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

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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 H$_2$O$_2$ (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 H$_2$O$_2$, whereas no improvement in the solubility of heated MP occurred with or without the incorporation of H$_2$O$_2$ (Fig. 1A). It is hypothesized that the HPH and H$_2$O$_2$ 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 H$_2$O$_2$ remarkably reduced ($P < 0.05$) the total SH content and the formation of disulfide bonds, evidencing the blocking effect of H$_2$O$_2$. Thermal treatment can accelerate the oxidation of SH reacted with H$_2$O$_2$ 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, H$_2$O$_2$ 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 H$_2$O$_2$ was facilitated by HPH pretreatment.

IV. CONCLUSION

The addition of H$_2$O$_2$ blocked the sulfhydryl (SH) groups, inhibited the formation of disulfide bonds, and suppressed thermal aggregation of MP. HPH facilitated the blockage effect of H$_2$O$_2$, 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.

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