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Effects of salt and ammonium hydroxide on the quality of ground buffalo meat

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ABSTRACT

The objective of this study was to evaluate the effects of ammonium hydroxide (AH) and sodium chloride on the quality of ground buffalo meat patties. Ground buffalo meat was treated with distilled water (control), 0.5% v/w AH, 1.0% v/w AH, 2.0% v/w AH and 1.0% w/w sodium chloride was added for all the samples. Treatment with AH increased ($P<0.05$) the pH and water holding capacity (WHC) of ground buffalo meat patties during storage relative to their controls. Hunterlab a^* (redness) and *chroma* values increased ($P<0.05$) and *hue* decreased ($P<0.05$) in all AH treated samples in comparison to controls during storage. Ammonium hydroxide significantly ($P<0.05$) inhibited metmyoglobin formation compared to control after 3rd day of storage. There was a significant ($P<0.05$) reduction in thiobarbituric acid reactive substances (TBARS) values in all AH treated samples compared to control throughout storage. These results indicate the potential antioxidant and myoglobin redox stabilizing effect of AH in ground buffalo meat patties.

1. Introduction

Buffalo meat is primarily produced in India from old and unproductive animals resulting in coarse meat with tough texture and dark color. Such meat can be profitably utilized by comminuting and using in a value added, convenience meat products like nuggets (Thomas, Anjaneyulu, & Kondaiah, 2006), sausages (Sachindra, Sakhare, Yashoda, & Narasimha Rao, 2005), loaves (Suresh, Mendiratta, & Kondaiah, 2004), burgers (Modi, Mahendrakar, Narasimha Rao, & Sachindra, 2003) and patties (Suman & Sharma, 2003). It is a common practice in India to preserve excess/left over buffalo meat at the end of each day's production in the form of kheema, i.e. ground meat mixed with salt and other seasonings. However, grinding of meat leads to rapid formation of metmyoglobin, the undesirable brown color and oxidative rancidity, thus seriously affecting the consumer acceptance (Madhavi & Carpenter, 1993; Sahoo & Anjaneyulu, 1997).

While ground buffalo meat is a staple in many consumers' diets, a significant drawback to ground meat is its very short retail display life. Many studies have shown that refrigerated ground meats stored under atmospheric conditions has a maximum of 2–3 days of simulated retail display until it is deemed unacceptable by most consumers because it lacks a bright-red color (Gill & Jones, 1994). Ground meat is a commodity produced from trimmings and low value cuts sourced from many animals, and because of the manner in which it is produced, ground meat remains a potential source of food borne illness (Jimenez-Villarreal, Pohlman, Johnson, & Brown, 2003). The major deteriorative changes in ground meat; microbial contamination and growth and oxidation of myoglobin and lipids contribute to color changes, off-flavors and off-odors compromising safety. In order to address many of these concerns different researchers have demonstrated the use of wide variety of antioxidants, antimicrobials, color stabilizing agents, humectants, acidulants and alkylating agents, etc. to maintain and improve the quality of ground meats during storage and retail display at refrigerated temperature.

Ammonium hydroxide (NH_4OH) is a strong alkali and is being used in food industry in baked goods, gelatins/puddings, cheeses etc. Ammonium hydroxide (AH) is listed as generally regarded as safe (GRAS) by Food and Drug Administration (FDA) (21 CFR 184.1139) with no limitation other than current good manufacturing practices for uses as leavening agent, pH control agent, surface finishing agent, boiler water additive, food additive. Ammonium hydroxide is included in the Codex Alimentarius and as such may be used in variety of meat and meat products including comminuted meats under the conditions of good manufacturing practices (GMP) as outlined in the Preamble of the Codex GSFA. Ammonium hydroxide is approved for use in most countries of the world including the European Union (EU), Australia/New Zealand (approved as a processing aid), US, and most others.

Beneficial effects of AH in beef steaks in improving shear force value, tenderness, and sensory traits are recently reported by few researchers (Hamling & Calkins, 2008; Hamling, Jenschke, & Calkins, 2008a). For example Hamling, Jenschke, and Calkins (2008b) have determined the retail shelf-life of beef chuck and round muscles enhanced with AH, salt, and carbon monoxide (CO) and found that enhancement of beef chuck and round muscles can improve the color stability under high and low oxygen packaging conditions. Enhancement of meat pH with a solution containing AH, CO and salt has been shown to improve consumer

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palatability ratings (Everts et al., 2006; Nath et al., 2006). Moreover, Gupta, Garg, and Tiwari (1988) investigated the preservative effect of AH in ground goat meat stored at 37 °C, 4 °C and −20 °C and reported a significant reduction in aerobic plate counts in AH treated samples.

Even though preservative effect of AH has been reported in the literature, it is not clear how it affects myoglobin redox stability and lipid oxidation in ground meats. Although few studies have revealed the multifunctional uses of ammonium hydroxide in meat and meat products, no systematic works on their effect on different quality attributes of meat and meat products are reported. Hence, the objective of this study was to evaluate the effects of ammonium hydroxide and sodium chloride on the quality of ground buffalo meat patties under aerobic packaging conditions during refrigerated storage.

2. Materials and methods

2.1. Materials

The round portion of buffalo carcasses comprising of *Biceps femoris*, *Semitendinosus*, *Semimembranosus* muscles was procured fresh from municipal slaughter house, Hyderabad, India and transported to laboratory under chilled condition in ice boxes. The meat cuts were chilled at 4 °C for 24 h followed by removal of all connective tissue and fat and then subjected to mincing twice through 8 and 4 mm plates of a meat mincer (SCHARFEN, Model X70, 58413 Witten, West Germany). Fresh meat samples were obtained in batches separately for each of the three replications ($n = 3$). The ammonium hydroxide (AH) (Liquor Ammonia, NH_3, 25% solution, sp. gr. 0.91) was procured from Fisher Scientific (Qualigens Fine Chemicals, Mumbai, India). The sodium chloride (NaCl), 2-thiobarbituric acid, trichloroacetic acid, sodium phosphate dibasic and monobasic anhydrous salts, TRIS (Hydroxymethyl) Aminomethane Hydrochloride, 1,1,3,3-Tetramethoxypropane (TMP) was obtained from MERK, Mumbai, India. Peptone and Plate count agar were procured from HIMEDIA, Mumbai, India.

2.2. Ground buffalo meat patty preparation and storage

Freshly ground buffalo meat was divided into four batches of 1 kg each and hand mixed with 0.5%, 1.0%, and 2.0% v/w ammonium hydroxide. To ensure uniform mixing 0.5, 1.0 and 2.0% v/w ammonium hydroxide was diluted with distilled water at 5.0% v/w of meat to obtain a final concentration of 0.065 M, 0.13 M and 0.26 M respectively. For control only distilled water at 5.0% v/w of meat was added instead of ammonium hydroxide. Sodium chloride (1.0% w/w) was also added to all the four batches and mixed by hand for 5 minutes. From each batch eight patties (100 g, 10 cm diameter and 1.5 cm thickness) were hand formed using glass Petri plates. Two patties each were packaged in oxygen-permeable low-density polyethylene pouches and assigned to either 3, 6 or 9 days of storage at 4 °C and analyzed for pH, instrumental color, water holding capacity (WHC), thiobarbituric acid reactive substances (TBARS) and microbial counts. The 0' day samples were analyzed prior to packaging immediately after patty formation.

2.3. Analysis of pH and water holding capacity

The pH of ground buffalo meat patties was determined according to Strange, Benedict, Smith, and Swift (1977) with modifications. The pH was determined by blending 10 g sample with 50 ml distilled water for 60 s in a homogenizer (Model: MICCRA D8-Si, ART Moderne Labortechnik, D-79379 Mullheim, Germany). The pH values were measured using a standardized electrode attached to a digital pH meter (Thermo Orion, Model 420A+, USA).

Water holding capacity (WHC) was determined according to Wardlaw, McCaskill, and Acton (1973). Ground buffalo meat patty (20 g) was placed in a centrifuge tube containing 30 ml of NaCl (0.6 M) and was stirred with glass rod for 1 min. The tube was then kept at 4 °C for 15 min, stirred again and centrifuged at 3000 *g* (CPR-24, Remi Instruments, Mumbai, India) and 4 °C for 25 min. The supernatant was measured and WHC was expressed in percentage.

2.4. Analysis of surface color

Surface color analysis was performed using a Hunter lab Miniscan XE Plus colorimeter (Hunter Associates Laboratory Inc., Reston, VA, USA) on day 0 and during storage with 25 mm aperture set for illumination D65, 10º standard observer angle. CIE L^* (lightness), a^* (redness) and b^* (yellowness) were measured on the surface of raw buffalo meat patties from five randomly chosen spots. *Hue angle* [Tan^{-1} (b^*/a^*)] and *chroma* $(a^{*2} + b^{*2})^{1/2}$ were calculated according to Hunter and Harold (1987).

2.5. Determination of metmyoglobin

Myoglobin was extracted from raw patties using a modified procedure of Warris (1979). Samples were blended with 5 volumes of cold 0.04 M phosphate buffer at pH 6.8 for 10 s in a homogenizer. After keeping at 1 °C for 1 h, the mixtures were centrifuged at 3500 g (CPR-24, Remi Instruments, Mumbai, India) and 4 °C for 30 min. The supernatant was further clarified by filtration through Whatman No. 1 filter paper. The absorbance of filtrate was measured at 525, 572, and 700 nm using a UV-VIS spectrophotometer (Model: UV-1700 PharmaSpec, SHIMADZU, Japan). The % metmyoglobin (Met Mb) was calculated according to Trout (1989).

$$\%\ \text{Met Mb} = \{1.395 - [(A572 - A700) / (A525 - A700)]\} \times 100$$

Where "*A* <lambda> = Absorbance at <lambda> nm"

2.6. Analysis of thiobarbituric acid reactive substances

The thiobarbituric acid reactive substances (TBARS) of ground buffalo meat patties was determined by using the extraction method described by Witte, Krauze, and Bailey (1970) with slight modifications. Four gram sample was homogenized with 20% trichloroacetic acid solution (20 ml) and the slurry was centrifuged at 3000 g (CPR-24, Remi Instruments, Mumbai, India) for 10 min. Two ml of supernatant was mixed with equal volume of freshly prepared 0.1% thiobarbituric acid in glass test tubes and heated in water bath at 100 °C for 30 min followed by cooling under tap water. The absorbance of the mixture was measured at 532 nm using UV–vis spectrophotometer (Model: UV-1700 PharmaSpec, SHIMADZU, Japan) and the TBARS values were calculated using a TBA standard curve and expressed in mg malonaldehyde/kg. The TBA standard curve was prepared using 10^{-3} M stock solution (10–100 µl) of 1,1,3,3-tetramethoxypropane (TMP).

2.7. Microbiological evaluation

For determination of microbial counts, 10 g of meat sample was homogenized with 90 ml, 0.1% sterile peptone water. Serial 10-fold dilutions were prepared by diluting 1 ml of homogenate in 9 ml of 0.1% peptone water. Appropriate serial dilutions were duplicate plated (Pour plate method) with plate count agar for aerobic plate count (APC) and psychrotrophic count (PPC) and plates were incubated at 37 °C for 48 h for APC and 7 °C for 10 days for PPC (ICMSF, 1983).