

# **Evaluate the Effect of Calcium Chloride Plus Sodium Alginate Treatment on Button Mushroom During Storage**

# Beyaz Şapkalı Mantarda Hasat Sonrası Kalsiyum Klorit ve Sodyum Aljinat Uygulamalarının Muhafaza Kalitesine Etkisinin Belirlenmesi

Ayse Tülin Öz\* <sup>©</sup> and Tülin Eker <sup>©</sup>

Osmaniye Korkut Ata University, Engineering Faculty, Food Engineering, Karacaoglan Campus, Osmaniye, Turkey.

#### ABSTRACT

n this study, distilled water (control), 1% calcium chloride  $(CaCl_2)$  (w/v), 1% sodium alginate (w/v), and combination with 1%  $CaCl_2 + 1\%$  sodium alginate (w/v) solutions were sprayed on freshly harvested button mushrooms. Treated mushrooms were stored in polypropylene boxes during 20 days at 4±1°C. Physical and chemical properties of treated mushrooms were investigated at five days intervals during storage. The present results showed that treatment of 1%  $CaCl_2$  revealed close effectiveness on weight loss, with 1.21%, L\* color value with 81,76, the and cap opening percentage with 11%. minimum decrease in firmness with 2.2 N of the mushrooms compared to other treatments at end of storage. The experiment results could suggest the 1%  $CaCl_2$  application for mushroom quality and shelf life were more observable up to 15 days.

#### **Key Words**

Button mushroom, edible coating, sodium alginate, shelf life.

## öΖ

Galışmada hasat edilen taze beyaz şapkalı mantarlara saf su (kontrol), % 1 kalsiyum klorür (CaCl<sub>2</sub>) (w / v), % 1 sodyum aljinat (w / v) uygulaması ile % 1 CaCl<sub>2</sub> +% 1 sodyum aljinat (w/v) kombinasyonu püskürtme yöntemi ile uygulanmıştır. Uygulama yapılan mantarlar polipropilen kutularda 4 ± 1°C'de 20 gün boyunca muhafaza edilmiştir. Uygulama yapılan mantarların fiziksel ve kimyasal özellikleri muhafaza süresinde 5 gün aralıklarla belirlenmiştir. Mevcut uygulama sonuçlarına göre % 1 CaCl<sub>2</sub> uygulaması yapılan , mantarların muhafaza sonunda ağırlık kaybı,%1,21, mantar L\* (parlaklık) renk değeri 81,76, şapka açılma oranı %11 ve şapka sertlik değeri 2.2 N ile diğer uygulamalar daha etkili olduğu belirlenmiştir. Uygulama sonuçlarına göre mantar kalitesi ve raf ömrü için %1 CaCl, uygulamasının 15 günlük raf ömrü süresince daha etkili olduğu belirlenmiştir.

#### Anahtar Kelimeler

Beyaz şapkalı mantar, yenilebilir kaplama, sodyum aljinat, raf ömrü.

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Correspondence to: Osmaniye Korkut Ata University, Engineering Faculty, Food Engineering, Karacaoglan Campus, Osmaniye, Turkey. E-Mail: aysetulinoz@osmaniye.edu.tr

# INTRODUCTION

he button mushroom (Agaricus bisporus) is one of the most popular edible mushrooms worldwide. The driving force behind this species are relatively reasonable cultivation methods and nutritional value [1]. The button mushrooms have a tendency to lose quality in a short time (3 to 4 days at room temperature) as a result of high respiration rate, bruising phenomena, water loss or a microbial deterioration. Because of high moisture content and respiration rate, shelf-life duration of mushrooms is very short. Especially delicate cuticle behavior, open caps and color browning are important for acceptability by consumers [2]. For delaying ripening and senescence process and extending shelf life of fresh mushrooms, there are several pre or postharvest treatments; modified atmosphere packaging and controlled atmosphere storage, UV-irradiation, vacuum cooling, low temperature storage and edible coating [3-5]. Among these, edible coating is the most used method, which can act as semipermeable barrier around fresh produce to prevent water loss, gases and volatiles and oxidation [6]. Edible coatings can also be used as carriers for some bioactive compounds, color agents, herbs and spices [7]. Several coating materials for A. bisporus mushroom such as chitosan, gum arabic, carboxymethyl cellulose, tragacanth gum, alginate with ergosterol or Tween 80 have been investigated [8, 9]. Polymeric coatings such as sodium alginate is popular one of the most studied edible coating because it meets almost all the advantages of edible coating. It has an excellent gel forming capacity. Solutions of alginate cerate form of gels when divalent or multivalent cations are added, through bridge formation between gluconate residues. The calcium treatment has important affects for maintaining of fresh product firmness and limiting physiological disorders during postharvest storage [10]. CaCl, supports texture of fresh produce and improves structure of cell membrane [11]. The advantages of alginate coatings have been reported in many studies when applied other fruits and vegetables [12-14]. In a few publications, the use of alginate for coating mushroom has been previously studied for mushroom, coated A. bisporus mushrooms with alginate + CaCl, solution and determined some physical quality parameters of mushrooms for 142 hours [15]. Whole button mushrooms coated with alginate + ergosterol + tween 80 combination showed better visual quality and protect weight loss when compared with the untreated samples [16]. Edible coating protect skin suface of fresh

produce and reduced moisture loss, prevent texture changes showed better appearance.Jiang et al. 2013, applied alginate in combination with nanomaterials (Ag) in fresh shiitake mushrooms and they reported that the alginate+nano-Ag coating showed better performance of the shiitake mushroom to promote its shelf-life quality for longer time [9]. It has been realized that previous studies focused only effect of alginate on physical properties of mushroom however less information the effect of coating treatment on chemical properties such as: antioxidant activity and MDA content of mushroom. In present study, it aimed to investigate the effect of treatment of CaCl<sub>2</sub>, coating of alginate and alginate combination with CaCl<sub>2</sub> on mushroom quality for 20 days cold storage.

# MATERIAL AND METHODS

# Plant material and coating treatments

Unwashed, whole, white button mushrooms with a 3.61 cm cap diameter and closed cap were hand-harvested from growing bags in the mushroom production houses facility at Osmaniye Korkut Ata University (Osmaniye, Turkey). Sodium alginate (Sigma-Aldrich, Co., St. Luis, MO, USA) (1%, w/v) was prepared by mixture of 5 g of alginate in 500 ml of distilled water and mixing at a controlled temperature of 70 °C to obtain a clear solution. CaCl<sub>2</sub> (1%, w/v) was dissolved in distilled water to form an aqueous solution [6]. The aqueous solutions of CaCl, and Na-alginate were mixed at 1:1 to prepare CaCl<sub>2</sub> + Na-alginate (1%, w/v). After harvest; on same day, mushrooms were divided into four groups and treated with i) distilled water (control), ii) CaCl, (ca), iii) alginate (na) and iv) CaCl<sub>2</sub> + alginate (ca+na). The mushrooms were not rinsed before coating, because they were clean after harvest. Solutions were applied by spray method on mushrooms. After, any excess solution was used to absorb by filter paper and mushrooms allowed to dry after coating around one hour at 25 °C room temperature (Figure 1). After treatment, mushrooms were stored in transparent PET (Petsa, 1000 cc, 18x20 cm) boxes during 20 days at 4 °C in the dark. Each treatment group comprises ten packages and two of them (two replicate) were used to analyses, at five days intervals and around 350 gr mushroom were put in for each boxes.

# Button mushroom quality evaluation

The button mushrooms quality assessments were measured during shelf life at 4°C. The weight loss was measured as the (%) of weight loss [17]. Mushrooms were squeezed with a juice extractor and the juice was used for further analysis. The opening caps value was calculated by proportioning the mushroom with the cap opened to total mushroom using following the equation [18]:

# % Cap opening=(Noc)/(Ntm)×100

where *Noc* expresses cap opened muhsrooms, and *Ntm* total mushroom.

# **TSS and pH measurements**

The total soluble solids (TSS) content was measured by the digital refractometer (Krüss, Hamburg, Germany).

Mushrooms pH value was measured with Thermo Scientific Orion 2 Star CG 710 pH meter for every treatments [11]

# **Firmness measurement**

Mushroom cap firmness was measured by using texture analyzer (Brookfield, US) with (TA39) probe speed was 2.0 mm/s. Penetration was performed through 3 mm on cap. The firmness value was defined as Newton [8].

# **Color measurement**

The mushroom color was measured with Chroma Meter (CR-400, Konica Minolta Inc., Tokyo, Japan) in  $L^*a^*b^*$ values. 5 measurements on each cap were performed.



Figure 1. a) Mushrooms in growing bag, b) Harvesting, c) Spraying of edible coating on mushrooms, d) Drying, e) Storing in PET boxes.

The browning (BI) were measured as the following equation [19]:

BI=  $[100 (x \ 0.31)]/0.172$ ; where x = (a + 1.75 L)/(5.645 L + a-3.012 b).

#### Malondialdehyde (MDA) content

The MDA of mushrooms was determined using the 2-Thiobarbituric acid reaction (TBARS) by [20] to evaluate the degree of lipid oxidation. Two mL of mushroom juice was mixed with 5 mL of 20 % (w/v) trichloroacetic acid (TCA) and then centrifuged (10000 × g, 20 min, 4°C). The clear upper phases were used for measurement of MDA level using TBA. MDA content was expressed as  $\mu$ mol/L fresh weight.

#### **Radical scavenging activity**

The mushroom juice was centrifugated at 3500 rpm for 10 minutes. The 0.1 mL of aqueous upper phase and 3.9 mL of DPPH solution (60 mM in MeOH) were mixed. The solution was incubated in dark condition for 1 hour. The at 517 nm absorbance was used monitored against MeOH. The radical scavenging activity was determined using formula which  $A_{control}$  is absorbance of DPPH solution and  $A_{sample}$  is absorbance of the sample solution [21].

Radical Scavenging Activity (%) =  $(A_{control} - A_{sample})/A_{sample} \times 100$ 



Figure 2. Effect of coating on total weight loss (A), cap opening (B), pH (C), and total soluble solid (D) changes of mushrooms stored at 4±1°C for 20 days.

# Statistical analysis

All data were analyzed using the SPSS statistical package program (SPSS, version 18.0, USA). Analysis of variance was performed to determine the significance of differences between means. Each analysis was replicated at least three times. Statistical significance was considered at p<0.05.

## **RESULTS and DISCUSSION**

# Weight loss, percentage of open caps, pH and TSS

There was some weight loss that occurred in all treatment groups during 20 days of storage (Figure 2-A). Although, results were not shown differences statistically between coated mushroom samples at the end of the storage  $CaCl_2$  exhibited a lowest moisture loss in comparison to the other groups. It appeared that the Ca -alginate showed much better performances than the alginate coating samples. Thus, the degree of weight loss of the fresh mushrooms (control) stored at 4 °C in 20 days was 1.29% while the weight loss for the same period of storage in alginate-coated mushrooms was even 1.9%. The alginate-coated samples showed the highest weight loss and it may be the result of the hygroscopic nature of hydrocolloids. Olivas et al. 2007, stated that alginate does not satisfy as good moisture barriers especially for high water content produce [22]. In that case, alginate coating was not satisfactory for prevention of moisture loss. However, the alginate in combination with CaCl, coating treatment showed better vapor barrier and made alginate films water-insoluble. The elevated moisture loss in mushrooms during shelf life affects a decrease in cohesive strength of water and proteins that are related for the closed position of the caps [19]. Mahajan et al. 2008 stated that when the weight loss in the products is between 5-10%, it is of marketable quality. This view supports our results [23]. The cap opening level of mushrooms increased in all the treatments, and it was 22.62%, 15.07%, 14.96% and 11.02%, for control, CaCl\_+alginate, alginate, and CaCl\_, coating, respectively (Figure 2-B). CaCl, treatment limited mushroom open cap percentage compared to all treatments. Calcium ions protect cell wall and middle lamella and keep texture of fresh produce [24]. There is a close correlation between aging and open cap percentage which is important parameter for marketable value [18,



Figure 3. Effect of coating on firmness of mushrooms stored at 4±1°C for 20 days.

25], pH values of mushrooms sharply decreased on the day of 5 in all groups. After the 5th day of storage, the pH values of all groups started to increase during the trial days. There were significant differences between groups and days for each treatment (Figure 2-C). The highest pH percent decrease over the 20 days of analysis was recorded for the alginate-coated samples. There was a reduction of TSS content compared to initial value of mushroom TSS during the storage in all the treatments while CaCl, treated mushrooms experienced a slight increase on day of 15 (Figure 2-D). In fact, it was found that mushrooms with high water loss have higher total soluble solids. As shown Huang et al. 2019 the control groups had the highest of TSS and increase significantly in TSS together with an extend in storage period and coated samples decreased respiration rate also

delayed use of metabolites result in lower TSS content [26]. Calcium treatment limited of water-soluble pectin [10], it may limit TSS content increased also.

# Firmness

Flesh firmness is one of main marketable value for mushroom after harvest and it shows produce degradation and loss in water content. Firmness value, which has important effect on shelf life (5) was quickly decreased in *A. bisporus* In the present study, all mushrooms gradually softened during storing period (Figure 3). As shown in Figure 3, the texture of alginate coated samples had fastly softened, and also losing 55.47% of their firmness value at the end of the experiment. It may be result of the hydrophilic character of alginate, soit could not act as a good barrier to water transfer



Figure 4. L\* value of mushrooms (A), appearance of alginate coated mushrooms on 10th day of storage (B), a\* value of mushroom (C), and b\* value of mushroom (D).

and delay dehydration [27]. It was indicated that firmness of fresh commodity was re correlated to loss of turgor pressure in the cell membrane reduced by loss of water [8]. The use of  $CaCl_2$  represented a beneficial effect for the coated mushroom by delaying its softening and showed the minimum loss of firmness (19.34%) at the end of the trial days. The addition of  $Ca^{2+}$  have been referred to raise the hardness of the cell membrane of fruits and vegetables and limit the flesh softening [9].  $CaCl_2$ -dipping application were reported to protect the cell wall and middle lamella structure and improve the texture of fresh produce [24]. Calcium treatment after harvest prevent water soluble pectin by constitute of calcium pectate that keep firmness of fresh produce [10].

### Color

Color is the most obvious indicator of quality of fruits and vegetables because it is the first parameter for consumer acceptance [8]. The lightness ( $L^*$ ) parameter is an showed the brightness and whiteness towards 100 values and decreasing to zero an indication of mushroom browning.  $L^*$  value was decreases progressively in was observed during storage in all groups (Figure3-A). The mushrooms treated with CaCl<sub>2</sub> treatment had the highest  $L^*$  value and they often appeared whiter than the other treatments at the end of study. The surface of mushroom coated with alginate seemed wet after 10 days of storage. It could be said that this was due to hygroscopic nature of alginate. [22]. The color a\* values of control group were significantly higher than the other treatment groups (Figure 4-C). While CaCl, coat-



# **Browning Index**

Figure 5. Effect of coating on browning index of mushrooms stored at 4±1°C for 20 days.

ing did not cause any significant differences in redness  $(a^*)$ , other groups showed significant increases and decreases during storage. The decrease in yellowness  $(b^*)$  of mushrooms occurred during storage, but variations of  $b^*$  values were significant among in all treatments (Figure 4-D). Browning of mushroom surface is the most important factor affecting consumer choice and marketing quality. Browning of button mushroom could be result of many factors include oxidation, mechanical damage, temperature, relative humidity, and atmosphere gas [28]. The treatments did not produce positive effect on browning index (BI) of samples. There were no significant differences between groups for each sampling day (Figure 5).

#### Malondialdehyde (MDA)

The level of lipid peroxidation was indicated by the amount of Malondialdehyde in the cells. MDA content is considered as indicator of cell membrane degradation and cell oxidative damage [26]. In this study, the formation of MDA, a secondary end-product of polyunsaturated fatty acid oxidation [5], showed sharp increase until the day of ten regardless of the treatments. As shown in Figure 6, the MDA level in control mushroom

at the end of the study was 1.46 times higher than the initial value MDA level and followed by alginate coated samples. Nevertheless, MDA amount in chitosan + guar gum (CH+GG )15% and CH+GG 25% treatments were relative lower during storage. When the experiment was terminated, MDA content in control and CH+GG 15% were 136.7% and 26.2% higher than those of initial day, respectively. The MDA values of mushrooms coated with CH+GG 5%, and CH+GG 25% were 104.1%, and 40.4%, respectively. MDA content, the oxidative enzymic product, in the coated mushrooms was significantly lower than control mushroom samples at the end of study and also CaCl, treated mushroom had the lowest MDA content. The present results showed that the combination of alginate and CaCl, treatment could decrease oxidative injury during the long storage days (Figure 6).

# Radical (DPPH) scavenging activity (RSA)

The influence of different coatings on RSA were given in Figure 6-B. The RSA activity determines non-enzymatic antioxidant activity in plants [27]. Instance of modification in RSA was similar for control mushroom samples and coated samples throughout storage. They all



Figure 6. Effect of coating on MDA content (A) and radical scavenging activity (B) of mushrooms stored at 4±1°C for 20 days

showed that little increase throughout the first 10 days of storage, after this the antioxidant activity decreased progressively during the last 20 days, regardless of the coating treatment. The increase in RSA in first 10 days could be characteristic to the progress of antioxidant compounds throughout cold storage and ripening [27]. There was a statistically significant change for scavenging activity during storage. Coating produced significant effect on RSA of mushrooms. Control mushrooms had the lowest RSA, while alginate coated samples exhibited higher RSA at the end of the storage. It could be seemed that alginate coating had beneficial effect on RSA. However, when we consider overall quality parameters, this phenomenon could be result of rapid ripening according to previous works [27-29]. Jimenez et al. reported that the liquid-phase antioxidants increased during the ripening action and there is a positive correlation between this increase and substantial changes in their redox state, becoming more reduced as ripening actions [29]. It could be explained that the alginate treated samples showed more rapid ripening resulted in significant changes of reducing status. Conclusion of this study, button mushroom was treated with CaCl., alginate, and in combination with alginate and CaCl, for the preservation during cold storage. The present results indicated that treatment of CaCl\_ decreased weight loss, kept L\* color value, limited firmness loss and cap opening percentage. However, alginate treatment caused the highest weight loss and softening. Alginate-CaCl, combination showed the lowest MDA value at the end of the storage. Alginate did not produce beneficial effect on coated samples as a moisture barrier very well. The weight loss, L\* color value, and cap opening are the first marketable quality parameters for mushrooms. In present study 20 day of storage period was chosen however coating advantages were more observable up to day of 15. As the results of this study, it could be recommended that the treatment of CaCl, and alginate-CaCl, for maintaining button mushroom quality and shelf life for two weeks storage. Moreover, further researches will be needed to reveal the practicable combinations of alginate coating material during storage.

#### **Conflicts of Interest**

The authors declare that there are no conflicts of interest.

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