Preparation, Properties and Application of Glycated Casein in Food Maillard Reaction

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Abstract: Natural casein is recognized as a green and safe product in the world. It is a material with good nutritional value. It is mainly used to transport hydrophobic nutrients, so as to improve the internal solubility and emulsification stability, and enhance the probability of bioutilization. This content has shown a very wide range of application prospects in the current market development. On the basis of understanding the content of glycosylated casein preparation, this paper analyzed the specific results in depth according to the experimental method.

1. Introduction

Protein and polysaccharide are the most important constituent molecules in food raw materials, and their functional characteristics have always been the focus of researchers. Proteins have ideal emulsifying ability and stable system, and polysaccharides have been widely used in food development for their solubility and water retention levels. However, in the process of technological innovation and development, protein-polysaccharide covalent complex shows superior performance compared with other substances, and the covalent complex product can be obtained through glycosylation reaction. At the same time, no chemical reagents need to be added during the actual reaction operation, so this change is also called the green method[1-4].

In this paper, the preparation and application of glycolated casein based on food Maillard reaction was proposed. The main method was to prepare PRP wall material with stability by Maillard reaction, so the selected proteins in the experimental study were casein and two kinds of polysaccharides, and the ultrasonic dry Maillard reaction was used to graft the two proteins. Then the grafting degree, dissolution level and emulsification properties of the two were studied under different conditions. Table 1 and Table 2 below are the materials and equipment required for this experiment respectively:

Table 1 Experimental Materials

Name of experimental material	The manufacturer
Casein (Cascin, food grade)	Sinopharm Chemical Reagent Co. Ltd
Carrageenan (food grade)	Aladdin (Shanghai) Biochemical Technology Co., Ltd
Carboxymethyl Chitosan (food grade)	Sinopharm Chemical Reagent Co. Ltd
Potassium bromide (analytically pure)	Sinopharm Chemical Reagent Co. Ltd
Coomassie Bright Blue G-250 (Analytical Pure)	Aladdin (Shanghai) Biochemical Technology Co., Ltd
Phthalaldehyde (analytical pure)	Aladdin (Shanghai) Biochemical Technology Co., Ltd
Sodium lauryl sulfate (analytical pure)	Chengdu Kelong Chemical Reagent Factory
Borax (analytically pure)	Chengdu Kelong Chemical Reagent Factory
β-mercaptopropionic acid (analytical pure)	Aladdin (Shanghai) Biochemical Technology Co., Ltd
Disodium hydrogen phosphate (analytically pure)	Sinopharm Chemical Reagent Co. Ltd
Sodium dihydrogen phosphate (analytical pure)	Sinopharm Chemical Reagent Co. Ltd
Magnesium chloride (analytically pure)	Sinopharm Chemical Reagent Co. Ltd
Potassium carbonate (analytically pure)	Sinopharm Chemical Reagent Co. Ltd
Potassium iodide (analytically pure)	Sinopharm Chemical Reagent Co. Ltd

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Table 2 Experimental Equipment

Name of instrument or equipment	The manufacturer
UV-2550 type ultraviolet spectrophotometer	Shimadzu Corporation of Japan
Nicolet-380 Fourier infrared spectrometer	THCRMO Inc
DZF-6050 vacuum drying oven	Shanghai Jinghong Experimental Equipment Co., Ltd
DF-101S electric thermostatic heating magnetic agitator	Gongyi Yuhua Instrument Co., Ltd
DGG-9240B type electric heating constant temperature air blowing drying oven	Shanghai Senxin Instrument Co., Ltd
Model 5804R EPPCNDORF centrifuge	Ebender AG, Germany
F-7000 type fluorescence spectrophotometer	Hitachi Corporation of Japan
Model AL204 electronic balance	Mettler Toledo Instruments Co., Ltd
PHS - 3 g PH meter	Mettler - Toledo Instrument Shanghai Co., Ltd
Model ZD-9556 decolorizing shaker	Jiangsu jintan xinhang instrument factory
Alpha Chemiluminescence Gel Imaging System	Protcin Simplc, USA
Complete electrophoretic system	Bio-Rad (USA) Company
Settram differential scanning calorimeter, France	Shanghai Lyser Spectral Instrumentation Analysis Technology Co., Ltd

2. Experimental Methods

To prepare casein glycation products, the casein is mixed into a phosphate buffer solution with a pH of 7.4 and magnetized at room temperature for 2 and a half hours to form a casein equalization solution of a certain concentration. The ultrasonic probe is then placed 2cm below the protein level. Correct processing on the basis of clear ultrasonic power. After the samples are collected, the polysaccharides are added in the prescribed proportion, and the mixture is balanced before being freeze-dried for 48 hours. Finally, the obtained samples are ground into a powder and filtered. The resulting sample is placed in a 25mL beaker with a container temperature of 60°C and a pH value of 7.4 for the saturated potassium bromide reaction. The cooling reaction is terminated after the specified reaction time is reached to obtain the casein polysaccharide mixture[5-9].

After completing the preparation of experimental samples, the analysis and calculation of experimental methods should be carried out from the following points: First, the free amino group and the corresponding grafting degree should be calculated; Second, to detect the degree of Browning; Third, to effectively control the relative humidity; Fourthly, the emulsification properties of Maillard reaction products were measured comprehensively. Fifthly, the solubility of Maillard reaction products was measured.

3. Result Analysis

Combined with the analysis in the figure below, it can be seen that, as the substrate ratio in this experiment, it has a profound influence on the bonding of the reactive group between protein and polysaccharide molecules during the glycosylation reaction. In the experimental operation structure, the selection of appropriate substrate ratio can effectively improve the reaction speed and control the occurrence of adverse phenomena. The reason for this is that the reactive groups between casein and polysaccharides are covalently bound together, so the bonding can only be achieved if the two are in the right mix. In the figure, the grafting degree of Cas-Ca would change with the ratio of substrate, and finally showed the first increase and then the decline. When the substrate ratio reached 0.5, the grafting degree was the highest. However, Cas-CC would decrease with the increase of the mass ratio of protein and polysaccharide. When the ratio of the two reached 1:3, the actual graft degree reached the maximum. It can be seen that the ratio of the two should be 1:2 and 1:3 respectively in practical application.

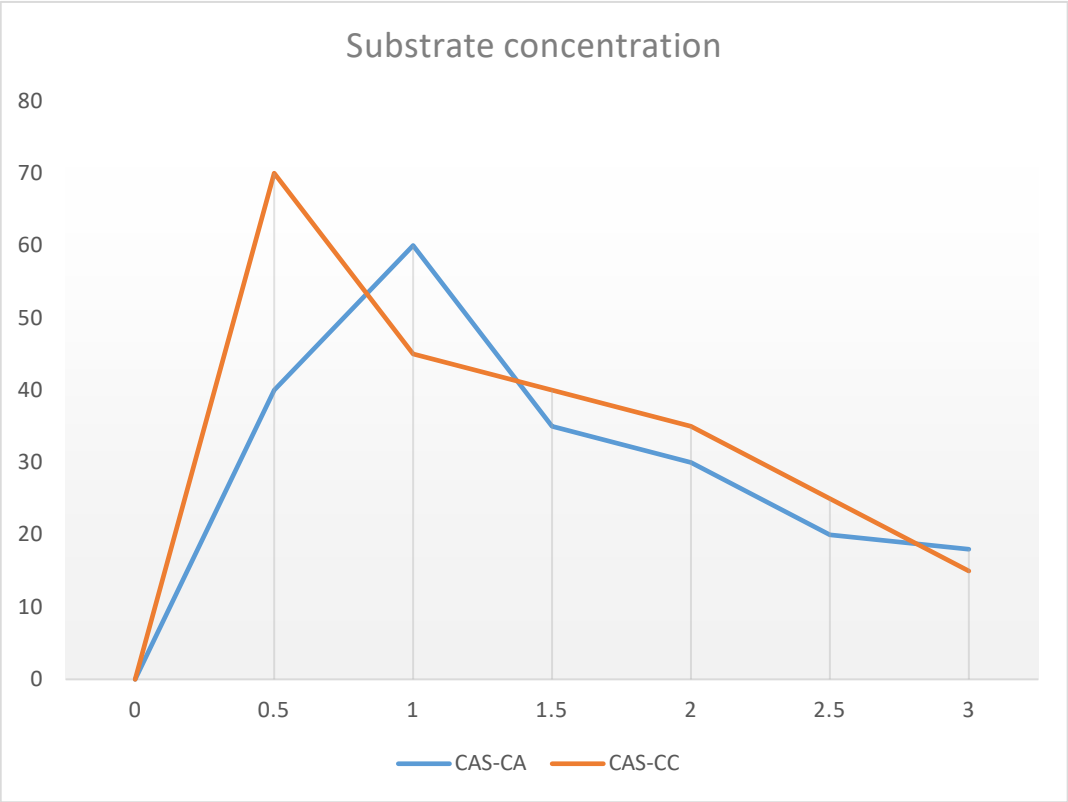

Fig.1 Analysis of the Influence of Different Substrate Ratio on the Degree of Grafting

In the analysis of the influence of relative humidity on the degree of grafting, the influence of this factor on the degree of grafting was also obvious. Combined with practical case studies, the glycosylation reaction is very fast between 30% and 80% humidity. The experiment designed in this paper mainly used a variety of saturated salt solutions to control humidity changes, and the proteoglycan mixtures were placed in different humidity environments, which were divided into 26%, 40%, 65% and 79%. The final results are as follows: First, the actual degree of grafting of Cas-Ca and Cas-CC increases with the increase of humidity. The reason for this phenomenon is that in a low humidity environment, the contact between proteins and polypeptide chains will be limited, and the probability of bond between groups is low. Second, in the context of continuously rising environmental humidity, with the increase of water content, the collision probability of protein and polysaccharide molecules will also be continuously increased. This increases the amount of water molecules, which inevitably causes the casein to swell more, increasing the flexibility and mobility of the polypeptide chain. In this process, the interaction between casein and polysaccharide can be further promoted, and more significant results can be achieved with the improvement of grafting degree. According to the analysis of experimental operation results, the optimal humidity needs to be controlled at 79%.

In addition to the above results, the reaction temperature and time, solubility, etc., were also discussed in depth. Finally, it was clearly recognized that the preparation process of casein - polysaccharide copolymer should ensure that the substrate ratio of Case-Ca graft reaction was 1:2, the reaction time was 24 hours, the reaction temperature was 60°C, and the humidity was 79%. The substrate ratio of CAS-CC should reach 1:3, the reaction time should be 36 hours, the reaction temperature should reach 60°C and the humidity should be 79%.

4. Conclusion

To sum up, the two brand-new glycosylated proteins proposed in the above experimental analysis have excellent value in practical application. Casein, for example, exhibits great stability and controlled release after covalently grafting polysaccharides through the Maillard reaction. Although there are few research projects and literature on this topic, with the continuous

improvement of scientific research and technology in various countries, it is inevitable that more valuable research ideas and contents can be found in the development of practice, so as to develop and prepare more glycosylated proteins with superior performance.

References

[1] Sun Y , Hayakawa S , Puangmanee S , et al. Chemical properties and antioxidative activity of glycated α-lactalbumin with a rare sugar, D-allose, by Maillard reaction[J]. Food Chemistry, 2006, 95(3):509-517.

[2] Chan W S , Yoo B . Preparation of milk protein isolate/κ-carrageenan conjugates by maillard reaction in wet-heating system and their application to stabilization of oil-in-water emulsions[J]. LWT- Food Science and Technology, 2020, 139:110542.

[3] Wang C , J Li, Li X , et al. Molecular forces and gelling properties of heat-induced gel from egg white protein glycated with isomalto-oligosaccharide[J]. Food Hydrocolloids, 2020, 99(Feb.):105356.1-105356.8.

[4] Li Q , H Sun, Zhang M , et al. Characterization of the flavor compounds in wheat bran and biochemical conversion for application in food[J]. Journal of Food Science, 2020, 85(3).

[5] Nooshkam M , Varidi M , Verma D K . Functional and biological properties of Maillard conjugates and their potential application in medical and food: A review[J]. Food Research International, 2020, 131:109003-.

[6] Wang C , Li J , Li X , et al. Emulsifying properties of glycation or glycation-heat modified egg white protein[J]. Food Research International, 2019, 119.

[7] Sun T , Qin Y , Xie J , et al. Effect of Maillard reaction on theological, physicochemical and functional properties of oat β-glucan[J]. Food Hydrocolloids, 2019, 89(APR.):90-94.

[8] Murata M . Browning and pigmentation in food through the Maillard reaction[J]. Glycoconjugate Journal, 2020.

[9] Jie H C , Hardacre A K , Parker M E . Effects of Maillard‐reacted beef bone hydrolysate on the physicochemical properties of extruded meat alternatives[J]. Journal of Food Science, 2020, 85(3).