CO_2 levels imply that the PP film could be adequate for passive modified atmosphere packaging of fresh cut processed *Fragaria vesca* strawberries.

d) Phenolic content, anthocyanin pigments and antioxidant activity

The results are shown in Figure 3. Fresh cut strawberries exhibited high amounts of phenolic compounds and showed not any significant differences during all storage days. This values are in agreement with the values found by other authors in the whole *Fragaria vesca* strawberry (4) and for other whole cultivars (13). In this way, other authors (7, 28) reported a concentration of total phenolics between 1730 and 3180 mg kg^{-1} in different strawberry cultivars determined by the Folin-Ciocalteu assay. Calculated total phenolics as the sum of each individual phenolic compound quantified by HPLC (23), the values ranging from 639 to 660 mg kg^{-1} . The higher phenolic concentrations found in our study, compared to those obtained by HPLC,

could be attributed to the analytical method used to determine these compounds. The Folin-Ciocalteu reagent usually overestimates the content of phenolic compounds compared with the sum of the individual phenolics, since other reducing agents present in food, such as ascorbic acid, can interfere (22).

Anthocyanins are responsible for the attractive red colors of strawberries. The initial concentration of total anthocyanins in strawberries agrees with the reports of other authors for whole *Fragaria vesca* strawberries (4) and for other whole cultivars (13). A slightly decrease was observed for the evolution of anthocyanin content during storage but showed not any significant differences.

Several investigations have reported significant differences in antioxidant activity among strawberry cultivars (17). In our study, fresh cut strawberries exhibited high total antioxidant activity during all storage days. Higher values were found by other authors for this cultivar (18). *Fragaria vesca* fruits were 2.5 times more active than cultivated strawberries in the TEAC assay (Trolox equivalent antioxidant capacity) (26). A slightly decrease was observed for the evolution of total antioxidant activity content during storage but not any significant differences were found. This decrease during storage period at 4 °C might be due to an effect on the activity of the major enzymes involves in the functional compounds degradation.

e) Microbiological assessment

The increase in cut damaged surface and availability of cell nutrients, in fresh-cut products, provide conditions that increase the number and types of microbes that can be develop. The specifications proposed by E.C (25) were used to determine the end of the shelf-life from the microbiological point of view, which are: 6 log CFU g^{-1} for total aerobic count and mesophiles. Figure 4 indicates a slight decrease of approximately 1.5 log unit CFU g^{-1} in mesophiles, psychrotrophs and yeast and moulds microbial growth after 10 days of storage when the UV radiation in the washing water was used. Higher values of psychrotrophic bacteria were found by other authors (2) for control samples of minimally processed strawberries. Previous results in our laboratory (20) concluded that the use of UV causes a microbial reduction in washing water, with possibility of reuse this water with the consequent minimization of water consumption and decrease in the wastewater discharge rates in the food industry, producing a good environmental impact.

f) Firmness determination

The values found are practically constant during all days of storage, not significant differences were found, however, there is a slightly decrease from day 5 due to that the minimal processing alters the integrity of the fruit (Figure 5) and induces surface damages increasing slightly the tissue respiration and leading

biochemical deteriorations such as texture breakdown (16).

g) Effect of washing treatment on sensory quality

The obtained results for the sensory evaluation are shown in Figure 5. The visual quality of minimally processed strawberries was excellent after washing and decreased slightly during storage. The same behaviour was found for promotion of browning in the cutting area. These samples maintained the typical flavour during storage. An off-odor was not detected in washed samples at any storage time, without evidence of anaerobic fermentations after 10 days of storage at 4 °C. The firmness of strawberries slightly decreased during storage but a day 10, all samples maintained a moderate crispy texture without significant differences among them. Therefore, water + UV radiation did not affect the sensory quality of fresh-cut strawberries during storage.

IV. CONCLUSIONS

It was concluded that the use of UV-C radiation in the washing water was effective in reducing the microbial counts, maintaining the antioxidant compounds and the sensorial quality of the fresh-cut strawberries during the 10 days at 4 °C. This treatment could be a good sanitizer and alternative technique for minimizing water consumption in the food industry.

V. ACKNOWLEDGEMENTS

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Figures

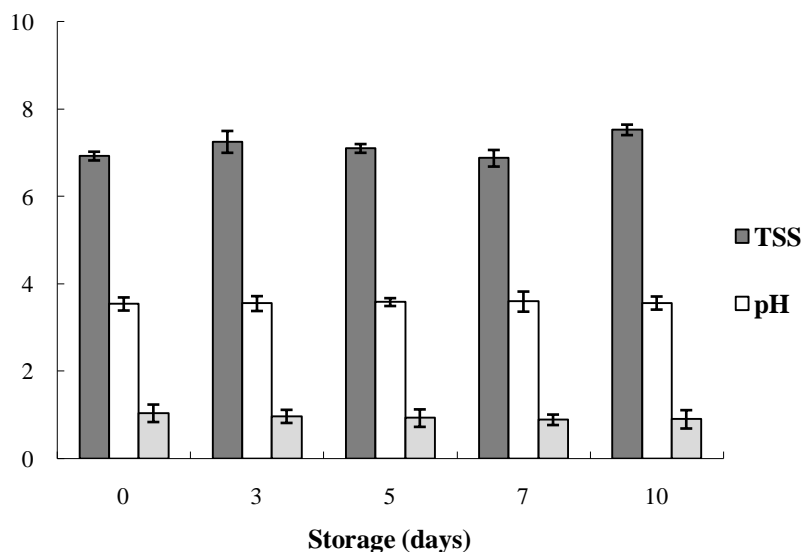


Figure 1 : Evolution of total soluble solids (TSS), titratable acidity and pH of fresh cut strawberries throughout the storage at 4 °C for 10 days. Values are the mean of four replicates and vertical bars represent the Standard Deviation. TSS (°Brix); acidity (g malic acid 100 g⁻¹ fresh weight (fw))

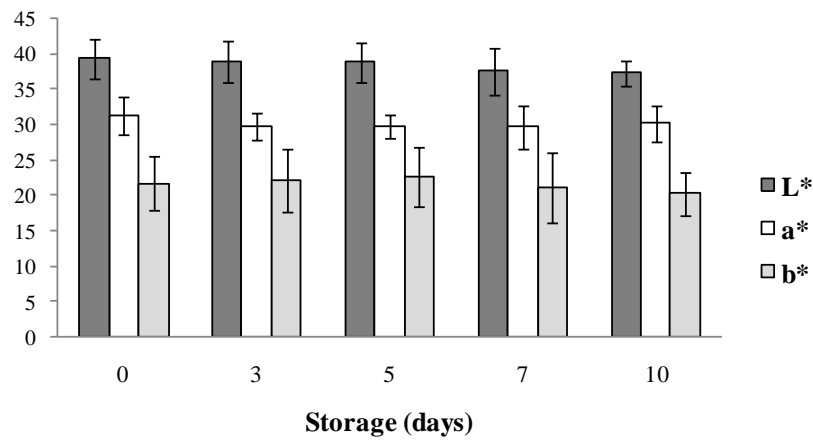


Figure 2 : Evolution of the colour parameters (L*, a* and b*) throughout the storage time at 4 °C in fresh-cut *Fragaria vesca* strawberries stored at 4 °C for 10 days. Values are the mean of four replicates and vertical bars represent the Standard Deviation

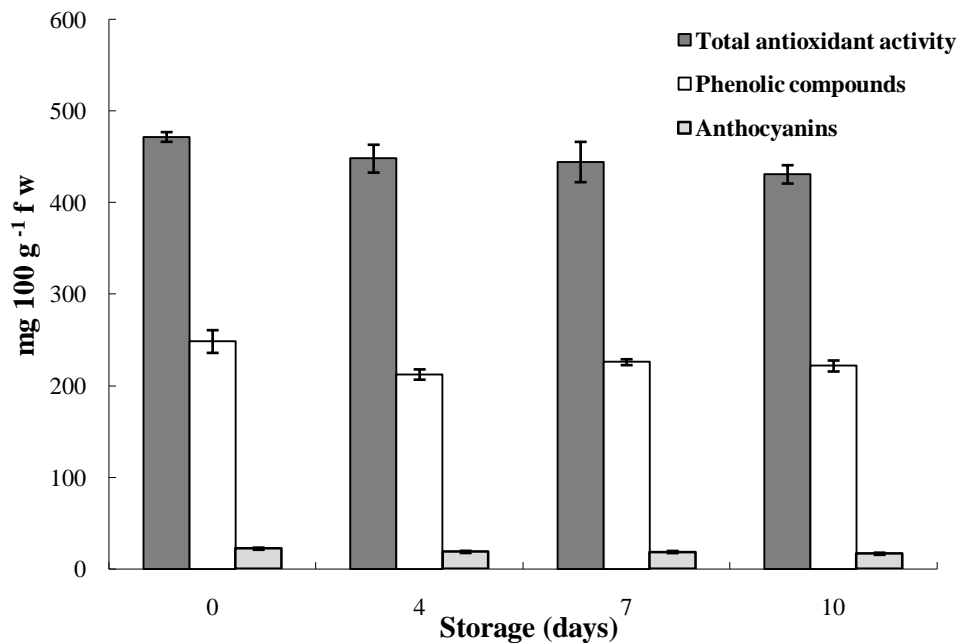


Figure 3 : Evolution of total phenolic content, anthocyanins content and total antioxidant activity of fresh cut *Fragaria vesca* strawberries at 4 °C throughout the storage. Values are the mean of four replicates and vertical bars represent the Standard Deviation. Total antioxidant (mg Trolox 100 g⁻¹ fw); Phenolic compounds (mg galic acid 100 g⁻¹ fw); Anthocyanins (mg cyaniding 3-O-rutinoside 100 g⁻¹ fw)

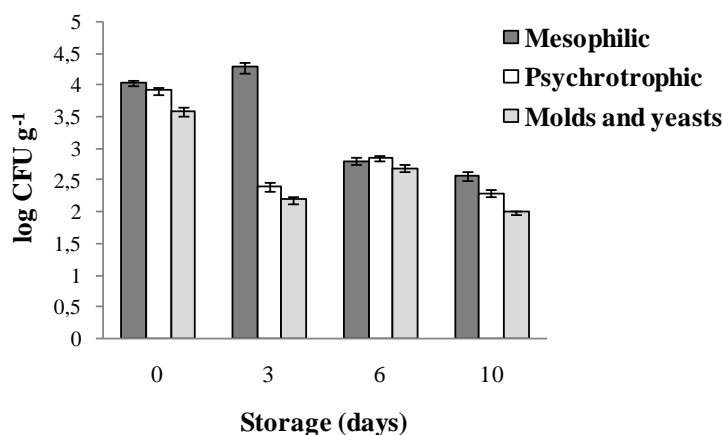


Figure 4 : Evolution of mesophilic, psychrotrophic, molds and yeasts (log CFU g⁻¹) counts in fresh-cut *Fragaria vesca* strawberries stored at 4 °C for 10 days. Values are the mean of four replicates and vertical bars represent the Standard Deviation

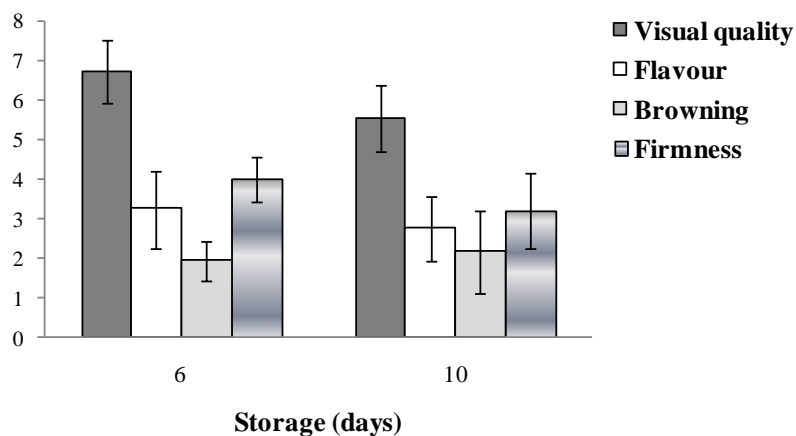


Figure 5 : Firmness and sensory quality of minimally processed *Fragaria vesca* strawberries stored at 4 °C for 10 days. Values are the mean of four replicates and vertical bars represent the Standard Deviation

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