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Myofibrillar protein-dextran conjugates: Linking the improved functional properties to the molecular and physicochemical characteristics (#147)

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Introduction

In recent years, consumer demand for foods with improved and desirable functional attributes has attracted a great deal of attention. Hence, the food industry is increasingly focusing on the search for procedures to obtain new multifunctional ingredients. These techno-functional issues have led to increasing research interest and effort devoted to glycation (Maillard reaction), which is an efficient, economical and safe strategy for improving the functionalities of proteins. Myofibrillar protein (MP) accounts for approximately 55%-60% of total meat proteins. What's more, it is highly digestible and non-allergic. Besides, MP is abundant in high biological value. In this regard, the current work was undertaken to systematically investigate the effect of glycation on the molecular structure changes at conformation levels of MP using dextran (DX) with various molecular weights, and to ascertain the implications of these physicochemical and molecular changes on the solubility property of resulting conjugates.

Methods

Measurement of UV absorbance and browning extent

The UV-Vis analyses were performed using a spectrophotometer (UV-1800, Shimadzu, Kyoto, Japan). Samples were dissolved in PBS (20 mmol/L, pH 7.5) to a final concentration of 1 mg/mL, and each sample was scanned at wavelengths ranging from 200 to 450 nm.

Far-UV circular dichroism (CD) spectroscopy

The CD curves of the samples (0.3 mg/mL) were obtained in the 200 to 260 nm range with a CD spectrometer (Applied Photophysics, Surrey, UK). The -helix content (%) was calculated using CD Pro software and was obtained from a Chirascan spectrometer.

Solubility profile

The solubility of samples was evaluated by adjusting the pH of each solution (1 mg/mL) to pH 5.5 (isoelectric point) and pH 7.5 (reaction conditions). The solutions were centrifuged at 10,000 \times g for 15 min. The solubility was calculated as the percentage of the protein content remaining in the supernatant relative to the total protein content in the sample.

Results

Evidence for the formation of MP-DX conjugates

The changes in absorbance at 294 and 420 nm were presented in Fig. 1. The greatest intensity difference among the samples was observed at the absorbance peak at 294 nm and indicated the formation of a Schiff base.

Specifically, the extent of glycation increased as the molecular weight of the DX increased, indicating that the conjugation was dependent on the size of the DX.

Changes in the secondary structure of MP

The CD spectrum of non-treated MP exhibited two negative bands near 208 and 222 nm (Fig. 2), demonstrating the predominance of -helix motifs. Glycated MP showed a different CD pattern. Obviously, the magnitude of each of these two peaks was greatly decreased after glycation, Implying significant disruptions and losses of the structure of myosin -helix of light meromyosin (LMM). Typically, -helix structures are buried in the interior sites of polypeptide chains and are related to stability of protein. In this sense, evaluation of the extended secondary structure showed that glycation caused MP molecules to unravel and become more flexible and disordered. **Solubility**

Fig. 3 showed the solubilities of non-treated MP, MPH and the glycated MP samples at pH 5.5 and 7.5. Non-treated MP showed a solubility of 44.7% at pH 7.5. At pH 5.5, close to its isoelectric point, its solubility was lower than that at pH 7.5. There was no net charge on the protein around its isoelectric point, and thus, the MP tended to aggregate. With respect to the conjugation effect at pH 7.5 (reaction conditions), the solubilities of the glycated MP samples were significantly higher (P < 0.05) than non-treated MP, and notably, DX-70 showed the highest solubility (approximately ~88.5%). The considerable increase in solubility was well correlated to the reason that more hydrophilic groups were present on the surface of the MP due to DX attachment, which caused steric hindrance that could prevent interactions among MP molecules and improved the affinity between MP and water molecules, thereby increasing solubility. At pH 5.5, with increasing DX molecular weights, the solubility gradually increased. However, the difference between the solubilities of MP-DX 20 and 40 was guite small. Nevertheless, MP-DX 70 was significantly (P < 0.05) more soluble. In this regard, these solubility differences could be attributed to glycation induced the shift of the isoelectric point to a lower pH value.

Conclusion

The current work provides important information about the glycation of muscle protein in a liquid system. Distinct changes in the secondary structure increased the solubility of MP, a key functional attribute for muscle food processing applications. Overall, understanding the mechanism for improv



ing the functionality of MP via glycation would broaden the applicability of MP in the food industry.







ments.

Solubility changes Fig. 3. Changes in solubility of MP following different treatments at pH 7.5 and 5.5. Means with different letters (a-e) within the same parameter group differ significantly (P < 0.05).



 ${\bf UV}\ {\bf absorbance}\ {\bf Fig.\,1.}\ {\bf Changes}\ {\rm in}\ {\rm the}\ {\rm UV-Vis}\ {\rm absorbance}\ {\rm of}\ {\rm MP}\ {\rm following}\ {\rm different}\ {\rm treatments}.$

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