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Extending shelf life and developing palatability of chicken meat by coating active edible film

Aktif yenilebilir film kaplanarak piliç etinin raf ömrünün uzatılması ve lezzetinin geliştirilmesi

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Abstract

In this study, the active film (AF) was produced from sodium caseinate-starch mixture (1:1) by adding salt, garlic extract, black pepper, and bay laurel oil. AF exhibited significant antibacterial activity against Salmonella. Coating chicken meats with active film reduced the total number of mesophilic and coliform bacteria by about 2.6 log cfu/g units during refrigerated storage. After 19 days, the psychrophiles count of the unwrapped meat increased from 6.80 cfu/g to 10.67 cfu/g, while the psychrophiles count of the AF-coated sample increased only to 8.13 cfu/g. Plane sodium caseinate-starch film (PF) and AF coatings increased the shelf life of chicken meat stored at +4 °C from 8 days to 11 and 19 days, respectively. On day 4, the samples were roasted at 200 °C for 30 minutes. Both PF and AF film reduced cooking loss and instrumental tenderness. While chicken meat pre-coated with PF was evaluated as softer, meat coated with AF was considered more delicious by the panelists.

Keywords: Active film, Bay laurel oil, Black pepper oil, Garlic extract, Chicken meat.

Öz

Bu çalışmada sodyum kazeinat-nişasta karışımına (1:1) tuz, sarımsak özü, karabiber ve defne yaprağı yağı ilave edilerek aktif film (AF) üretilmiştir. AF, Salmonella'ya karşı önemli antibakteriyel aktivite göstermiştir. Piliç etlerini aktif filmle sarmak, buzdolabı koşullarında toplam mezofilik ve koliform bakteri sayısını yaklaşık 2.6 log cfu/g birim azaltmıştır. 19 gün sonra, ambalajsız etin psikrofil sayısı 6.80 cfu/g'den 10.67 cfu/g'ye yükselmişken, AF sarılı numunenin psikrofil sayısı 8.13 cfu/g'ye yükselmiştir. Sade sodyum kazeinat-nişasta filmi (PF) ve AF ile sarma, +4 °C'de saklanan piliç etinin raf ömrünü sırasıyla 8 günden 11 ve 19 güne çıkarmıştır. 4. gün numuneler 200 °C'de 30 dk. pişirilmiştir. Hem PF hem de AF filmi, pişirme kaybını ve sertliği azaltmıştır. Panelistler tarafından PF ile sarılmış piliç eti daha yumuşak olarak değerlendirilirken, AF ile sarılmış et daha lezzetli olarak değerlendirilmiştir.

Anahtar kelimeler: Aktif film, Defne yaprağı yağı, Karabiber yağı, Sarımsak özü, Piliç eti.

1 Introduction

The consumption of chicken meat and its products worldwide is constantly rising because of its relatively low price, easy digestibility, and low-fat content, as well as being high quality protein source. Raw chicken meat usually has a shelf life of 8-12 days, depending on the production technique and packaging. Chicken meat is rapidly spoiled due to the growth of the lactic acid bacteria, *Enterobacteriaceae, Pseudomonads spp.* and *Brochothrix thermosphacta* under aerobic conditions at refrigeration temperature [1]. In addition, chicken meat could be contaminated with a variety of potential food-borne pathogens such as *Salmonella.* and *Listeria* [2].

Spices are mostly utilized for their natural flavors, but their bioactive components enable them to be used as food preservatives. Spices essential oils or extracts obtained from plants such as garlic, black pepper, bay leaf, thyme, marjoram, sumac, and rosemary have strong antimicrobial effects on pathogens and food spoilage microorganisms due to their high amount of phenolic compounds [3]. Fresh garlic extracts have been found to inhibit the growth of *Salmonella Typhi Staphylococcus aureus* and *Staphylococcus epidermidis* [4]. Han and coauthors [5] stated that the antibacterial activity of 1 mg allicin, an organosulfur compound obtained from garlic, was equal to that of 15 IU of penicillin. Teneva and coauthors [6] showed that black pepper oil exhibited antimicrobial activity against *Salmonella sp, S. aureus, Proteus vulgaris*, and *E. coli.* It was found that laurel leaf oil showed high antibacterial activity against *E. coli, S. aureus, S. Enteritidis, S. typhimurium, Listeria innocua, L. monocytogenes* and *Campylobacter jejuni* [7],[8].

Antimicrobial edible films were obtained by adding various essential oils (thyme, rosemary, garlic, laurel, pimento, cinnamon, and black pepper) [9]-[12]. Whey protein isolate films including 4% garlic oil showed significant inhibition against S. aureus and L. monocytogenes [13]. The alginate film containing 0.1% garlic oil decreased the bacteria counts in the nutrient broth for S. aureus, E. coli and S. Typhimurium, by 4.31, 2.28 and 1.24 log units, respectively, after 24h incubation [14]. Coating in gelatin films containing 1% laurel oil raised the shelf-life of vacuum-packed fish fillets by 7 days. And the film wrapping reduced the total viable, total psychrotrophic bacteria and Enterobacteriaceae counts around 2 log units during storage [15]. Carrageenan film containing black pepper oleoresin increased the shelf life of buffalo steaks two times and decreased viable and psychrotrophic counts [9].

Juiciness, tenderness, and flavor are the three main components of meat palatability, which is defined as the eating quality of meat and related to consumer acceptance [16].

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Cooking conditions have a major impact on these three palatability components. The water, fat-binding properties, and texture that affect meat tenderness are closely related to the applied heating conditions [17]. Flavor development during cooking poultry meat occurs due to amino acid and sugar interactions, lipid thermal oxidation, and thiamin degradation [18]. In a study, it was shown that roasting chicken meat after wrapping it with caseinate-starch edible film containing thyme and cumin oleoresin improved the tenderness and flavor of the meat. Especially the meat wrapped in the film containing oleoresin was found to be the most delicious because of the aroma migration from the film [19].

In this study, it was aimed to extend the shelf life of fresh chicken meat and add spice flavor to roasted meat by coated active film. Therefore spices (garlic, laurel leaf and black pepper), which are frequently used during the roasting of chicken meat, especially in the Middle East countries, were considered, and active films were prepared by using the extract or oil of these spices and salt.

2 Material and methods

2.1 Edible films production

Black pepper and bay laurel essential oil were obtained from Defne & Doga Company (Antalya, Turkey). Garlic was purchased from a local market and garlic juice was extracted from 50 grams of crushed garlic by mixing 100 ml of reverse osmosis water with a magnetic stirrer for 60 minutes. The juice was kept in the refrigerator for two hours and it was filtered using a 45µm wire sieve. The filtrate was filled in tubes and stored at 4 °C [20].

Plane Film (PF) prepared from sodium caseinate (Protap SHV6, Asturias, Spain) and potato starch (Avebe, Veendam, Netherlands) mixed (1:1) by the casting method. The mixture of sodium caseinate-starch (21 g) and glycerin (5.25 g) was added into distilled water (279 g) and was homogenized with a hand blender (Bosch, Germany) for 10 minutes. The mixture was heated with a magnetic stirrer (480 rpm, Memmert, Schwabach, Germany) to 200 °C for 30 minutes. For active film (AF) production salt (3.75 g), garlic extract (1.05 g), black pepper (1.05 g), and bay laurel essential oil (1.05 g) were added to the plane film solution and homogenized with a hand blender for 10 minutes. The solutions were degassed in a water bath at 90°C for 15 min and then casted onto polyvinyl chloride film and dried at 70°C on a reflectance windows dryer. Antibacterial effect and water vapor permeability of the films

Salmonella enterica subs. enterica serovar Typhimurium LT2 (DSM 18522) culture was grown in Tryptic Soy Broth medium at 37 °C for 48 hours. 0.1 ml of inoculum including in the range of 10^5-10^6 cfu/mL was evenly spread onto nutrient agar plates. Discs (20 mm) of plane and active films were put on the nutrient agar surfaces and then plates incubated at 37 °C for 24 h. The inhibition zone were measured in mm using a caliper [14].

Water vapor permeability (WVP) of the films was determined by modifying ASTM standard method E 96-95 (ASTM 1995). The film sample was fixed on top of the test cup (25 mm depth and 80 mm diameter) containing 90 ml water. The ring lid of the cup was fixed with a screw. The test cups were placed in an environmental chamber (ID 400 Nüve, Ankara, Turkey) with 2.5 m/s airflow rate at 25°C and 50% relative humidity. The cups were weighted for 12 hours at a 2-hour interval after a steady state was reached. The WVP was calculated according to Parris and coauthors [21].

2.2 Coating chicken meat

Skinless and boneless broiler chicken thigh meat (Banvit, Balıkesir, Turkey) was obtained the day after slaughter. Skinless and boneless thigh was completely coated with plane and active films. The wrapped and unwrapped (control) samples were put on expanded polystyrene trays, covered with stretch film, stored at +4 °C and analyzed on days 0, 4, 8, 11, 15 and 19.

2.3 Microbial analysis and pH value

Chicken meat (10 g) was aseptically weighted into a sterile bag, 90 ml of peptone water was added, and the mixture was homogenized for 1 minute in a stomacher (Stomacher 80 Biomaster, Seward Ltd., UK). Serial decimal dilutions of total mesophilic aerobic, psychrophilic and coliform bacteria were generated using 0.1% peptone water and spread on Plate Count Agar (Oxford, MS, USA) and Violet Red Bile Agar (Merck, Darmstadt, Germany). The plates were incubated at 30 °C for 24 hours for mesophiles, 4 °C for 10 days for psychrophiles, and 37 °C for 48 hours for coliform bacteria. Log10 colony forming units per gram of chicken meat (cfu/g) were used to calculate microbial counts [22].

The pH value of the meat samples was measured with a pH meter (WTW Multi 3410 Set 1, Weilheim, Germany) after homogenizing 10 g sample with 90 ml distilled water using a homogenizer.

2.4 Cooking loss, texture and sensory analysis of cooked chicken meat

After 4 days of storage, the wrapped chicken meats were roasted at 200 °C for 30 minutes in an electrical oven for evaluation of cooked meats. Cooking loss was calculated from the weight of samples before and after roasting [19].

The texture of roasted meat were measured using TA-XT Plus texture analyzer equipped with a Meullenet-Owens Razor Shear Blade (Stable Micro Systems, Surrey, England). The samples were sheared at right angles to the fiber axis - across the muscle fibers with a standard razor blade at a crosshead speed of 10 mm/s and to a depth of 10 mm. The maximum force (N) and total energy (N.mm) were recorded as shear force and shear energy, respectively [19].

The simple ranking test was used for sensory analysis. After the panelists were trained about the firmness and flavor of chicken meat, the chicken meat samples were presented to 19 panelists (Master and PhD students of the Department of Food Engineering, Akdeniz University). The hardness of the samples was first evaluated by ranking between 1 (least) and 3 (highest), then their flavor was ranked between 1 (best) and 3 (worst). The rank sums of samples were calculated and analyzed by Friedman's test. Fisher's least significant difference procedure was performed to determine significant differences between the samples [23].

2.5 Statistical analysis

The study was performed in two replications and analyses were made in duplicate. Except for sensory evaluation, the data were evaluated by analysis of variance (ANOVA) and Duncan's multiple range was used to determine significant differences between the samples [24].

3 Results and discussion

3.1 Antibacterial activity and water vapor permeability of the films

Chicken meat has long been considered as a significant source of Salmonella, which causes Salmonellosis, one of the most widespread foodborne diseases. *Salmonella enterica Enteritidis* and *Salmonella enterica Typhimurium* are responsible for most Salmonellosis in human. There has been a substantial decrease in the occurrence of salmonellosis due to effective Salmonella control strategies (including monitoring, bio-security, and immunization) in poultry and egg production in Europe. However, cases of salmonellosis are still common in developing countries [25]. For this reason, innovative approaches to prevent salmonella development in chicken meat maintain their importance.

The active film (AF) containing salt, garlic extract, black pepper, and laurel oil exhibited an antibacterial activity on Salmonella enterica Typhimurium. A zone of inhibition with a mean diameter of 33.60±0.58 mm was formed around the AF. However, no inhibition zone against Salmonella enterica Typhimurium was observed around the plane film. It was thought that the refractive window drying technique, which shortens the drying time of the films at the same temperature compared to convective hot air drying, contributed to the very high antibacterial activity of the AF film.

The antibacterial activity of spices extract and essential oils for Salmonella has been reported in many studies. 192 mg/mL garlic extract with 5 mm inhibition zone [26] and 5 mg/mL black pepper oil with 9 mm zone diameter [27] showed good antimicrobial activity on S. Typhimurium. Laurel oil has been found to exhibit strong antibacterial activity against a lot of food-born bacteria, including Salmonella[28]. The antibacterial effect of garlic extract results from organosulfur compounds (S-allyl cysteine, S-allylmercapto-L-cysteine, and S-methyl cysteine)[26], While the antibacterial effect of essential oils is due to monoterpenes and their derivatives (e.g. ß-caryophyllene, limonene and sabinene for black pepper [27] and e.g. 1,8-cineole, α-terpinyl acetate and sabinene for bay leaf oil[28]).

Water vapor permeability (WVP) of AF (1.921 ± 0.128 g.mm/kPa.h.m^2), was significantly higher than that of PF (1.386 ± 0.320 g.mm/kPa.h.m^2) ($p<0.05$). While the added laurel and black pepper oil in this study was expected to have a lowering effect on the WVP value of the AF film, the 17% salt content of the AF film increased both water vapor absorption and the gaps between the polymer chains, possibly causing an increase in the WVP of the film.

3.2 Microbiological analysis and pH value of the chicken meat

The counts of total mesophilic aerobic bacteria (TMAB), total coliform bacteria (TCB) and total psychrophilic aerobic bacteria (TPAB) raised continuously over storage time in both unwrapped and wrapped chicken meats. However, especially the active film significantly reduced the growth of TMAB, TPAB and TCB.

The change in the odor of chicken meat is closely related to microbial growth. Malodor in chicken meat is related to sulfur-containing compounds (hydrogen sulfide, dimethyl sulfide, and dimethyl disulfide) and biogenic amines (dimethylamine and trimethylamine), which are formed due to the decomposition of proteins by microorganisms [1].

TMAB in the control sample reached 8.91±0.21 log cfu/g, after 8 days of storage and the fresh chicken smell disappeared. After the 8th day, off-odor was detected in the control sample. Wrapping with AF film reduced TMAB count between 1.7 and 2.6 log cfu/g in chicken meats. Even the TMAB count (8.62±0.06 log cfu/g) of the AF wrapped sample after 19 days of storage was lower than that of the control sample on day 8 and the AF wrapped sample was free of malodor (Figure 1). Also, plain sodium caseinate-starch film partially reduced TMAB count and in the sample, off-odor was perceived after the 11th day. In the present study, malodorous in the chicken meat was detected when the TMAB number exceeded 9 log cfu/g. Gonzalez-Fandos and coauthors [29] previously reported that chicken leg meats were rejected when TMAB counts surpass 9 log cfu/g.

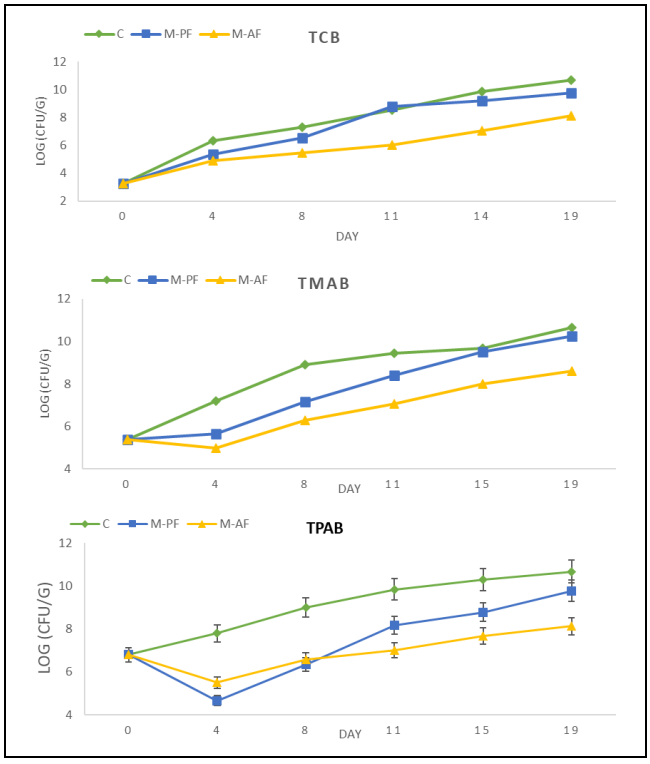

Figure 1. Changes in viable bacteria count during refrigerated storage. C: unwrapped meat, M-PF: meat wrapped in plane film, M-AF: meat wrapped in active film.

As with TMAB counts, the AF-wrapping of the chicken meat decreased the TCB counts between 1.45 and 2.55 log units at days 4 and 19 of storage, respectively. Also, PF wrapping reduced the TCB growth by around 0.9 log unites in chicken meat until 11 days. Both PF and AF suppressed the development of psychrophilic strains, even at days 4 and 8, the psychotropic aerobic bacteria (TPAB) count in the PF and AF-wrapped samples were lower than TPAB count (6.80 cfu/g) on day 0 (Figure 1). After 19 days, the PAB count of the AF wrapped meats increased only 1.25 log unite. Similarly, rainbow trout wrapped in pure and gelatin film including 1% bay essential oil had lower psychrophilic bacteria count than that of the control [15].

Covering the chicken meat completely with the edible films, which were reported to have low oxygen permeability in the

many kinds of literature [30], suppressed the growth of aerobic microorganisms, possibly by limiting the presence of oxygen in the package. In addition, garlic extract, laurel, and black pepper oil in the active film likely showed antimicrobial activity and further reduced the growth of bacteria.

Antimicrobial activities of garlic extract, laurel, and black pepper oil have been reported in the many studies [31],[32].

The pH value of fresh chicken often changes between 5.3-6.7 depending on the level of muscle glycogen at death [33]. Glycolysis is the main source of the decline in the pH value of meat after slaughter [34]. The pH value changes in chicken meat throughout storage are closely related to the number and type of bacteria affected by the initial flora and storage conditions. Under normal conditions, the pH of meat rises after a certain time due to volatile amine compounds, biogenic amines, and ammonia formed as a result of the activities of aerobic bacteria growing in meat [35].

However, when chicken meat is stored under low oxygen partial pressure, the initial flora of the meat shifts to one dominated by Gram-positive facultative anaerobic microflora (Lactic acid bacteria and Brochothrix thermosphacta). And the pH of the meat decreases as a result of the lactic, acetic and formic acid produced by these bacteria [36].

The pH values of the unwrapped chicken meat remained constant at around 6.8 for the first 8 days, and then the value began to increase with spoilage. However, the pH value of the samples wrapped in plain and active film decreased until the 8th and 12th day, respectively (Figure 2). After these days, the pH of the samples started to rise. The decrease in pH in wrapped meats indicated the growth of facultative aerobic bacteria, possibly due to the limited oxygen availability in the wrap. On day 19, the pH of the samples wrapped in the AF reached 6.58 ± 0.01, which is even lower than on day 0. The main reason for this was probably that AF film slowed down bacterial growth due to its antimicrobial activity, as seen in the bacterial counts. Similarly, the pH of chicken meat treated with rosemary and clove extract decreased from 5.65 to 5.48, while the pH of the control sample increased to 6.66 during storage at 4°C for 15 days [37].

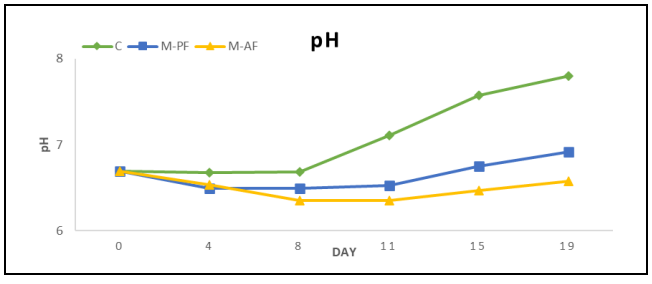

Figure 2. Change in pH values of the samples during storage at +4 °C.

3.3 Cooking loss, texture and sensory analysis of the cooked chicken meat

The cooking loss is result of water evaporation and fat that has melted and dripped from roasting meat tissue [38]. Compared to the unwrapped meat, wrapping with PF and AF reduced cooking loss about 11.8% and 9.1%, respectively (Table 1). The greater success of PF in reducing cooking loss may be due to its lower water vapor permeability and thus more inhibition of water evaporation during cooking. Similarly, Küçüközet and Uslu [19] were reported wrapping chicken meat in the film decreased cooking loss about 6.6-10.3%.

Table 1. Cooking loss, instrumental tenderness and sensory analysis results of roasted meats (means ± standard deviation).

Sample	Cooking Loss (%)	Shear Force (N)	Shear Energy (N.mm)	Hardness (Sensory)	Flavor
C	44.2±1.1^{a}	9.3±0.5^{a}	52.9±6.9^{a}	48^{a}	45^{a}
M-PF	32.4±3.5^{b}	6.2±1.8^{b}	40.4±11.6^{b}	29^{b}	40^{ab}
M-AF	35.1±4.6^{ab}	6.5±1.2^{b}	42.7±9.1 ^{b}	37^{ab}	29^{b}

The shear force and shear energy values determined by the Merlene-Owens razor shear blade technique have been reported to be a useful technique to evaluate the tenderness of chicken thigh [19] and breast meats [39]. Wrapping meat with the edible films significantly reduced the shear force and shear energy values of the cooked chicken meats ($p < 0.05$) (Table 1). This study and the previous study [30] showed that the decrease in these values was due to barrier property of the edible films against the water vapor and preventing cooking loss. The water vapor barrier property of edible films has been reported to improve moisture retention, juiciness, and texture of meat products. [40]. Spice extract, essential oil, or oleoresin added to the films did not have any effect on the tenderness of the meat.

The mean values of sensory hardness and flavour of roasted chicken meat are presented in Table 1. A significant effect of the films was noticed on the hardness ($p < 0.01$) and flavour of the chicken meat ($p < 0.05$). The chicken meat wrapped in the PF was evaluated the most tender sample because of lower cooking loss. However, meat wrapped in the AF was more appreciated by the panelists, due to a migration of salt and spice aroma from the film to the chicken meat.

4 Conclusion

This study revealed that active edible film with strong antimicrobial activity could be obtained by adding laurel, black pepper oil and garlic extract. Wrapping chicken meat with the active film is promising because it delays the deterioration of the meat by suppressing the growth of psychrophilic strains, many of which cause spoilage. Also, it increases the safety of the product by inhibiting growth of salmonella. Additionally, wrapping the chicken meat with active film will make the cooked chicken meat more tender and delicious.

5 Author contribution statement

In the conducted study, Yahya Imar was responsible for conducting the experiments and reviewing literature; Mustafa Kemal Uslu generating idea, designing the study and supervising the experiments; Ahmet Oktay Küçüközet assisted in conducting the experiments; and Firuze Ergin Zeren assisted in microbial analyses and evaluation of their results.

6 Ethics committee approval and conflict of interest statement

"Ethics committee approval is not required for the prepared article."

"In the prepared article, there is no conflict of interest with any person/institution."

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