The effects of *Aloe vera* (*Aloe barbadensis*) coating on the quality of shrimp during cold storage

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Abstract Green tiger shrimp (*Penaeus semisulcatus*) is an important aquaculture species worldwide. Its perishable nature, however, needs preservation methods to ensure its quality and shelf life. In this study, the effects of *Aloe vera* coating on the quality and shelf life of shrimps during cold storage were investigated. Shrimp samples were dipped in aqueous solutions containing 25%, 50%, 75%, and 100% *Aloe vera* gel before storage at 4 °C for 7 days. Drip loss, pH, TBA, TVB-N, and texture of both the control and treated shrimp samples were analyzed periodically. There were significant differences between coated shrimps and the control group in all parameters evaluated. *Aloe vera* at 75% and 100% concentrations was able to prevent lipid oxidation and drip loss properly; however, coatings containing 25% *Aloe vera* did not have the desired effects on these characteristics. Shrimps coated with higher concentrations of *Aloe vera* had better textural properties during cold storage. Results also indicated the positive effects of *Aloe vera* coating on the sensory quality of shrimp.

Keywords *Aloe vera* · Coating · Shrimp · TBA · TVB-N

Introduction

Fresh shrimp is usually more perishable than most other foodstuff due to enzymatic and microbial activities, the latter effect

being often the dominant factor limiting the shelf life and wholesomeness during refrigerated storage and shipping. Over the past few decades, reliable methods have been developed to enhance shrimp shelf life in an attempt to avoid consumer health risks (Al-Dagal and Bazaraa 1999). In addition to such traditional methods as storage on ice and cook-chill processes commonly used for preventing the spoilage of fresh shrimp, various chemical and natural antimicrobial agents and polymers, antioxidants, and biotechnology products have also been developed and effectively used in the food industry. In addition, natural plant extracts, especially plant essential oils and bacteriocins such as nisin, have been found desirable additives for the preservation of food products (Lu et al. 2010). When it comes to methods and technologies, the role of bio-preservation methods using *Bifidobacterium breve* cannot be overemphasized in the improvement of shrimp quality during storage (Al-Dagal and Bazaraa 1999). Another recent development in this field is the use of edible coatings used to extend shrimp shelf life (Nowzari et al. 2013). In this method, a thin layer of edible, biocompatible coating protects the shrimp against physical and chemical damages, delays microbial growth, and enhances its safety. Moreover, the method has the additional advantages of reducing packaging waste and production of new food products (Valverde et al. 2005).

Environmental concerns and the growing consumer concerns about the effects of chemical preservatives on human health have encouraged recent studies to concentrate on edible coatings with natural antimicrobial and antioxidant properties. Chitin, chitosan, and their derivatives have been used for years in processing seafood products because of their antibacterial and antioxidative activities (Kamil et al. 2002). In addition, sepia, the ink produced by mollusks, has been found capable of retarding the spoilage of shrimp as an edible coating (Sadok et al. 2004). More recently, *Aloe vera* has been used in the food industry as an edible coating for preserving grape fruit

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quality in addition to its uses as a functional ingredient in drinks, beverages, and ice cream (Valverde et al. 2005).

Aloe barbadensis belongs to the Liliaceal family with about 360 species. Although this cactus-like plant grows in hot and dry climates as its natural habitat, it has been cultivated in many areas of the world. The presence of 75 active components in *Aloe vera* meets the needs of cosmetic and pharmaceutical industries for the production of natural products (Vogler and Ernst 1999). Anthrone, chromone, aloe verasin, hydroxyaloin, glycoprotein aloctin A, glucmannan, acemannan, and many other active ingredients have been found to have wound healing, anti-inflammatory, anti-tumor, anti-ulcer, anti-neoplastic, and anti-viral effects (Hu et al. 2003). A wide range of medicinal products made from *Aloe vera* are used for the treatment of diabetes, cancer, allergy, and AIDS as well as ulcerous, gastrointestinal, kidney, and cardiovascular disorders (Valverde et al. 2005).

The surface coating of *Aloe vera* has been used for extending the shelf life of tomato (Chauhan et al. 2013), apple slice (Song et al. 2013), mushroom (Mohebbi et al. 2012), sour cherry (Ravanfar et al. 2012), and many other fruits and vegetables due to its antimicrobial and antioxidant activities. The aim of this study was to investigate the effects of different concentrations of *Aloe vera* gel and low processing temperatures on the quality of shrimp during storage.

Materials and methods

Preparation of Aloe Vera gel

Aloe vera gel was extracted from the plant's green leaves by slicing each leaf in order to separate the epidermis from the parenchyma. The parenchyma pieces thus obtained were then crushed and squeezed to yield a mucilaginous gel which was filtered to remove the fibrous fraction (Guillén et al. 2013).

Proximate analysis of Aloe vera gel

The proximate composition of the gel obtained in the preceding stage was determined according to the AOAC methods. Moisture content was determined in an oven at 105 °C until the weight became constant. Total crude protein and lipid contents were determined using the Kjeldahl method and the Soxhlet method, respectively (AOAC 1996).

Determination of phenolic compounds

To determine the phenolic compounds in the ethanolic extract of *Aloe vera*, an aliquot (0.1 ml) of the extract was reached to 4.6 ml using distilled water and 0.1 ml of Folin-Ciocalteu reagent (diluted with 2 volumes of distilled water) and 0.3 ml of 2% aqueous sodium carbonate solution were added

before the tubes were shaken well. Absorbance of the mixture was measured at 760 nm via a spectrophotometer (Unico, Shanghai, China) after 2 h. In this experiment, calibration curve was prepared using gallic acid. The experiment was replicated three times and expressed as a mg gallic acid equivalent per 100 gram of dry matter (Ozsoy et al. 2009).

DPPH Radical scavenging activity

The antioxidant activity of the *Aloe Vera* extract solution was measured in terms of its radical scavenging ability using the stable radical DPPH. For this purpose, a total of 2 mL of 80% ethanol (v/v) solution of the *Aloe Vera* extract was mixed with 2 mL of a 2×10^{-4} mol/L ethanol solution of DPPH and the absorbance at 517 nm was immediately recorded. Measurements were continued at intervals of 5 min until the absorbance reached a steady state (Hu et al. 2003). All the determinations were performed in triplicates. The inhibition of the DPPH radical by the samples was calculated using the formula proposed by Yen and Duh (1994).

Shrimp preparation and coating

Green tiger shrimps (*Penaeus semisulcatus*) used in the experiments were caught in the Persian Gulf (Iran), immediately beheaded, gutted, washed with tap water, and transferred to laboratory on ice after 15 h. Shrimp samples were randomly assigned into four groups consisting of a control group (only immersed in water) and three experimental groups immersed in 100 g 100 g⁻¹ *Aloe vera* gel and a water solution of 75 g 100 g⁻¹, 50 g 100 g⁻¹, and 25 g 100 g⁻¹ *Aloe vera* gel.

Approximately 200 g of shrimp sample was used for each coated lot. The samples were plunged in the coating material for 10 min to ensure complete contact of the sample with the *Aloe vera* gel before they were brought out. The residual solution was allowed to drip off for 5 min. The coated samples were then dried using a gentle air flow for about 10 min and packed in polyethylene bags for storage at 4 ± 1 °C for a week. Samples were taken on days 1, 3, 5, and 7 after storage and all the experiments were repeated at each sampling interval.

pH Determination

An IKA homogenizer (ULTRA-TURRAX, Germany) was used to homogenize 5 g of shrimp with 45 mL distilled water for 30s before pH values of the samples were measured using a pH meter (JENWAY, USA).

Drip loss

To measure drip loss, all the samples were weighed after coating. Refrigerated shrimps were removed from packing and placed on a metal net for 10 min to release the

liquid drip. The drip loss percentage was calculated using the equation below:

$$\text{Drip loss (\%)} = \frac{W_{br} - W_{ar}}{W_{br}} \times 100$$

where, W_{br} and W_{ar} represent the weight of the shrimp samples before refrigeration and after elimination of surface water, respectively.

Determination of total volatile base nitrogen (TVB-N)

10 g shrimp sample and 2 g of MgO was homogenized in 300 mL of distilled water. The prepared sample was then distilled in 50 mL of boric acid and methyl red indicator until its volume reached 150 mL. The boric acid solution was then titrated with 0.1 N hydrochloric acid. TVB-N was reported as mg N/100 g of the sample (Goulas and Kontominas 2005).

Determination of thiobarbituric acid reactive substances (TBARS)

A shrimp sample 10 g in weight was blended with 25 ml of 20% trichloroacetic acid (TCA) for 2 min. The homogenized sample was mixed with 25 ml of distilled water and filtered through a Whatman filter paper (No. 1). From the TCA extract, 5 mL of the aliquot was mixed with 5 ml of 0.01 M 2-thiobarbituric acid in 90% glacial acetic acid and incubated for 1 h in boiling water. The TBA value was reported as mg of malonaldehyde/kg of the sample (Strange et al. 1977).

Evaluation of texture

Textural properties were measured at 4 ± 1 °C using a texture analyzer (Instron, model 1140, USA) equipped with a flat-ended cylindrical probe of stainless steel 5 mm in diameter for the penetration study. The sample was placed under the probe that moved downwards up to a depth of 5 mm at a constant speed of 20 mm/min while the penetration force was measured.

Sensory evaluation

From among the students, administrative staff members, and faculty members at the College of Agriculture, Isfahan University of Technology, Iran, 20 trained panelists (aged 19–30 years) were selected to carry out the sensory evaluation of the shrimp samples. The samples were randomly taken from each group and prepared by soaking in boiled water (95 ± 2 °C) for 3 min. Each sample was randomly designated by a three digit number and placed individually in a closed container to retain the aroma before they were presented to the panelists in a random order. Water and crackers were provided

to the panelists as palate cleansers. Panelists scored the samples for odor, taste, color, and texture quality as well as overall acceptability using a seven-point hedonic scale ranging from −3 (extreme dislike) to 3 (extreme like) (Weaver and Daniel 2003).

Statistical analysis

Each group of shrimps was prepared in four replicates and the mean values $\pm$ standard deviations were reported for each group. The split-plot analysis of variance with confidence intervals was set for a level of significance at $p < 0.05$ using the Statistical Analysis System (SAS) to evaluate the significance level of differences among the mean values.

Results and discussion

Proximate analysis

Table 1 shows the mean values and standard deviations for the *Aloe vera* gel composition. Clearly, the fresh gel contained a large amount of water trapped in 0.7% solid matter. This is in agreement with other reports indicating the presence of 99.5% water in *Aloe vera* gel (Eshun and He 2004). The remaining 0.5–1% solid matter included water- and fat-soluble vitamins, minerals, enzymes, polysaccharides, phenolic compounds, and organic acids. Fats comprised the smallest fraction in the *Aloe vera* composition with a value of 0.65% dry matter consisting of arachidonic acid, γ -linolenic acid, steroids (campesterol, cholesterol, β -sitosterol), and triglycerides (Hamman 2008). Aloe gel also contained a small amount of proteins including lectins and lectin-like substances.

Phenolic compounds and DPPH radical scavenging activity

Table 1 presents the composition of the phenolic content of the gel. The most important polyphenolic compounds in the *Aloe vera* gel are benzoic acid, *p*-toluic acid, *p*-coumaric acid,

Table 1 Chemical characteristics of *Aloe vera* gel

Parameters	Fresh <i>Aloe vera</i> gel
Moisture (g/100 g w.w. *)	99.3 $\pm$ 0.1
Protein (g/100 g d.m. **)	1.0 $\pm$ 0.0
Fat (g/100 g d.m.)	0.65 $\pm$ 0.08
Phenolic compound (mg gallic acid/100 g d.m.)	82.7 $\pm$ 3.04
DPPH radical scavenging activity (%)	67.51 $\pm$ 0.71%
pH	4.64 $\pm$ 0.06

* w.w.: based on wet weight

**d.m.: based on dry matter

psalicylic acid, protocatechuic acid, hydroxyphenylacetic acid, ferulic acid, aloe emodin, and vanillic acid (Nejatzadeh-Barandozi 2013). The phenolic compounds seem to play a role in the antioxidant activity of the *Aloe vera* extract due to their good hydrogen donating properties.

The *Aloe vera* extract exhibited a DPPH radical scavenging activity of about 68% (Table 1). This value is comparable with the results reported by Hu et al. (2003) and Ozsoy et al. (2009) who reported oxidation inhibitory values of 71% and 72% for *Aloe vera*, respectively. A high correlation has been reported between the radical scavenging activity and the phenolic and flavonoid contents of this plant (Ozsoy et al. 2009). Hu et al. (2003) evaluated the antioxidant potential of the *Aloe vera* extract in different growth stages of the plant and found that its antioxidative activity was age-dependent and comparable to those of BHT and α -tocopherol. The antioxidant mechanisms of the *Aloe vera* constituents have not been yet clarified, but it contains polysaccharides and flavonoids which inhibit lipid oxidation. The main antioxidant compounds identified in the different *Aloe* species include 7-O-methylaloesin A and ORAC as well as indoles and alkaloids (Zapata et al. 2013).

pH value

Figure 1 shows the pH values in all the treatments. Results showed that storage time and *Aloe vera* concentration had a significant effect on the pH value of the coated shrimp ($p < 0.05$), which decreased on day 1 with increasing *Aloe vera* concentration. This may be explained by acid accumulation in the *Aloe vera* gel (Zapata et al. 2013) as its concentration in the coating increased. Moreover, the data obtained indicate that pH variations followed the same trend in all the *Aloe vera* gel-

coated samples; however, the increasing pH level exhibited a faster trend in the control sample during cold storage.

Not only have previous studies demonstrated the antimicrobial effect of *Aloe vera* (Gontijo et al. 2013), but the lower pH of the *Aloe vera* gel coating (pH of coating=4.64) has also been found to have a positive effect on the inhibition of microbial growth. By inhibiting the endogenous proteases activity and decreasing the rate of microbial growth, *Aloe vera* could have a preservative effect on shrimp. The production of some compounds, like trimethylamine and dimethylamine by enzymatic activity increases pH value of samples (Song et al. 2011). Finally, the production of volatile basic nitrogen in the interior shrimp muscles is inevitable due to the presence of the external *Aloe vera* coating, which led to increasing pH with time.

Drip loss

Based on the results obtained, both storage time and *Aloe vera* concentration had significant effects on drip loss ($p < 0.05$). Fig. 2 shows the effects of cold storage and *Aloe vera* treatment on drip loss. The control samples exhibited a higher weight loss than those treated with *Aloe vera*; however, the treatment with 25% *Aloe vera* showed the same results as the control. The losses ranged from 3.7% (on day 3) for samples treated with 100% *Aloe vera* to 11.3% (on day 5) for the control samples. Coating with 100% *Aloe vera* had the highest efficiency in preventing water loss from shrimp muscles so that drip loss increased with reducing *Aloe vera* concentration in the coating. In general, the hygroscopic properties of the edible coating had a positive effect on water retention such that *Aloe vera* formed a water barrier between the shrimp and

Fig. 1 Changes in pH values of shrimp samples coated with different concentration of *Aloe vera* during cold storage

Fig. 2 Changes in drip loss of shrimp samples coated with different concentration of *Aloe vera* during cold storage

the ambient air, thereby preventing water release into the environment.

Water loss showed an increasing trend up to day 5 of the storage period when it declined. It may be claimed that rising pH was a possible cause for the reducing drip loss after 5 days of storage. The water holding capacity of meat was influenced by the increasing pH change so that more water could be held in the space between actin and myosin at higher pH levels. Also, the dehydrated surface layer formed during cold storage reportedly produces an additional barrier to mass transfer in biological substances (Pham and Willix 1984). This mechanism might have also led to the slower trend of water loss in shrimp samples after a certain period of storage.

Total volatile base nitrogen (TVB-N)

Crustacean meat is known to be rich in free amino acids and soluble nitrogen; thus, one indicator of shrimp spoilage is the increased total volatile base-nitrogen. The reaction that leads to the production of volatile base nitrogen involves the transformation of trimethylamine oxide (TMAO) to trimethylamine (TMA) using reductive enzymes in the micro-organism (Lu 2009). Fig. 3 shows the effect of different *Aloe vera* concentrations on TVB-N production in shrimp samples stored at 4 °C. In this study, the initial TVB-N value of the shrimps prior to storage was ~34–36 mg N/100 g meat, which was far greater than the recommended level of 25 mg N/100 g (Ojagh et al. 2010). However, this acceptable limit may be questionable for shrimps as higher values have been observed in fresh shrimp. This is also confirmed by the findings of Zeng et al. (2005). TVB-N value seems to have increased during the time the shrimps were being transferred to the laboratory. This transfer time might also have had the effect of inhibiting direct

contact between ice and shrimp which obviously entailed no later washing out of TVB-N (Cobb et al. 1976)

During storage, TVB-N value tended to increase in all the samples due to the activities of bacteria and endogenous enzymes (Kyrana et al. 1997). This is while no significant increase was observed during the first 3 days of storage in the TVB-N values measured in samples coated with 75% and 100% *Aloe vera*. In both the control shrimps and those coated with only 25% *Aloe vera*, a marked increase was observed during the storage period. These effects might be somehow related to the inhibitory effect of *Aloe vera* on microbial growth. This is in agreement with the findings of Asamenew et al. (2011) who reported strong antimicrobial activity by aloin and 7-O-methylaloeserin A in a wide range of Gram-negative and Gram-positive bacteria and fungi strains.

On day 5 of the storage period, a significant increase was observed in shrimps coated with 75% and 100% *Aloe vera*. It is likely that the surface bacteria would not grow during the first days of storage, but that the interior microbes and enzymes would increase the TVB-N value after some time. In the latest stage of the storage period, TVB-N values rose significantly, but the highest increase was observed in the control that lacked *Aloe vera*. The coating containing 75% and 100% *Aloe vera* had lower volatile nitrogen content than other treatments. This can be attributed to either the reduced microbial population or the diminishing oxidative deamination of non-protein nitrogen compounds by enzymes (Song et al. 2011). Identical patterns were observed in TVB-N and pH variations. While no study in the past has investigated the effect of *Aloe vera* on TVB-N variations, the results of the present study indicate that coating shrimps with *Aloe vera* leads to a significant reduction in TVB-N values during storage at 4 ± 1 °C ($p < 0.05$).

Fig. 3 Changes in TVB-N value of shrimp samples coated with different concentration of *Aloe vera* during cold storage

Thiobarbituric acid reactive substances (TBARS)

As can be seen in Fig. 4, TBA value was higher in the control than in the experimental groups during storage. At the beginning of the storage period, TBA values varied between 0.68 ± 0.09 and 0.62 ± 0.06 mg malonaldehyde/kg in the control and coated shrimps, respectively, whereas these values rose to 1.5 ± 0.04 and 1.1 ± 0.1 mg malonaldehyde/kg at the end of the storage period. During cold storage, the TBA value increased in all the samples; however, the rate of oxidation was higher in shrimps lacking *Aloe vera*. The shrimp flesh must have been susceptible to oxidation due to the presence of a higher ratio of unsaturated fatty acids, which resulted in increased hydroperoxide content followed by an increase in the malondialdehyde content whose formation is indicated by the rising TBA value during storage (Kilincceker et al. 2009).

The results of DPPH radical scavenging activity indicated the good antioxidant activity of *Aloe vera*. The compound seems to have been able to prevent satisfactorily the oxidation of unsaturated fatty acids of shrimps since TBA values did not exhibit any significant increase after 3 days of storage in the 50%, 75%, and 100% *Aloe vera* gel treatments. The antioxidant effect of *Aloe vera* gel receded and oxidation proceeded at a faster rate when its concentration was reduced to 25%, although better results were still achieved in this treatment up to day 5 of the storage period as compared to the control. Moreover, the *Aloe vera* coating prevented the reaction of oxygen with fatty acids. At lower gel concentrations, oxygen easily passed through the coating layer such that after 7 days of storage, no significant differences were observed between the 25% *Aloe vera* treatment and the control while TBA values also increased in the 50% *Aloe vera* treatment.

Fig. 4 Changes in TBA value of shrimp samples coated with different concentration of *Aloe vera* during cold storage

Fig. 5 Changes in penetration force of shrimp samples coated with different concentration of *Aloe vera* during cold storage

Texture

Figure 5 shows the effects of storage time and *Aloe vera* concentration on the texture of the shrimp samples. Clearly, the treatments had a significant effect on the toughness of the shrimp flesh. During the first 3 days of storage, the toughness of the shrimp texture increased. After this period, two different trends were observed. Shrimps coated with water and 25% *Aloe vera* became softer while those in the 75% and 100% *Aloe vera* treatments did not exhibit any significant differences in their textural properties. The predomination of autolysis to rigor mortis in the shrimp muscles decreased the firmness of the muscular tissue, which might have been mainly due to the enzymatic degradation of muscle proteins (Ocaño-Higuera et al. 2011). After fish death, postmortem autolysis was triggered, causing the shrimp flesh to soften and muscle elasticity to decrease. This process might have been accelerated by microbial activity (Olafsdottir et al. 2004). In this study, *Aloe vera* served to slow down the process and whereby the loss of toughness. A major problem in the seafood industry is the higher rate of flesh softening of aquatic life than in other types of meat. *Aloe vera* might, therefore, be satisfactorily exploited to retain flesh toughness and maintain the textural properties of shrimp muscle.

Sensory evaluation

The sensory attributes of shrimp coated with different concentrations of *Aloe vera* gel are presented in Table 1. Aroma and overall acceptability of the samples were significantly different on day 5 of storage ($p < 0.05$) while no significant differences were observed in taste, color, and texture. The enhancement of TVB-N in the control and the 25% and 50% *Aloe vera* treatments might be claimed as the reason for the lower acceptability and the undesirable odor in these samples.

Panelists were able to detect off-odor in both the control samples and those coated with 25% *Aloe vera*, whereas none of the panelists detected the off-odor in samples coated with 75% or 100% *Aloe vera* gel. Fortunately, *Aloe vera* did not have any undesirable effects on the taste of the samples and could be used for coating of shrimps without any adverse effects on shrimp quality.

Conclusion

A major problem in the seafood industry is the limited shelf life of the products, especially shrimps. Different methods have been so far developed and used for extending shrimp shelf life but all at the expense of fresh flesh quality. Among the possible solutions to this problem is coating of shrimp with edible compounds for preservation of the quality. The results of the present study indicate that *Aloe vera* gel has positive effects on retarding lipid oxidation. The treatments in our experiments led to reduced TBA, TVB-N, pH, and drip loss values in shrimps treated with higher *Aloe vera* gel concentrations. Moreover, coating green tiger shrimps with *Aloe vera* concentrations of 75% and 100% led to the retention of the fresh flesh quality during cold storage.

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