DIFFERENTIATION IN THE PERFORMANCE OF PROTEIN CROSSLINKS INDUCED BY TRANSGLUTAMINASE IN THE MUSCLE FIBER OF CHICKEN AND BEEF

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Introduction

Microbial transglutaminase (MTG) induces crosslinking in meat proteins; many scientists have found the addition of MTG to be very important in the process of meat manufacturing. The proper ratio between MTG and muscle proteins can be determined through the use of certain techniques, such as the evaluation of ε(γ-glutamyl)lysine (G-L) content, in order to present a product with good textural properties. To the best of our knowledge no study has as yet explored the reaction manner of MTG to different meat types. We hypothesized that MTG catalyzes meat proteins in a different manner depending on meat type. In a previous publication (Ahhmed et al., 2007), the present authors observed a degree of variation in gel improvement level between chicken and beef sausages. This resulted from the variation in meat proteins in response to MTG, as well as the original glutamyl and lysine content. As a second step in the differentiation the gel improvements of meat products induced by MTG, the current study investigated whether the variation in gel improvement remains when MTG is added to the myofibrillar protein. The G-L content was determined in chicken and beef myofibrillar after adding MTG.

Materials and Methods

The thighs of chickens and the biceps femoris of Japanese black cattle were utilized in this research. Firstly, the cuts of the chicken and beef were minced in a meat grinder. The minced meats were homogenized with borate-KCl solution by a homogenizer. Eventually the homogenates were centrifuged with the use of a centrifuge at 12,000 rpm for 20 min. The homogenates were centrifuged consecutively three times by borate-kcl buffer, the supernatant of the homogenates were discarded and the upper layers of precipitations were removed and used as myofibrillar. The samples made of 50 g myofibrillar, 30 ml distilled water, 1.4 g NaCl, 0.21 g of sodium pyrophosphate and MTG was added at different levels (from 0.1 ml, 0.2 ml, until 1.0 ml) the addition of MTG was gradually increased gradually by 0.1 ml. The samples were divided into two groups, based on the heat treatments. The first group was incubated at 40°C for 30 min and the second group was cooked at 78°C for 30 min in a water bath. The samples were subjected to a textural test by a creep meter and further experiments such as evaluating the protein crosslinks through SDS-PAGE technique. The G-L content was determined by HPLC and protein concentrations were examined using the biuret method. Data expressed as mean ± SEM and one factorial analysis about MTG treatment was carried out by means of ANOVA, followed by Tukey method.

Results and Discussion

There is a definite need to determine the reason that MTG differently improves meat texture and how it acts with proteins in muscle fiber of different types of meat. This research was designed to access the exact level at which MTG acts properly with meat protein in chicken and beef muscle fiber. MTG was added at 10 levels from 0.1 ml until 1.0 ml; however this presentation illustrates MTG at just three levels. Remarkably, myofibrillar showed gradual developments in the gel strength by adding MTG. Figure 1 clarifies how MTG improves the texture in chicken and beef samples incubated at 40°C, though surprisingly the values of chicken samples undesirably decreased as MTG addition increased. However, the samples that were incubated in 78°C showed a significant ($P < 0.01$) increase (Figure 2).

In this research, beef samples generally showed a higher improvement than the chicken samples. In both heat treatments the MTG functions with beef proteins systematically. Samples of both meat types that treated with 1ml of MTG, in contrast to control samples, had significantly higher breaking strength values, a coveted increase in the present study. Progressively the breaking strength values were increased as the addition of MTG increases at all MTG levels. The molecular bands of MHC, measured by SDS-PAGE, were decreased by adding MTG (Figure 3). MTG was found to react positively with muscle fiber proteins. The majority of the MTG reactions occurred with proteins in the muscle fiber, not with water-soluble proteins, as protein concentrations were significantly reduced (*P*<0.01) by the action of the MTG in samples of chicken and beef which were dissolved in Guba-Straub-ATP (GS-ATP) solution in both meat types (Figure 4). G-L content was quantitatively determined through the use of biological assay techniques. Proteases used in this study were pronase, leucine aminopeptidase, prolidase, and carboxypeptidase, intentionally used to degrade all peptides to amino acids with the exception of the complex peptide of G-L. The G-L contents in the muscle fibers of both chicken and beef increased significantly in the treated samples at two level of MTG (data not shown). MTG reacts differently with meat proteins possibly as a factor of the age of the meat, the sarcolemma composition of the filaments and the original amount of amino acids that have the ability to react with MTG, such as glutamyl and lysine.

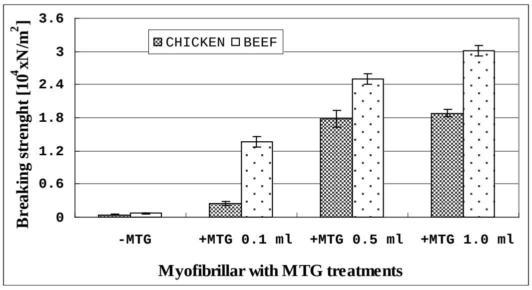

Figure 1. The breaking strength in muscle fiber proteins of chicken and beef induced by MTG. Samples incubated at 40°C.

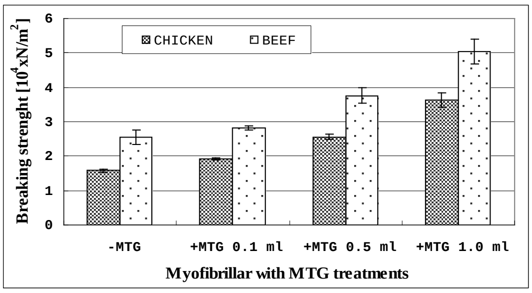

Figure 2. The breaking strength in muscle fiber proteins of chicken and beef as induced by MTG. Samples cooked at 78°C.

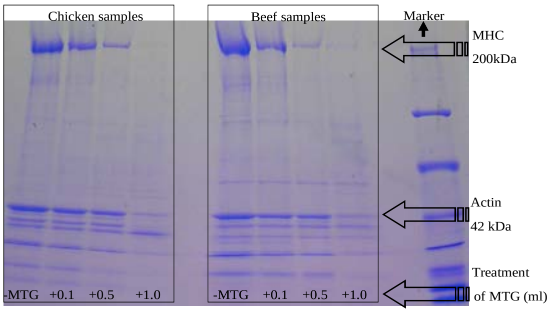

Figure 3. Changes in SDS–PAGE pattern in muscle fiber proteins of chicken and beef proteins. Samples were dissolving in GS–ATP solution.

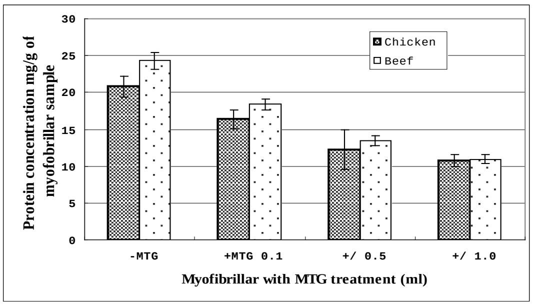

Figure 4. Protein concentrations of chicken and beef sausages as affected by MTG. Proteins were extracted in GS–ATP solution.

Conclusions

A difference was found in the improvement in the gel strength in chicken and beef myofibrillar even when these samples were treated using the same method. The values of the breaking strength in the chicken samples were inferior to that of the beef samples. MTG develops the texture of meat products by means of its strong reactant correlations with meat protein. This reaction takes place in the muscle protein, most likely between the amino acids of myofibrillar and the intramuscular proteins. However, this reaction is limited and it may strongly depend on the residue of amino acids, as suggested in the data collected from the chicken samples. It is dependent upon the amino acids in sarcolemma of myofibrillar filaments. Once the reactable amino acid residues are depleted (crosslinked) by MTG activity, the functions of MTG will be insignificant. Consequently, the protein cross-linking performances induced by MTG were based on the specificity and mount of amino acid within intramuscular of meat muscle. Values of G-L content were also increased in both meat types.

Data suggest that the textural characteristics were developed in a different manner between beef and chicken as affected by MTG. In conclusion, we suggest that MTG distinguishingly functions the muscle fiber proteins between chicken and beef.

Reference

1. Ahhmed, M. A., Kawahara, S., Ohta, K., Nakade, K., Soeda, T., and Muguruma, M. (2007). Differentiation in improvements of gel strength in chicken and beef sausages induced by transglutaminase. *Meat Science,* 76, 455-462.