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Effect of whey protein based edible coating on the microbial properties of mutton during cold storage

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***Abstract*— Food packaging, an important discipline in the area of food technology, concerns preservation and protection of foods, as well from oxidative and microbial spoilage. The objective of this research is evaluation of the effect of whey protein based edible coating on the microbial properties of fresh mutton after 0, 1, 3 and 5 days at refrigerator storage. The microbial properties (total count and psychrophilic bacteria) of the coated and uncoated samples were analyzed. The results showed that, total count, psychrophilic bacteria of the coated and uncoated samples did not have significant differences (p>0.05). Therefore whey protein based edible coating could not reduce the microbial load of fresh mutton.**

Keywords- Edible coating, Whey protein, Microbial properties, mutton, cold storage

I. INTRODUCTION

Man has many competitors for the food he produces. Animals, particularly rodents, insects and micro-organisms (moulds, yeasts and bacteria) all cause wastage at various stages in the growth, harvesting, processing, storage, transport and sale of food. These organisms, particularly bacteria, can affect food and render it poisonous to man by causing sickness and even death. The provision of food, which is good and safe, is essential to food industry and to a nation´s economic well-being food, in interesting variety, must be available all the year. It must be presented in a way that is convenient to purchase and use, and in most instances, this means that it must be packaged. Therefore, the packaging plays a decisive role in achieving the objectives of safety and waste prevention. Coatings involve formation of films directly on the surface of the objects they are intended to protect or enhance in some manner. In this sense, coatings become part of the product and remain on the product through use and consumption. The principle nature polymers utilized in forming films are a variety of polysaccharides, proteins and lipids. Due to the high consumption of meat and the fact that it is an excellent source of protein to dietary needs, meat packaging technology has evolved rapidly over the past two decades. In spite of the major developments in packaging materials and systems, the fundamental principles of packaging muscle foods remain the same. Packaging fresh meat is carried to delay spoilage, permit some enzymatic activity to improve tenderness, reduce weight loss and, where applicable, to ensure an oxy-myoglobin or cherry red color in red meat at retail or customer level [1]. Selection of a preservative packaging that is appropriate for a particular fresh meat product for a particular niche market requires the principal knowledge of deteriorative processes to which the product is subjected. The microbial population of fresh meat is affected by a number of factors such as species, health and handling of the live animal, slaughtering practices, chilling of the carcass and sanitation. The current traditional wrapping materials for fresh meat are only slightly permeable to water vapor, but highly permeable to oxygen and carbon-dioxide.Therefore, conditions for microbial growth are favorable.The spoilage bacteria of chilled meat stored is air dominated by Gram-negative, psychotrophilic, aerobic, rod-shaped bacteria. Although a wide range of genera are present on meat, pseudomonas, Acinetobacter and psychrobacter species are normally of importance, of which the species of *pseudomonas* are of the greatest importance. Pseudomonas Spp. typically accounts for >50% of the flora and sometimes for up to 90%. The spoilage of psychrobacter is not severally dangerous for man, but it changes the appearance and lowers shelf life of the meat products.

Therefore, our main objective in the present study was evaluated the effect of whey protein edible coating on the microbial properties of mutton during cold storage.

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II. MATERIALS AND METHODS

A. Materials

Whey protein concentrate (WPC) was obtained from Pooyan Food Supplementary Co. (Tehran, Iran). The WPC powder had a dry content of %75 ± 2 protein (N×6.35). The used mono-glyceride was mono-stearate from Spar-Uso, Spain. Edible glycerol (with the purity of %99), obtained from Merck Inc. (Germany), was used as a plasticizer in the formulation. Another chemical materials including peptone water, PCA media, ringer tablet, potassium iodide, boric acid, magnesium oxide, methyl red, bromocerzol and titrazole sulfuric acid was purchased from Merck Co. in Iran (Merck, Germany) for testing.

B. Film formation method

Film formulation was prepared according to McHugh et al [2], Perez-Gago et al [3], Miller et al [4], Anker et al [5] and Osses et al [6]. Aqueous solutions of 5% (w/w) WPC were prepared and heated at 90°C for 30 min. in water bath. The solutions were then cooled at room temperature and vacuum was applied to remove dissolved air. Weight glycerol (G) relative to the weight of WPC originally dissolved was then added as a plasticizer for film. Then the solutions were emulsified for 10 min at 140 rpm using a shaker. Weight mono-stearate glycerol relative to the half weight of WPC originally dissolved was then added and heated at 70°C for 30 min. The solutions were then cooled down at room temperature, emulsified and homogenized for 5 min at 500 rpm.

C. Coating fresh Mutton

Six-pack of 1Kg fresh muscles of mutton, each one containing 250 g four muscles wrapped in cellophane with pad absorbent were purchased from the meat packaging industries from (shahrvand store Tehran, Iran) and stored at 2°C overnight for testing [7,8]. After the solution formation, twelve samples were randomly selected and followed by immersion in formation solutions (1min). The Samples were then kept overnight in refrigerator for 1, 3 and 5 days at storage conditions with exposed surface area (23 ± 2°C, RH 50%). The interval between mutton purchase and sample coating was < 5 hr. The control samples were wrapped in cellophane in retail plastic trays with placing absorbent pads at the bottom of trays.

D. Statistical Analysis

SPSS version 15 was used for all statistical analyses. The using General Linear Model with replication measurement were tested and significance level ($\alpha = 0.05$) was applied.

III. RESULT AND DISCUSSION

Fig.1 and Fig.2 show the mean of log CFU/g values for the total microbial count of coated and uncoated samples after 0, 1, 3 and 5 days at refrigerator storage conditions. The results obtained from variance analysis showed no significant differences in total count and psychrophilic bacteria of coated and uncoated samples ($p>0.05$). Evaluation of the effect of edible whey protein-mono-glyceride film on the total microbial and psychrophilic bacteria counts showed that the total and psychrophilic bacteria counts were not significantly different. Although, the coating samples were consistently inclined to lower microbial load, but edible coating did not significantly ($p>0.05$) influence on the microbial count of fresh meat. At 5 days storage conditions, the coated samples had higher psychrophilic microbial count than the allowed limit (exceeds 10^7 CFU/g). This would suggest that bacteria off-flavor and drip which developed during the weighing of uncoated lamb samples. Therefore, the shelf-life of coated meat was through 3 day storage when compared to the uncoated samples.

The results of a study by Williams et al [9] revealed that calcium alginate film not have statistically significant effect on the microbial growth rate of beef cuts, so that the total aerobic plate counts (APC/g) increased overtime during the storage from the initial level of 1.4×10^4 APC/g for the coated beef cuts to the final level of 4.0×10^6 APC/g after 7 days storage at 5 °C.

Nortje et al [10] suggested that the casein-whey protein edible coating of raw, moisture and pickled with salt showed no significant effect on the rate of microbial growth. Protein films have lower Oxygen Permeability (OP). This may be related to their more polar nature and higher moisture content.

Lazarus et al [11] showed that at 2 days postmortem, the carcasses wrapped with plastic wrap had significantly higher microbial count ($\log_{10} = 3.65$) than both the control ($\log_{10} = 3.04$) and the alginate treated ($\log_{10} = 2.87$) carcasses. This difference was maintained through the days 5 and 7 with plastic wrap carcasses having a significantly higher microbial count than either the control or alginate treated carcasses. The elevated microbial count of plastic wrap carcasses was due to the reduction in surface evaporation, thereby maintaining a more favorable water activity (a_w) for growth. The calcium alginate coated carcasses tended to have lower surface microbial count from the sirloin area at all time periods, although not always significant. The ionic effect of calcium chloride would be expected to influence the microbial growth on the carcass surface.

Although protein-based films are good barriers to the transport of gases, the limited moisture barrier ability of protein-based films is mainly due to the hydrophilic nature of proteins. The barrier and mechanical properties of protein

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films are both affected by water absorption. Therefore, the film conditioning is an important factor when characterizing protein films, because relative humidity (RH) can substantially affect film properties. At low RH conditions (0 to %50), protein films are good oxygen barriers. However, McHgh et al [2] and Miller et al [4] suggested that at higher RH, their oxygen barrier ability decreases due to the plasticization effect of the absorbed water.

Also Mate et al [12]showed that the roasted peanuts coated with WPI/ glycerol (Gly) coatings had hexane levels reduced by more than %75 as compared to the uncoated nuts after 70 days of storage at %21 and %53 RH. Gontard et al [13] also concluded that the oxygen and CO2 permeability values of wheat gluten films increased substantially with increasing RH (%40-%100). The increase of permeability may be related to the hydrophilicity of wheat gluten due to its high content of amide groups. Interactions between water molecules and amide groups contribute to higher water content in the films as well as to a modification of the protein network structure. Disruption of hydrogen bonds may create additional sites for dissolution of oxygen and increase mobility of the gas molecules within the polymer bulk phase.

The results of this study showed that the effect of whey protein edible coating on the microbial properties of mutton during 0, 1, 3 and 5 days storage conditions had no significant different ($p>0.05$). An edible coating may be used as a vehicle for incorporating food additives such as antioxidants and antimicrobial agents onto the surface of the food, where deterioration of many solid foodstuffs by microbial growth or oxidation begins. Therefore, the inclusion of a preservative in an edible coating enrobed onto the product positions the preservative at the point of the food's greatest susceptibility to deterioration.

A. Figures

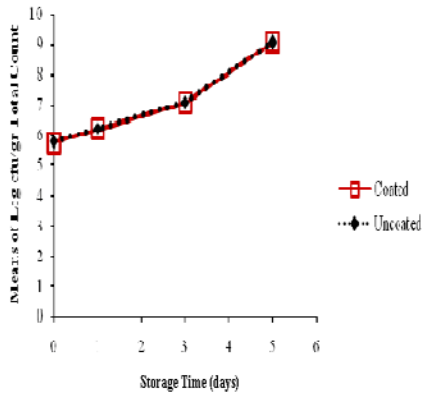

Fig1. Mean of log CFU/g values for the total microbial count

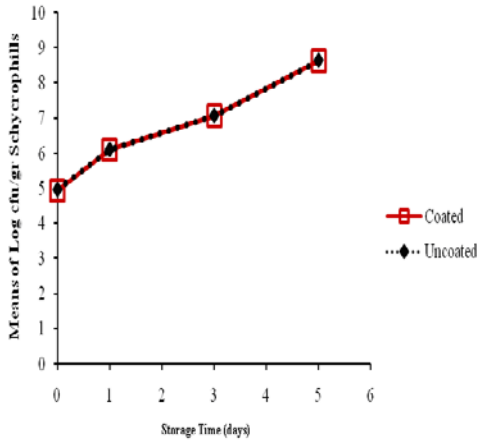

Fig2. Mean of log CFU/g values for the psychrophilic bacteria count

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