

A NEW STRATEGY TO ENHANCE THE THERMAL STABILITY OF MYOFIBRILLAR PROTEIN AQUEOUS SOLUTION

Xing Chen¹, Youling L. Xiong^{2*}, Xinglian Xu^{1*} and Guanghong Zhou¹

¹ National Center of Meat Science and Safety Control, Ministry of Science and Technology, Jiangsu Synergetic Innovation Center of Meat Production and Processing, and College of Food Science and Technology, Nanjing Agricultural University, Nanjing, Jiangsu 210095, People's Republic of China;

² Department of Animal and Food Sciences, University of Kentucky, Lexington, Kentucky 40546, United States

*Corresponding author email: xlxus@njau.edu.cn (X. Xu); ylxiong@uky.edu (Y.L. Xiong)

I. INTRODUCTION

As nutritive food ingredients, meat proteins have not been exploited to the same extent as milk or soybean proteins, for example, as supplementary protein beverages or liquid diet to meet the demand of certain special populations, including those suffering from masticatory and swallowing dysfunctions (dysphagia). The major functional challenge hindering the development of muscle protein-based beverages is that myofibrillar protein (MP), the major protein fraction in muscle, is generally insoluble at low ionic strengths [1] as well as its strong susceptibility to heat-induced aggregation. In this study, we hypothesize that the dispersion effect of high pressure homogenization (HPH) combined with sulfhydryl (SH) blocking function of H₂O₂ [2] can inhibit the heat-induced aggregation of MP in neutral aqueous solution. Therefore, the objective of the present study was to investigate the effects of HPH and H₂O₂ treatment on the solubility of MP in aqueous solution upon heating (95 °C for 10 min).

II. MATERIALS AND METHODS

Myofibril isolations were carried out as described previously [3]. For HPH treatment, MP aqueous suspensions (15 mg/mL protein in 5 mM sodium phosphate, 1 mM EDTA, pH 7.0) were prepared at 4–6 °C and subjected to 69 MPa treatment according to our previous procedure [4]. For H₂O₂ addition, 0, 40, 80, 160 and 320 µmol/g of hydrogen peroxides (final concentrations) were added to MP and HPH treated MP (H-MP) samples. Protein solubility (2 mg/mL) was defined as the protein concentration of the supernatant divided by that of the original myofibril suspension. Total SH contents and disulfide bond content (in a 2 mg/mL MP solution) were determined based on the procedures of Liu *et al.* [5] and Thannhauser *et al.* [6], respectively.

III. RESULTS AND DISCUSSION

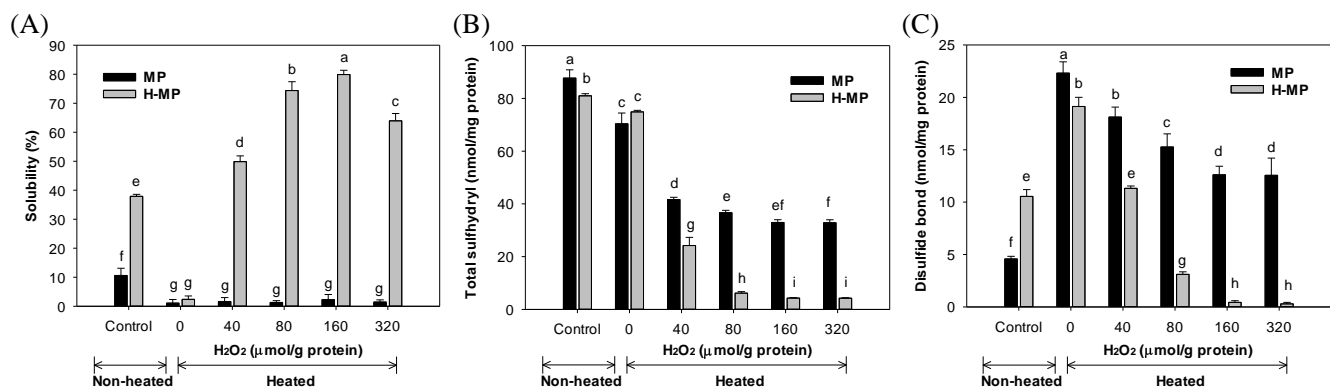


Figure 1. Effects of HPH and H₂O₂ on the solubility (A), total SH (B) and disulfide bond (C) of heated MP in neutral and low ionic strength solution. Control: Non-heated MP and H-MP. Means without a common letter differ significantly ($P < 0.05$).

By the addition of H₂O₂ (40-320 µmol/g protein), the solubility of H-MP increased remarkable ($P < 0.05$), with more than 75% of H-MP remained soluble after thermal treatment in the presence of 160 µmol/g H₂O₂, whereas no improvement in the solubility of heated MP occurred with or without the incorporation of H₂O₂ (Fig. 1A). It is hypothesized that the HPH and H₂O₂ might interfere with protein-protein association during heating, enhancing the solubility of heated H-MP.

The decrease of SH groups and formation of disulfides bridges are essential for the formation of aggregated structures of muscle proteins upon heating [7]. As shown in Fig. 1B and 1C, the addition of H₂O₂ remarkably reduced ($P < 0.05$) the total SH content and the formation of disulfide bonds, evidencing the blocking effect of H₂O₂. Thermal treatment can accelerate the oxidation of SH reacted with H₂O₂ to form sulfonic acid [8], thereby decreasing the total SH group content and irreversibly inhibiting the formation of disulfide bonds by masking the reactive group. Notably, H₂O₂ trapped the SH groups in a similar but more pronounced fashion in H-MP aqueous suspension than it did in MP sample, especially at the concentration of 160 µmol/g, most of the SH groups were blocked while the formation of disulfide bonds was completely inhibited in H-MP. It seemed that the blocking effect of H₂O₂ was facilitated by HPH pretreatment.

IV. CONCLUSION

The addition of H₂O₂ blocked the sulfhydryl (SH) groups, inhibited the formation of disulfide bonds, and suppressed thermal aggregation of MP. HPH facilitated the blockage effect of H₂O₂, leading to a further improved solubility of MP. HPH pre-dispersion combined with SH blockage proved to be an efficient strategy in suppressing the thermal aggregation of MP and enhancing the heat stability of MP in aqueous solution at neutral pH. It would be of interest to discover the high stability of MP towards heating at low ionic strength so that muscle proteins can be possibly utilized in tailor-making of protein-rich beverages for the people who have difficulty to masticate foods.

ACKNOWLEDGEMENTS

This study is supported by the USDA National Institute of Food and Agriculture (Hatch Project 1005724), the National Natural Science Foundation of China (No. 31671875), an Oversea Study Fellowship from the China Scholarship Council (to Xing Chen), and the Priority Academic Program Development of Jiangsu Higher Education Institutions (PAPD).

REFERENCES

1. Hultin, H. O., Feng, Y., & Stanley, D. W. (1995). A re-examination of muscle protein solubility. *Journal of Muscle Foods* 6(2): 91-107.
2. Sutariya, S., & Patel, H. (2017). Effect of hydrogen peroxide on improving the heat stability of whey protein isolate solutions. *Food Chemistry* 223: 114-120.
3. Ito, Y., Tatsumi, R., Wakamatsu, J. I., Nishimura, T., & Hattori, A. (2003). The solubilization of myofibrillar proteins of vertebrate skeletal muscle in water. *Animal Science Journal* 74(5): 417-425.
4. Chen, X., Xu, X. L., & Zhou, G. H. (2016). Potential of high pressure homogenization to solubilize chicken breast myofibrillar proteins in water. *Innovative Food Science & Emerging Technologies* 33: 170-179.
5. Liu, G., Xiong, Y., & Butterfield, D. (2000). Chemical, physical, and gel-forming properties of oxidized myofibrils and whey-and soy-protein isolates. *Journal of Food Science* 65(5): 811-818.
6. Thannhauser, T. W., Konishi, Y., & Scheraga, H. A. (1987). Analysis for disulfide bonds in peptides and proteins. *Methods in Enzymology* 143: 115-119.
7. Cortés-Ruiz, J. A., Pacheco-Aguilar, R., Ramírez-Suárez, J. C., Lugo-Sánchez, M. E., García-Orozco, K. D., Sotelo-Mundo, R. R., & Peña-Ramos, A. (2016). Conformational changes in proteins recovered from jumbo squid (*Dosidicus gigas*) muscle through pH shift washing treatments. *Food Chemistry* 196: 769-775.
8. Chang, K., Marshall, H., & Satterlee, L. (1982). Sulfur amino acid stability. Hydrogen peroxide treatment of casein, egg white, and soy isolate. *Journal of Food Science* 47(4): 1181-1183.