EFFECT OF AMMONIUM HYDROXIDE ON DIFFERENT PHYSICO-CHEMICAL AND HISTOLOGICAL CHARACTERISTICS OF BUFFALO MEAT CHUNKS

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***Abstract*- This study was conducted to develop a method for improving tenderness of tough buffalo meat using ammonium hydroxide. Buffalo meat chunks from *Biceps femoris* muscle were marinated with distilled water (control), 0.1 %, 0.5% and 1 % (v/w) ammonium hydroxide for 48 hours at 4±1°C and subjected to various physico-chemical analysis and histological studies. Ammonium hydroxide treatment significantly ($P<0.05$) increased the pH, water holding capacity (WHC), collagen solubility, total and myofibrillar protein extractability and cooking yield. Significant ($P<0.05$) reduction in Warner-Bratzler shear force values were observed in all ammonium hydroxide treated samples compared to non-treated control. Electrophoretic pattern of muscle proteins also revealed reduction in the intensity and number of certain protein bands in all treated samples compared to control. These results suggest that ammonium hydroxide might be used to tenderize tough buffalo meat.**

Index terms: Ammonium hydroxide, Buffalo meat, tenderness

I. INTRODUCTION

Majority of buffaloes in India are slaughtered after completion of their productive period resulting in tough meat with poor quality characteristics. However, consumer research suggests that tenderness is a very important element of eating quality and the variations in tenderness affect the decision to repurchase (Maltin, Balcerzak, Tilley, & Delday, 2003). In order to increase the tenderness and other quality parameters marination technology has been widely used to improve palatability to increase the acceptance of lower-value cuts of meat (Naveena & Mendiratta, 2001; Naveena, Mendiratta, & Anjaneyulu, 2006).

Ammonium hydroxide is being used in food industry in baked goods, gelatins/puddings, cheese etc. Ammonium hydroxide 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. Beneficial effect of ammonium hydroxide in beef steaks in improving shear force value, tenderness, and sensory traits are recently reported by few researchers (Cerruto-Noya, Van Overbeke, & Mireles

DeWitt, 2009; Hamling & Calkins, 2008; Hamling, Jenschke, & Calkins, 2008). Even though, few published reports reveal the multifunctional uses of ammonium hydroxide in meat and meat products, no systematic studies on their effect on different quality attributes of meat and meat products are reported. Hence, this work was carried out to determine the tenderizing efficacy of ammonium hydroxide in buffalo meat chunks.

II. MATERIALS AND METHODS

Biceps femoris muscles from buffalo carcasses were collected within 2-3 hrs post slaughter from a selected retail meat shop of Hyderabad and stored in refrigerator at 4±1°C for 24 hrs. Uniform sized buffalo meat chunks (3 x 3 x 3 cm) were cut and divided into four batches of 1 Kg each. The 0.1%, 0.5% and 1.0% ammonium hydroxide (Liquor Ammonia, NH_3, 25% solution, sp. gr. 0.91) solution of pH 10.09, 10.46, and 10.6 respectively were prepared with distilled water and added to each 1 Kg batch @ 15% v/w. For control batch only 15% v/w of distilled water was added. After thorough mixing by hand, chunks were placed in polyethylene bags and kept at 4±1°C for 48 hrs. Four different treatments used were as follows:

(a) Control : 15 ml, distilled water
(b) 0.1% AH : 15 ml, 0.1 % ammonium hydroxide
(c) 0.5% AH : 15 ml, 0.5 % ammonium hydroxide
(d) 1.0% AH : 15 ml, 1.0 % ammonium hydroxide

After 48 hrs marination raw meat chunks were evaluated for pH, water-holding capacity (Wardlaw, Maccaskill, & Acton, 1973), hydroxyproline content (Nueman & Logan, 1950), collagen solubility (Mahendrakar, Dani, Ramesh, & Amla, 1989), protein extractability (Joo, Kauffman, Kim, & Park, 1999), muscle fibre diameter (Tuma, Venable, Wuthier, & Henrickson, 1962) and Sodium Dodecyl Sulphate-Polyacrylamide gel electrophoresis (SDS-PAGE) (Laemli, 1970). Cooked meat chunks were also evaluated for Warner-Bratzler shear force values. The overall experiment was replicated on three separate occasions. Statistical analysis was performed with the analysis of variance (ANOVA) using SPSS (SPSS version 13.0 for windows; SPSS, Chicago, IL, USA) and differences among mean values were obtained by Duncan's multiple range tests.

III. RESULTS AND DISCUSSION

The results of various Physico-chemical and histological analysis of control and ammonium hydroxide (AH) treated buffalo meat chunks are shown in Table 1. Significant (P<0.05) increase in pH was observed in all AH treated buffalo meat chunks compared to control. Water holding capacity increased (P<0.05) with increase in AH concentration and are significantly higher in all treatment samples than control. No difference in collagen content was observed between control and AH treatment groups, however significant increase (P<0.05) in collagen solubility was observed for all treated samples. With increase in AH concentration progressive increase in collagen solubility was observed. These findings are important as collagen degradation post mortem is quite limited and collagen content and its degradation are important determinants of eating quality (Maltin et al., 2003). Significantly (P<0.05) higher myofibrillar and total protein extractability was observed in all AH treated samples compared to control. On the other hand sarcoplasmic protein extractability was not influenced by AH treatment. The higher protein extractability in treated samples might be

due to increase in permeability of myofibrils which will disintegrate easily, however, in control samples regularly aligned filaments of myofibrils prevent buffer penetration, thus making action seemingly resistant to extraction (Naveena et al., 2004). The muscle fibre diameter of all the treated samples were significantly (P<0.05) lower than control sample. This is in accordance with findings of Tuma et al., (1962) who indicated that fibre diameter and tenderness are negatively related. SDS-PAGE (figures not shown) also revealed reduction in the intensity of few protein bands in treated samples compared to controls.

No significant difference in cooking yield was observed between control and treatments, except higher (P<0.05) yield for 1.0% AH treatment. Ammonium hydroxide treated buffalo meat chunks were significantly tender when compared to control as evidenced by lower (P<0.05) Warner-Bratzler shear force values. This increase in tenderness may be due to increased pH, as higher pH improves protein functionality in terms of WHC and tenderness (Cerruto-Noya et al., 2009).

IV. CONCLUSION

Marination of buffalo meat chunks with different levels of ammonium hydroxide for 48 hrs at 4±1°C has significantly increased (P<0.05) the pH, water holding capacity, total and myofibrillar protein extractability, collagen solubility and reduced (P<0.05) the Warner-Bratzler shear force values. These results suggest the action of ammonium hydroxide on both myofibrillar proteins and collagen tissue resulting in tenderization of buffalo meat.

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Table 1

Physico-chemical and histological qualities of buffalo meat chunks treated with different concentrations (0, 0.1%, 0.5%, and 1% v/w) of ammonium hydroxide (AH)

Parameters	Control	0.1% AH	0.5% AH	1.0% AH
pH	5.56±0.06^{a}	5.74±0.01^{b}	5.85±0.02^{bc}	5.89±0.04^{c}
Water-holding capacity (%)	15.33±0.33^{a}	20.67±1.76^{b}	23.00±2.08^{b}	25.00±1.26^{b}
Cooking yield (%)	53.53±0.71^{a}	55.47±0.32^{ab}	55.37±0.57^{ab}	55.60±1.02^{b}
Collagen content (mg/g tissue)	0.67±0.09^{a}	0.68±0.12^{a}	0.71±0.24^{a}	0.71±0.17^{a}
Collagen solubility (% total collagen)	44.18±0.98^{a}	64.38±0.85^{b}	73.59±0.70^{c}	87.6±0.88^{d}
Warner-Bratzler shear-force (kg/1.25 cm core)	14.27±1.19^{a}	11.04±0.96^{b}	10.24±1.09^{b}	8.67±0.60^{b}
Sarcoplasmic protein solubility (mg/g)	51.33±2.90^{a}	48.00±1.15^{a}	48.33±4.10^{a}	53.00±2.89^{a}
Myofibrillar protein solubility (mg/g)	90.67±4.05^{a}	108.67±4.67^{b}	117.33±2.91^{b}	116.00±2.00^{b}
Total protein solubility (mg/g)	142.00±1.15^{a}	156.67±3.53^{b}	165.67±1.20^{c}	169.00±1.53^{c}
Muscle fiber diameter (μ)	41.72±0.46^{a}	34.24±0.66^{b}	34.62±1.75^{b}	36.27±1.05^{b}

Means bearing same superscripts row-wise do not differ significantly (P>0.05)