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Towards Drylands Biorefineries: Valorisation of Forage *Opuntia* for the Production of Edible Coatings

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Abstract: Species of the genus *Opuntia* may be a well-suited feedstock for biorefineries located in drylands, where biomass is scarcer than in humid or temperate regions. This plant has numerous uses in Mexico and Central America, and its mucilage is a specialty material with many promising applications. We extracted the mucilage from a forage species, *O. heliabravoana* Scheinvar, and mixed it with a thermoplastic starch to produce an edible coating. The coating was applied to blackberries, which were then evaluated in terms of several physicochemical and microbiological variables. During a 10-day evaluation period, the physicochemical variables measured in the coated fruits were not significantly different from those of the control group. However, the microbiological load of the coated fruits was significantly lower than that of the uncoated fruits, which was attributed to a decreased water activity under the edible coating. Multivariate analysis of the physicochemical and microbial variables indicated that the storage time negatively affected the weight and size of the coated and uncoated blackberries. Although some sensory attributes have yet to be optimised, our results support the use of the mucilage of forage *Opuntia* for the formation of edible coatings, as well as their valorisation through a biorefinery approach.

Keywords: biomass; cactus; food waste; mucilage; shelf-life; food innovation; sustainable biorefinery

1. Introduction

Biorefineries are increasingly seen as a feasible source of chemicals, materials, bioenergy, and biofertilisers, that can contribute to the energy sovereignty and the economy of both developed and developing countries. To be sustainable, a biorefinery must be based on biomass feedstocks that are renewable and widely available and that do not compete with food crops for water and fertile soil, such as agricultural and forestry residues, municipal and industrial waste, or perennial plants grown on degraded lands. These biomass feedstocks are particularly well suited for biorefineries located in drylands, because in these zones the availability of biomass and the variety of agricultural activities are limited compared with humid or temperate regions [1].

In Mexico, products derived from *Opuntia* spp. (mainly vegetables, fruits and forage) have been used since 9000 B.C. [2]. In particular, *O. ficus-indica* produces edible cladodes that are highly regarded, and consumed as vegetables. This is one of the most economically important crops in this country and in Central America, and its range of industrial uses spans from food additives to cosmetics to adhesives.

The main advantage of *Opuntia* spp. lies in the fact that it exhibits crassulacean acid metabolism, or CAM photosynthesis, thereby improving the efficiency of water use and carbon assimilation under conditions of low water availability. This crop also presents high biomass productivity, adaptation to changing climate conditions, rapid growth, and minimal nutrient input. Accordingly, *Opuntia* spp. has potential for valorisation in drylands through the production of methane [3], bioethanol [4,5], and specialty chemicals [6].

In particular, its mucilage presents physicochemical and rheological properties that give it promising food, pharmaceutical, cosmetic, and environmental applications [7,8]. The mucilage is a heteropolysaccharide of high molecular weight that can contain over 30,000 subunits, mostly arabinose, galactose, rhamnose, xylose, and uronic acids. It generates gels that retain large amounts of water and have good emulsifying characteristics. Among other applications, the mucilage of *O. ficus-indica* has been employed to produce edible films with good mechanical properties [9,10]. Edible films and coatings are defined as any material used for encapsulating food to extend the shelf-life of the product, that may be eaten with the food with or without removal of the material [11]. The difference between the two materials lies in the way they are applied: films are structures formed separately and then applied to the food surface, while coatings are formed directly on the food surface [12].

In Mexico, approximately 37% of food produced is wasted; for fruits, this percentage is greater than 50% [13]. Edible films and coatings represent a sustainable way to increase the shelf-life of numerous products, minimise food waste, and eventually reduce the amount of petroleum-based synthetic packaging used [12,14]. However, reduction of the environmental impact of *Opuntia*-derived edible films and coatings involves their production from species not used for human consumption.

The aim of this work was to valorise an emblematic drylands plant, *O. heliabravoana* Scheinvar, which is only used as cattle feed, to produce an edible coating. The coating was characterised physicochemically and was applied and tested on blackberries, a highly perishable fruit, through physicochemical and sensory analyses. The environmental impact of a baseline scenario, i.e., landfilling of the crop instead of its valorisation, was also evaluated through the calculation of greenhouse gas (GHG) emissions by a simulation tool.

2. Materials and Methods

2.1. Biological Materials

O. heliabravoana Scheinvar is an endemic species of Mexico. It abounds in Hidalgo State, where it is found as perennial shrubs or creeping plants. Neither its cladodes nor its fruits are exploited commercially [15]. Samples of one-year-old cacti were manually obtained from Mineral de la Reforma, Hidalgo, Mexico (N 20°4′20.39″, O 98°44′57.43″). The samples were processed no more than 36 h after collection. Blackberries (*Rubus ulmifolius*) used to test the edible coatings were obtained from a local market.

2.2. Extraction of Opuntia Mucilage

The mucilage was extracted according to Sepúlveda et al. [16], with some modifications. After removing the spines, the cladodes were washed, cubed (1 cm^3) and heated in water (1:4 w/v) at 80 °C for three hours. Then, the cladodes were removed, and the liquid obtained was centrifuged (3500$\times$ g for 20 min). The supernatant was recovered, and the mucilage was precipitated by adding 96% food-grade ethanol (1:3 v/v). The mucilage was oven-dried at 70 °C for two hours, and ground in a mortar.

2.3. Preparation of the Edible Coatings

The edible coatings were prepared following Schlemmer et al. [17]. First, starch, water, and glycerol were mixed in a 50:15:35 ratio ($w/w/w$) for 15 min. They were transformed into thermoplastic starch (TPS) by heating at 95 °C in a water bath with continuous stirring for 30 min. TPS was mixed with two different mucilage solutions (1% and 3%) in different ratios (1:9, 9:1 w/w), leading to four mixtures. Each mixture was heated for three hours at two different temperatures (50 °C and 80 °C) in a water bath, thereby producing eight coating-forming formulations. The viscosity of each was measured using a Brookfield LVT viscometer. Blackberries were coated to observe the appearance of the fruit after application of each of the eight coating-forming solutions, and to choose the most appropriate formulation.

2.4. Physicochemical Characterisation of the Materials

Both the mucilage and the edible coating were characterised by proximate and elemental analyses, scanning electron microscopy (SEM), and thermogravimetric analysis (TGA). For the proximate analysis, the contents of moisture, ash, and fibre were determined in triplicate samples by methods 934.01, 942.05, and 978.10 of the AOAC, respectively [18]. The protein quantity was analysed using the Bradford method [19]. The nitrogen-free extract (NFE) was obtained by subtracting the percentages calculated for each nutrient from 100 [20]. The elemental composition was determined using a 2400 Series II CHNS Elemental Analyser (Perkin–Elmer™, Waltham, MA, USA). The surfaces of the samples were studied by SEM (JSM-6010LA InTouchScope). The samples were fixed on aluminium supports using double-sided adhesive carbon tape, and covered with gold. The SEM images were obtained at 15 kV. TGA was performed using Mettler Toledo equipment, model TGA/SDTA 851e (Switzerland). Approximately 3–6 mg of sample was positioned in an alumina cell and heated from 30 to 600 °C at a heating rate of 10 °C/min. The analysis was performed under nitrogen flow at 50 mL/min.

2.5. Physicochemical and Microbiological Evaluation of the Coated Fruits

The fruits were washed and disinfected with a 200 mg/L solution of sodium hypochlorite and then separated into two batches: one batch was tagged as the control, and the fruits in the other batch were coated by immersion in the coating-forming solution. Both batches were stored at 0 °C in a refrigeration chamber for 10 days. Weight loss (WL), size, pH, soluble solids content (SSC), colour, and microbial load were evaluated in both batches.

Weight loss (WL) was determined by recording the weight of a given sample from both batches daily, and expressed as the percentage loss of the initial weight. In the same way, longitudinal and transversal measurements were made with a Vernier to determine the surface size (S, cm^2) of given samples from the control group, and from the batch of coated fruits. The pH and SSC were measured in the juice obtained from a sample using a pH meter (Hanna Instruments pH 210, Ann Harbor, MI, USA) and an ABBE refractometer (Reichert AR200, Depew, NY, USA), respectively. Colour determination was performed with a colorimeter (MiniScan XE PLUS model 45/0-L, Reston, VA, USA), using CIELAB as the colour space. The obtained colour coordinates were L (luminosity), a (redness), b (yellowness), and c (chroma), but only L was used for monitoring the fruits during storage. The microbial load (ML) was determined by measuring the content of yeasts according to the Mexican standard technique [21]. All the determinations were carried out in triplicate.

2.6. Sensory Evaluation of the Coated Fruits

To determine the acceptability of the coated fruits, a sensory evaluation was performed after 0 and 6 days of storage using a five-point hedonic scale (5 = I like it a lot, 4 = I like it, 3 = I neither like it nor dislike it, 2 = I dislike it, and 1 = I dislike it a lot). Brightness, colour, and flavour were evaluated by 30 untrained panellists; no specific criteria were taken into consideration (age or sex).

2.7. Statistical Analyses

We used multivariate statistical analysis (Pearson correlation and factor analysis, FA) to evaluate the interactions between the physical and chemical characteristics of blackberries and the two applied treatments. The normal distribution of the data was verified by the Kolmogorov-Smirnov and Shapiro-Wilk tests. Pearson correlations were also calculated ($0.01 \leq p \leq 0.05$) to identify the minimal significance of the senescence stage, but only the 0.05 significance level was further used to assess the loadings to be considered in FA [22]. Finally, FA with Varimax rotation was conducted either with the original data set or with standardised data, according to the method of Lucho-Constantino et al. [22].

We evaluated the effect of the organoleptic characteristics (brightness, colour, and flavour) on the blackberries samples using a Latin square design test ($n = 30$); $0.05 \leq p \leq 0.10$; i (two treatments: with and without edible coating); j (five-point hedonic scale: 5 = I like it a lot, 4 = I like it, 3 = I neither like it nor dislike it, 2 = I dislike it, and 1 = I dislike it a lot) at two shelf times, 0 and 6 days. Additionally, we used a post hoc multiple comparison test (least significant difference, LSD) to determine the significant differences ($0.01 \leq p \leq 0.05$) of the means of the variables in the sensorial analysis (between the organoleptic characteristics and the different applied treatments, with and without edible coating). The statistical package SPSS v21 (SPSS Inc., Chicago, IL, USA) was used to perform this analysis. Graphs showing the effect of storage time on the study variables were created in Excel 2010 v14.0, Microsoft Office™.

2.8. Analysis of the Environmental Impact of the Valorisation of Forage Opuntia

The environmental impact of a baseline scenario, i.e., landfilling of the crop instead of its valorisation, was estimated from the CO_2 emissions from the amount of *Opuntia* required for the production of ten kilograms of mucilage per day. For this, we used the open-source EPA Waste Reduction Model (WARM) v14. According to the U.S. Environmental Protection Agency (U.S. EPA) [23], this model calculates the greenhouse gas (GHG) emissions "for baseline and alternative waste management practices, including source reduction, recycling, combustion, composting, and landfilling". In the WARM worksheet, the option of "*Opuntia* waste" does not appear as an input, so we chose the "fruits and vegetables" option instead. The fruits and vegetables category includes "a wide variety of cultivars produced worldwide, all with widely varying inputs, processing stages, and transportation distances", and the emission factors are calculated as a mixed weighted average, which represents the relative contribution of diverse fruits and vegetables to the total waste stream in the USA [23]. Further explanation of the methodology is provided by the WARM documentation [24]. The equivalencies of the GHG emissions given by the WARM model were also calculated [25].

3. Results and Discussion

3.1. Mucilage Extraction

The mucilage was extracted from the cladodes of *O. heliabravoana* Scheinvar in nine batches. The dry end product was a homogenous, opalescent, beige-coloured powder. The extraction yield was 0.30 ± 0.08%, which is notably lower than previously reported values. Ruíz [10] obtained an extraction yield of 0.44%, while Sepúlveda et al. [16] reported a mean extraction yield of 1.2%, both from *O. ficus-indica*. The variability in the yield may be due to differences in the used species and extraction methods, as well as in their age and environment. These last factors are particularly important because the function of mucilage in the plant is to buffer climate fluctuations, which can be extreme in arid environments. Consequently, long drought periods could induce the overproduction of mucilage to further protect the plant [26].

3.2. Preparation of the Edible Coatings

Eight coating-forming formulations were prepared with different proportions of mucilage and TPS, and then heated at two different temperatures. The viscosity of six of the formulations and the appearance of the coated fruits are shown in Figure 1. Two of the formulations were too viscous and were discarded (formulations 2 and 6 in Figure 1). The solution chosen for further use was the mixture composed of nine parts mucilage solution (1%) and one part TPS, heated at 50 °C (formulation 3 in Figure 1). This formulation was chosen because it had the lowest viscosity (87.5 cps) and provided the most homogeneous coverage of the fruits. The berries coated with this formulation also had the best general appearance.

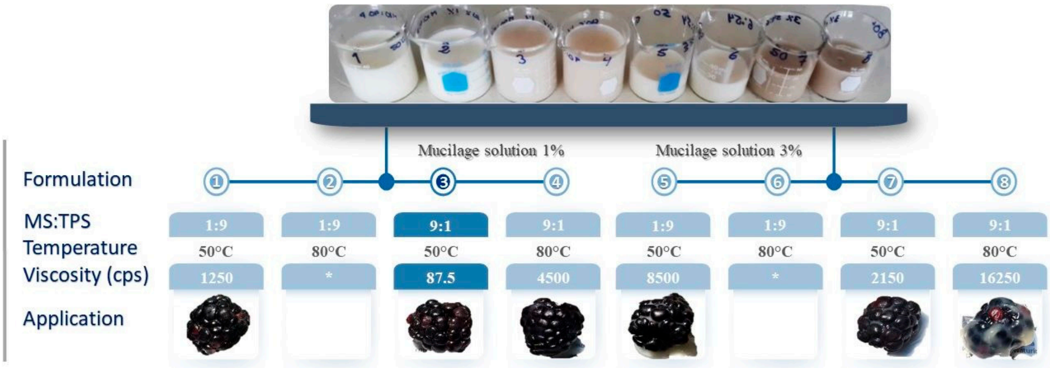

Figure 1. Preparation of the edible coatings from eight formulations. MS: mucilage solution; TPS: thermoplastic starch. The asterisks identify the discarded solutions for which the viscosity was not determined and which were not used to cover the fruits.

3.3. Physicochemical Characterisation of the Mucilage and the Edible Coating

The mucilage extracted from *O. heliabravoana* Scheinvar and the edible coating prepared from this mucilage were characterised physicochemically via proximate and elemental analyses; the results are shown in Table 1. The average moisture content in the mucilage was 7.75%, which is lower than that reported by Espino-Díaz et al. [9] and Rodríguez-González [27], who obtained mean values of 12.43% and 9.31%, respectively, in the mucilage of *O. ficus-indica*. These differences may be due to the storage temperature and drying time, since the mucilage is highly hygroscopic. The ash content in the mucilage was 13.14%, which is similar to the values reported by Rodríguez-González et al. [28] and Rodríguez-González [27] for the mucilage of *O. atropes* and *O. ficus-indica*, i.e., 11.91% and 14.01%, respectively. The protein content in the mucilage was 1.11%, which is quite similar to the value measured by Espino-Díaz et al. [9] in *O. ficus-indica* (1.04%) using the same analytical method. Finally, the crude fibre content was 55.92%, which is also similar to the values (57.71% and 57.23%) measured in the mucilage of *O. atropes* and *O. ficus-indica*, respectively, by Rodríguez-González et al. [28] and Rodríguez-González [27]. The nitrogen-free extract (NFE) includes mostly soluble organic compounds (as starch and sugars). The value estimated for the mucilage (22.08%) is slightly higher but still similar to the range of values reported for other *Opuntia* species (1.52–17.01%) [28]. Quantification of the ethereal extract was not performed because the values found were less than 0.1% [28].

Table 1. Results of the proximate and elemental analyses of the mucilage and the edible coating. Data are the mean ± the standard deviation of three independent determinations.

Material	Proximate Analysis					Elemental Analysis		
	Moisture	Ash [1]	Protein [1]	Dietary Fibre [1]	Nitrogen-Free Extract (NFE) [1]	C	N	H
Mucilage	7.75 ± 1.15	13.14 ± 1.58	1.11 ± 0.01	55.92 ± 1.76	22.08	36.91	<1.0	6.00
Edible coating	93.82 ± 1.69	1.21 ± 0.11	<0.01 ± 0.003	0.24 ± 0.04	4.73	37.66	<1.0	7.65

[1] % calculated on a dry weight basis.

Elemental analysis indicated the mucilage was composed of 36.91% carbon, 6.0% hydrogen and less than 1% nitrogen. The last value is consistent with the low protein content in the plant. These results are similar to those reported by Trachtenberg and Mayer [29], who obtained values of 42.57% for carbon and 6.31% for hydrogen, discarding the content of nitrogen.

The edible coating presented a high moisture content (93.82%; Table 1), which was expected since water is the main component of the coating. The low contents of ash and fibre (1.21% and

0.24%, respectively) are due to the small proportions of starch and mucilage contained in the coating, which contribute these components (Table 1). In the same way, the low protein concentration in the edible coating is related to both the low proportion of mucilage in the coating and the reduced protein content in the mucilage. A value of 4.73% was obtained for the NFE, mainly provided by the starch.

Surface optical micrographs obtained for the mucilage and the edible coating are shown in Figure 2a,b, respectively. The observed surface morphology of the coating (Figure 2b) showed no cracks or pores, resulting in the formation of a dense and homogeneous coating.

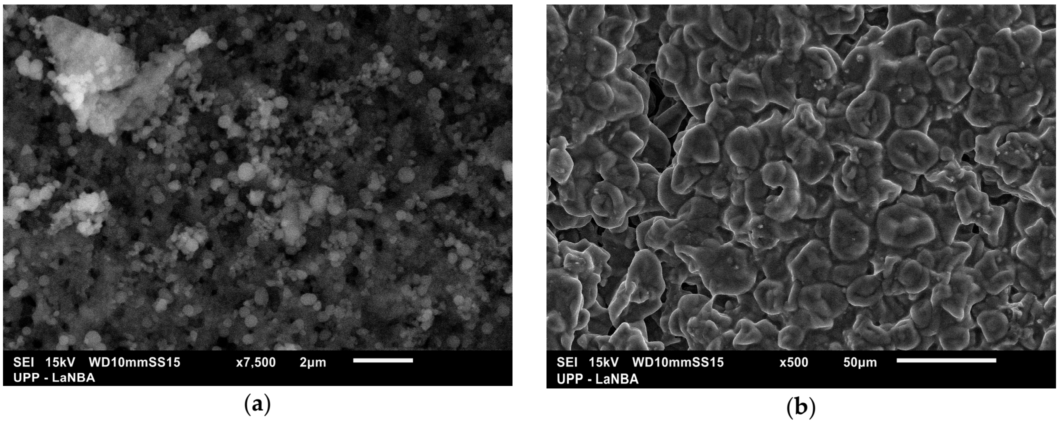

Figure 2. SEM micrographs of (**a**) The mucilage of *Opuntia heliabravoana* Scheinvar; (**b**) The surface of the edible coating.

TGA curves of the mucilage and the edible coating are shown in Figure 3a,b, respectively. The first weight loss stage was attributed to the removal of free water, and values of 5.1% and 4.45% were measured in the mucilage and the edible coating, respectively. For mucilage, the second stage began at 227.52 °C and corresponded to the decomposition process, which resulted in a loss of 51.2% of the total mass (Figure 3a). The second stage for the edible coating was detected at 134.58 °C (with a maximum at 195.60 °C). This stage was attributed to the evaporation of glycerol from the coating. Additionally, a third stage for the edible coating was observed at 213.38 °C, corresponding to the decomposition process, after which 74.1% of the sample mass was lost (Figure 3b).

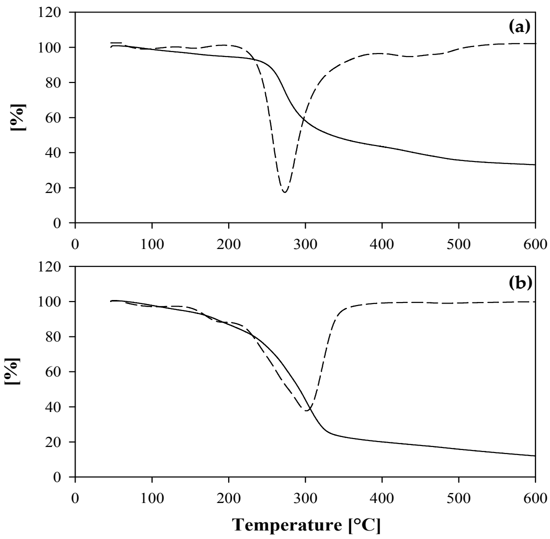

Figure 3. Thermogravimetric (continuous lines) and derivative (discontinuous lines) curves of (**a**) the mucilage of *Opuntia heliabravoana* Scheinvar; (**b**) the edible coating.

3.4. Physicochemical and Microbiological Evaluation of the Coated Fruits

Figure 4 shows the changes in quality of both the coated and uncoated blackberries during the storage period. The quality of the fruits was assessed in terms of weight loss (WL), size (S), luminosity (L), pH, soluble solids content (SSC), and microbial load (ML).

The coated and uncoated blackberries lost weight throughout the ten days of storage at almost the same rate (Figure 4a). After this period, the coated blackberries had lost 21.44 ± 0.31% of their initial weight, while the control blackberries had lost 19.41 ± 2.42%. No significant difference was found between treatments ($p \leq 0.05$) concerning this variable. The weight losses registered in this study appear to be higher than those in previous studies. Pérez-Gallardo et al. [30] measured a WL of 7.6 ± 0.13% in uncoated blackberries and of 9.72 ± 0.42% in blackberries coated by a starch-beeswax emulsion after a 16-day storage period at 4 °C and 88% relative humidity. In another work, a WL of 9% after 18 days of storage at 0 °C was reported for uncoated blackberries [31]. The differences in the reported weight losses are likely to arise from differences in the physicochemical environments, such as the temperature or humidity during storage. In fact, for two series of experiments carried out at 0 and 10 °C, Oliveira et al. [32] determined weight losses of approximately 8% and 32%, respectively, for uncoated blackberries after a storage period of 12 days. The edible coatings prepared by these authors—made of chitosan, cassava starch, and kefir grains—proved to be effective in reducing the WL at both storage temperatures.

Although carbohydrate-based edible coatings are inexpensive, give fruits a good appearance, and are non-toxic to humans, they present a high water permeability, which leads to significant moisture loss [33]. Most fruits and vegetables lose a significant fraction of weight in the form of water vapour due to the transpiration process. This is the main cause of physiological deterioration in fruits, because it leads to a diminution in the organoleptic and nutritional quality of the product [34]. A WL of 6% is the maximum permissible value in fruits [35]; in our experiments, both coated and uncoated blackberries reached this value after 2 days.

Concerning the measurements of S (expressed in terms of surface, Figure 4b), the results indicated a progressive reduction in both groups, although this reduction was faster in the control fruits. After 10 days of storage, the S reduction of the control group was more than 40%, while the S of the coated fruits diminished by approximately 26%. This difference was found to be significant at $p \leq 0.1$ but not at $p \leq 0.05$.

Figure 4c shows the pH of the fruits measured during storage. At the beginning of the experiment, blackberries presented pH values (3.09 and 2.97 for the coated and uncoated fruits, respectively) comparable to those suggested by Tosun et al. [36] as representative of mature blackberries (i.e., 3.14). During the storage period, the pH values of both groups increased to a similar extent (to 3.55 and 3.54 for the uncoated and coated fruits, respectively). The increase in the pH is attributed to the natural process of fruit senescence; during this process, the content of organic acids declines, as they are either respired or converted to sugars [36]. The pH values obtained for both groups after the 10 days of storage were not significantly different ($p \leq 0.05$).

The effect of the edible coating on the SSC throughout the storage period is shown in Figure 4d. At the beginning of the experiment, the uncoated and coated blackberries presented a mean SSC of 8.1 and 8.8, respectively. The initial SSC of the control fruits (8.1) was comparable to the values for uncoated blackberries reported by Oliveira et al. [32], which were between 7.5 and 8.3. In our study, the SSC increased in the uncoated and coated blackberries throughout the storage period, following a pattern similar to that observed for the pH (Figure 4c). This increase in the SSC is possibly related to the moisture loss observed during the storage period. In contrast, Oliveira et al. [32] signalled an important decline in the SSC of uncoated blackberries after 12 days of storage at 10 °C. These authors also detected a significant diminution in the SSC of blackberries coated with edible films based on chitosan, cassava starch, and kefir grains after 12 days of storage under refrigeration at 10 °C.

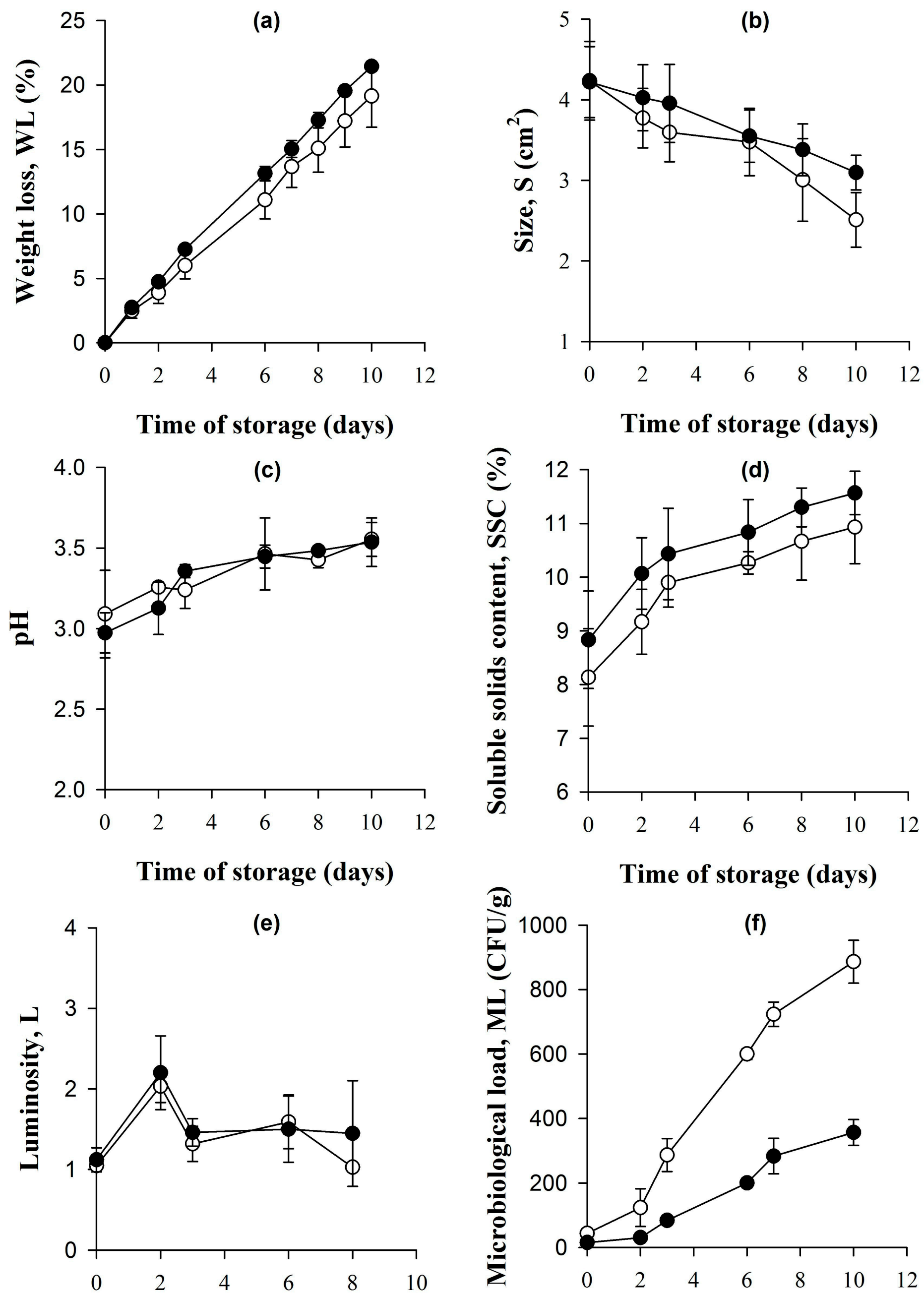

Figure 4. Changes in quality of the uncoated (○) and coated (●) fruits: (**a**) weight loss (WL); (**b**) size (S); (**c**) pH; (**d**) soluble solids content (SSC); (**e**) luminosity (L); (**f**) microbiological load (ML). Data points and error bars represent means and standard deviations, respectively, based on three independent samples.

The results of the luminosity (L) measurement throughout the storage period are shown in Figure 4e. This parameter indicates the darkness (0) or lightness (100) of the colour, and can be used as a surrogate for colour [37]. As coated fruits presented statistically similar L values to the control fruits directly after being coated, it can be stated that the edible coating did not modify the natural luminosity of the fruits. The L values increased thereafter and reached their maximal values at day 2 in both the uncoated and coated fruits. Afterwards, the lightness diminished and remained at relatively similar levels throughout the rest of the storage period for the two groups of fruits. After ten days, no significant differences in luminosity ($p \leq 0.05$) were found.

Blackberries are a highly perishable fruit due, on the one hand, to the fast changes in their physicochemical properties and, on the other hand, to the fast growth of moulds and yeasts (such as *Botrytis cinerea* or *Alternaria* spp.) that occurs during the postharvest period [38]. Figure 4f depicts the growth of yeasts on the uncoated and coated blackberries during storage at 4 °C. Although a certain amount of microbial growth was observed in both groups, a significant difference ($p < 0.05$) between them was found after only two days of storage. This significant difference remained for the rest of the experiment.

The lower microbial counts in the coated fruits may be due to the *Opuntia* mucilage added to the edible coatings. On the one hand, there is evidence that this plant contains compounds related to CAM photosynthesis, such as camphor, flavonoids, and other aromatics, which hinder the growth of some microorganisms. For instance, Ortiz-Rodríguez et al. [39] studied the microbiological quality of raw milk supplemented with the mucilage of *O. ficus-indica* at 0.5, 1 and 2% (v/v). These authors found that the addition of mucilage diminished the contents of mesophilic aerobic bacteria and of total coliforms after two hours of contact, which was attributed to the aforementioned compounds. On the other hand, the mucilage is a hydrocolloid that strongly retains water by the formation of high-molecular-weight gels. Thus, in the edible coating, it could act as an inhibitor of microbial growth by diminishing water activity. This property of the mucilage contributed to increasing the shelf-life of the coated blackberries by about two days. However, further research must be done to confirm the antimicrobial activity of the mucilage in the edible coating.

3.5. Results of the Multivariate Analysis of the Characteristics of the Coated Fruits

A factorial analysis (FA) was carried out to determine the relationships between storage time and some of the parameters (WL, S, pH, SSC, and ML) evaluated in the coated and control fruits (denoted with the subscripts "c" and "u", respectively). The S and ML values were log transformed to normalise them. The results of the analysis are shown in Figure 5.

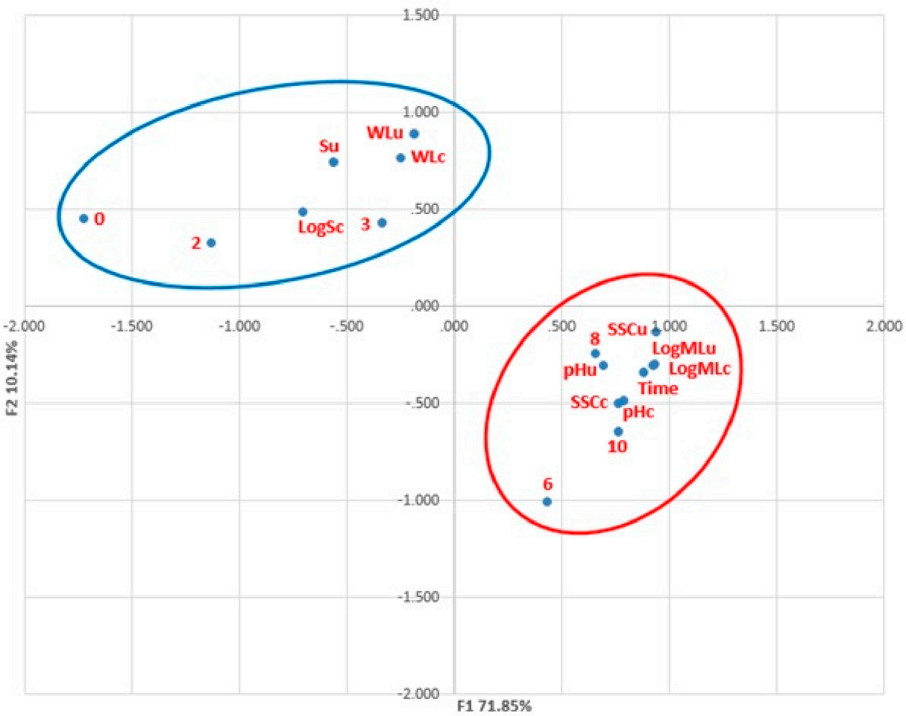

Figure 5. Loading plot (F1 vs. F2) from the multivariate analysis of the storage time and the parameters measured in the uncoated fruits (denoted with the subscript u) and the coated fruits (denoted with the subscript c).

The FA explains 81.98% of the total variance with two factors, while F1 explains by itself 71.85%. F1 was contributed to positively by the storage time, pH_u, pH_c, SSC_u, SSC_c, $LogML_u$, and $LogML_c$, while it was negatively contributed to by $LogS_c$ and S_u. F1 mainly involved changes in some of the fruit quality parameters (pH, SSC, ML) that occurred during storage independently of the application of the edible coating. The changes in pH and SSC are due to the loss of humidity, and therefore, of weight. Thus, this "senescence factor" can be linked to the effect of the shelf-life on the characteristics of the berries, particularly after the sixth day of storage.

F2 explains 10.14% of the total variance and shows a positive association between S_u, $LogS_c$, WL_u, and WL_c during the first three days of storage. Consequently, F2 can be also associated with the senescence of the fruits. The above variables showed no statistically significant differences ($p \leq 0.05$) in any of the treatments. F2 was contributed to negatively by SSC during the first three days of storage (Figure 5).

3.6. Results of the LSD Statistical Analysis of the Sensory Evaluation of the Coated Fruits

The main results of the sensory evaluation of the uncoated and coated berries are presented in Table 2. At the beginning of storage, the brightness of the fruits in both groups was significantly different ($p \leq 0.05$), as determined from the scores of the hedonic scale "I like it a lot" and "I like it" against "I dislike it" and "I dislike it a lot" ($0.01 \leq p \leq 0.05$). More than 90% of the panellists (n = 30) perceived the brightness of the blackberries (coated or not) as acceptable (i.e., taking together the scores 5 to 3: "I like it a lot", "I like it", and "I neither like nor dislike it") at the beginning of storage.

Table 2. Results of the sensory evaluation of the uncoated and coated blackberries before and after a 6-day storage period.

Attribute	Days (D)	Treatment (T)		Mean	Standard Error	Significance (T)
		Uncoated	Coated			
Brightness	0	4.03	3.50	3.77	0.27	s
	6	4.17	2.70	3.43	0.73	s
	Mean	4.10	3.10			
	Standard error	0.07	0.40			
	Significance (D)	ns	s			
Colour	0	4.07	3.90	3.98	0.83	ns
	6	4.13	3.00	3.57	0.57	s
	Mean	4.10	3.45			
	Standard error	0.03	0.45			
	Significance (D)	ns	s			
Flavour	0	3.53	3.67	3.60	0.70	ns
	6	3.40	3.37	3.38	0.02	ns
	Mean	3.47	3.52			
	Standard error	0.07	0.15			
	Significance (D)	ns	s			

s: significant; ns: non significant ($0.01 \leq p \leq 0.05$).

The sensory evaluation of the brightness carried out on the sixth day showed significant differences ($0.01 \leq p \leq 0.05$) between the treatments. In the case of the coated blackberries, 67% of the panellists perceived the brightness of the fruits as acceptable (scores 5 to 3), while 100% of the panellists did not find significant differences in the control fruits ($0.01 \leq p \leq 0.05$) after the same storage period. The colour of both the coated and uncoated fruits was not found to be significantly different ($p \leq 0.05$). More than 93% of the panellists concluded that the colour was acceptable (scores 5 to 3) in both groups. However, on the sixth day, score 3 ("I neither like nor dislike it") was found to be significantly different from score 2 ("I dislike it"; $p \leq 0.05$) and score 1 ("I dislike it a lot"; $0.01 \leq p \leq 0.05$). The comparison of the colour on days 0 and 6 in the control fruits did not reveal significant differences ($p \leq 0.05$), as 100% of the panellists agreed that the colour of the fruits was acceptable (scores 5 to 3) at both storage times. In contrast, only 67% of the panellists found that the colour of the coated fruits on the sixth day was acceptable (scores 5 to 3), which is significantly different ($p \leq 0.05$) from the colour evaluated at the beginning of storage. By considering the results obtained from the sensory analysis of both the brightness and colour of the blackberries, we can conclude that the fruits are negatively affected by the edible film after a storage time of six days due to changes in their general appearance.

The flavour of the blackberries in both groups did not show significant differences ($p \leq 0.05$) at the beginning of storage, as 80% of the panellists perceived the flavour of the control fruits to be acceptable (scores 5 to 3), while 83% had the same opinion for the coated fruits. Similar results were obtained after six days of storage, as over 70% of the panellists stated that the flavour of the fruits in both groups was acceptable (scores 5 to 3). The flavour of the coated blackberries was found to be significantly different at days 0 and 6; in fact, the acceptance of the flavour diminished 13% after 6 days of storage.

3.7. Environmental and Social Impacts of the Proposed Edible Coatings

Each Mexican (in general, any inhabitant of the Latin American region) wastes 267 kg of food per year due to inefficient management practices, inadequate transport, distribution, and storage systems, and the use of improper packaging and containers, among other causes [40]. This annual food loss is equivalent to the emission of 300 kg of CO_2e per capita, the consumption of 32 m^3 of available freshwater per capita, and the use of 2193 m^2 of soil per capita [40]. Therefore, any technological innovation leading to a decrease in food waste is of key environmental interest, as it involves resources and biodiversity conservation, as well as global food security [41].

Due to its resilience towards water and nutrient shortages, *Opuntia* spp. (specifically, *O. ficus-indica* and *Nopalea cochenillifera*, which are both edible species) has been proposed as a raw material for the sustainable production of biofuels in arid zones of Brazil through anaerobic digestion [4]. In our approach, the edible coating proposed herein was prepared with mucilage extracted from an *Opuntia* species used only for animal feeding. The extraction of mucilage from *O. heliabravoana* Scheinvar appears to be a way to add value to this crop. It may foster the economy of drylands, where other vegetal resources are scarce, with no competition with food production. In addition, the valorisation of *Opuntia* would avoid the environmental consequences of its open-air decomposition or landfilling. We took the daily production of ten kg of mucilage as the basis for calculation, and we estimated a raw entry of 3.164 tons of *O. heliabravoana* Scheinvar, corresponding to the processing of 1155 tons per year from the mean yield obtained in this study (0.3% in dry basis). WARM analysis showed that if this amount of vegetal material was not valorised but instead landfilled, it would lead to the emission of 1607 tons of CO_2e. According to U.S. EPA [25], these tons of CO_2 are equivalent to the consumption of 3718 oil barrels, 180,713 gallons of gasoline, 344 passenger vehicles driven over one year, the electricity requirements of 241 homes for one year, or the carbon sequestered by the growth of 41,624 tree seedlings over ten years.

We also used the WARM model to simulate the valorisation of *Opuntia* waste per year to produce methane. The analysis showed that the anaerobic digestion of 1155 tons of fruit and vegetable waste generates ~1776 tons of CO_2e, considering the landfilling of the waste as the baseline scenario. This value implies that the anaerobic digestion not only avoids the 1607 tons of CO_2e generated annually by the landfilling of 1155 tons of *Opuntia* but also captures 1776 tons of CO_2 already present in the atmosphere.

The valorisation of forage *Opuntia* through methane production is an environmentally sound way to develop alternative energy sources in drylands. However, it is our view that a biorefinery approach would be more advantageous for communities, especially if, as in the case of Mexico, subsistence economies prevail in these zones. In such an approach, the production of edible coatings from the mucilage of *O. heliabravoana* Scheinvar would be accompanied by (i) ethanol recuperation from the liquid waste of the mucilage extraction process and its further reuse, and (ii) the anaerobic digestion of both the liquid waste produced by (iii) and the solid waste generated by the mucilage extraction.

The viability of this proposal has yet to be validated experimentally, as does its environmental impact assessed, as by simulation tools such as the WARM model.

4. Conclusions

The mucilage extracted from *O. heliabravoana* Scheinvar exhibited characteristics similar to those reported for the mucilage extracted from other *Opuntia* species. An edible coating was prepared from thermoplastic starch and the extracted mucilage, which was evaluated upon application to blackberries. The evaluation of the weight loss, size, pH, soluble solids content, and luminosity in these fruits during a 10-day storage period did not show significant differences from the characteristics of the control group. In contrast, the edible coating significantly diminished the microbiological load of the coated fruits and allowed their shelf-life to be increased by about two days. This can be attributed to the strong water retention capacity of the mucilage added to the coating, which may have diminished both water activity and microbial growth.

Multivariate analysis of the physicochemical and microbial variables indicated that the storage time negatively affected the weight and size of the coated and uncoated blackberries. The two main factors identified were related to the senescence process of the fruits and to three days of storage. The sensory analysis showed that the edible coating modified the brightness and colour of the fruits during the evaluation period. The flavour was the variable that was least affected by the edible coating and storage time. These findings support the use of the mucilage of forage species of *Opuntia* for the formation of edible coatings and films. However, attributes such as brightness and colour of the coated fruits must be optimised through further formulations of the mucilage and thermoplastic starch mixture.

The integral valorisation of forage and waste *Opuntia* through a biorefinery approach is a sustainable alternative to foster economic growth in drylands. This approach considers the scarcity of resources (such as biomass and water), as well as the necessity to diminish the environmental impact of food waste and the improper disposal of valuable plant material.

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