Synthesis of Novel Amino Lactose and Evaluation of Its Antioxidant Property

Qing Li, Yan Feng, Guodong Gu, and Zhanyong Guo*

In order to improve the bioactivity of lactose, a novel lactose derivative is designed and synthesized by introducing an amino group to the C-6 of lactose. The in vitro antioxidant activities of synthesized lactose derivatives are assessed. It shows better scavenging activity against hydrogen peroxide (IC50 < 0.1 mg mL−1), hydroxyl radicals (IC50 0.55 mg mL−1), and DPPH radicals (IC50 0.37 mg mL−1) than lactose. Based on this data, it is reasonable to propose that the introduction of amino group through chemical modification is significant to enhance the free antioxidant activity of lactose. This derivatization strategy might provide an effective tool to broaden utilization of lactose.

1. Introduction

Reactive oxygen species (e.g. \( \text{O}_2^+ \), \( \text{OH} \), \( \text{H}_2\text{O}_2 \), DPPH etc.) are of great threat to the health as metabolic products of human body. It was found that their concentrations in blood were strongly related to various pathological events such as aging, cellular injury, and DNA degradation. For example, oxidative stress resulting from the toxic effects of free radicals on tissue played an important role in the pathogenesis of various neurodegenerative diseases such as Alzheimer’s disease, Parkinson’s disease, and ischemic-reperfusion injury.\(^{[1]}\) Therefore, it is of great interest among researchers to develop antioxidant supplements to help the human body reduce oxidative scratch. Moreover, restrictions over the use of synthetic antioxidants such as butyl hydroxy anisd (BHA) and butylated hydroxytoluene (BHT) in food further strengthen the concept of using naturally occurring antioxidants.\(^{[2]}\)

Lactose (\( \beta\)-D-galactosyl-D-glucose) is a necessary nutrition for infants and plays a vital role in the development of intelligence. As a natural disaccharide, it is only present in the milk of mammals. Raw lactose could be easily separated from milk or whey at low costs.\(^{[3–7]}\) Its related products are characterized by low sweetness, hygroscopicity, and high stability. With above benefits, lactose has been widely used as an ingredient in food and drug, especially in pharmaceutical industry as diluents of tablets and carriers of medicines.\(^{[8–9]}\)

Compared with lactose, lactose-based bioactive substances have similar characteristics and applications. They have been proven to be beneficial to human health (especially that of guts) as lactose.\(^{[10]}\) The presence of lactose matrix enables such derivatives to act as cross-linking agents upon oxidative enzymatic treatment in a cascade reaction.\(^{[11]}\) However, except as a main food source, native lactose has very limited industrial applicability for the lack of highly bioactive functional groups such as amino, sulfate ester, and carboxyl groups, compared with chitosan, heparin, and hyaluronic acid. In recent years, much attention has been paid to the study of novel materials, which could at least partially replace commodity polymers made from petroleum-based materials.\(^{[12–15]}\) Further evidences showed that, when lactose was drafted to chitosan, these derivatives would cause cell leakage and inhibit the production of enterotoxin.\(^{[16]}\) On the other hand, such derivatives benefited from ligands modified onto lactose and showed some other advantages.\(^{[17,18]}\) In order to effectively broaden the applications of new valuable products and materials based on lactose in non-food industry such as biomedical, cosmetics, and textile areas, lactose can be processed into modified lactose derivatives with chemical methods via the introduction of individual functional moieties to native lactose molecules.

Among the polysaccharide derivatives, aminated polysaccharides have received most attention for their interesting enhanced bioactivities, which have suggested the potential to be developed as valuable biomaterials. Because amino group is reductive and electropositive, most relative compounds can prevent or retard deleterious reactions, scavenge DPPH radicals, reduce Fe³⁺ cations, and inhibit lipid peroxidation effectively.\(^{[19,20]}\) In our previous work, it was found that amino inulin and relative derivatives exhibited obviously improved antifungal activity and antioxidant activity compared with inulin.\(^{[21–23]}\) These observations inspired us to modify lactose by introducing amino groups. With active amino groups, amino lactose was expected to have advantageous features, namely high antioxidant activity and/or high antifungal activity. As lactose is non-toxic, biodegradable, and biocompatible, amino lactose has the potential of becoming the alternative of free radical scavenger or fungistat in the food and medicine area.

In this study, 6-amino-lactose was synthesized by the introduction of amino group into the C-6 of lactose for the
first time. The aim of this study was to test the potential effects of amino group on antioxidant activities of lactose. The chemical structures of the derivatives were characterized by FTIR, \(^1\)H NMR, and \(^{13}\)C NMR. The antioxidant activities of lactose and the synthesized lactose derivatives were evaluated in vitro, and the relationship between structure and antioxidant activity of lactose was discussed.

2. Experimental Section

2.1. Materials

Anhydrous lactose was purchased from Aladin Chemical Corp (Shanghai, China), and was employed without further purification. N-bromobutanimide (NBS), acetic anhydride, sodium azide, triphenylphosphine (Ph\(_3\)P), and hydrazine hydrate were purchased from Sigma–Aldrich Chemical Corp (Shanghai, China). Other reagents are analytical grade and were supplied by Sinopharm Chemical Reagent Co., Ltd., (Shanghai, China).

2.2. Analytical Methods

The FT-IR spectra were measured on a Jasco-4100 Fourier Transform Infrared Spectrometer (Japan, provided by JASCO China, Co., Ltd., Shanghai, China) at 25°C in the transmittance mode. The samples were mixed with analytical grade KBr at a weight ratio of 1:100. All spectra were scanned against a blank KBr pellet back-ground by accumulating 16 scans with a resolution of 4.0 cm\(^{-1}\) in the range of 400-400 cm\(^{-1}\). The mass spectrometry (MS) spectra were collected on a Thermo Fisher LCQ Fleet Mass Spectrometer (Thermo Fisher Scientific, Waltham, MA). The \(^1\)H Nuclear Magnetic Resonance spectra (\(^1\)H NMR) was measured using a Bruker AVIII-500 Spectrometer at 25°C using DMSO-\(d_6\) as solvent. Chemical shifts (8 ppm) were referenced to tetramethylsilane (TMS). The solid-state \(^{13}\)C CP/MAS NMR spectra were obtained from a Bruker AC-300 spectrometer (Bruker BioSpin CN / Bruker (Beijing) Tech. and Serv. Co., Ltd. Beijing, China.).

2.3. Synthesis

2.3.1. Preparation of 6-Bromo-Lactose\(^{[25]}\)

Lactose (1.80 g, 5 mmol), 3.4 g NBS, and 5.7 g Ph\(_3\)P were added to 100 mL of N,N-dimethylformamide (DMF) and stirred for 30 min at 0°C. Then the mixture was heated and stirred at 70°C for 3 h under nitrogen atmosphere. The mixture was poured into 300 mL acetone and filtered by suction. The unreacted NBS, Ph\(_3\)P, and other outgrowth were extracted in a Soxhlet apparatus with acetone and ethanol for 48 h. The product was dried at 60°C for 24 h, yield: 65.1%.

2.3.2. Preparation of Hydrophobic 6-Bromo-Lactose (Intermediate Product A)

Hydrophobic 6-bromo-lactose (A) was synthesized as follows\(^{[21]}\): 6-bromo-lactose (5 mmol) and 10 mL acetic anhydride were dissolved in pyridine. After stirring at room temperature under nitrogen atmosphere for 12 h, the mixture was poured into 450 mL of distilled water. The precipitate was then filtered and washed with water and ethanol, respectively. The product was freeze-dried for 24 h, yield: 53.6%; MS (ESI, m/z): calculated [M+1]\(^+\) for C\(_{24}\)H\(_{15}\)Br\(_2\)O\(_5\): 718.01, found 719.13; \(^1\)H NMR (500 MHz, DMSO-\(d_6\)): \(\delta\) 1.91–2.11 ppm (CH\(_3\)-C = O), \(\delta\) 3.68–5.25 ppm (pyranose rings); FT-IR (thin film): \(\nu\) 1751 (C = O of acetyl).

2.3.3. Preparation of Hydrophobic 6-Azido-Lactose (Intermediate Product B)

Hydrophobic 6-azido-lactose (B) was synthesized as follows\(^{[26]}\): sodium azide (10 mmol) and A (5 mmol) were added to 80 mL of DMF at room temperature. After stirring at 80°C for 4 h under nitrogen atmosphere, the mixture was cooled to room temperature and slowly poured into 200 mL of distilled water. The precipitate was collected, washed with water and ethanol, respectively. After Soxhlet extraction with water for 48 h to remove the probable remained sodium azide, the obtained product was freeze dried in vacuum for 24 h, yield: 53.6%; MS (ESI, m/z): calculated [M+1]\(^+\) for C\(_{24}\)H\(_{24}\)N\(_2\)O\(_{14}\): 644.19, found 645.16; \(^1\)H NMR (500 MHz, DMSO-\(d_6\)): \(\delta\) 1.91–2.11 ppm (CH\(_3\)-C = O), \(\delta\) 3.67–5.25 ppm (pyranose rings); FT-IR (thin film): \(\nu\) 2105 (C-6-azido), \(\nu\) 1751 (C = O of acetyl).

2.3.4. Preparation of Hydrophobic 6-Amino-Lactose (Intermediate Product C)

Hydrophobic 6-amino-lactose (product C) was synthesized as follow\(^{[27-28]}\): Ph\(_3\)P (10 mmol) and B (5 mmol) were dissolved in 200 mL of anhydrous acetone and stirred for 24 h at room temperature under nitrogen atmosphere. Thereafter 1 mL of water was added and the solution was stirred for another 24 h. The solvent was removed by reduced pressure distillation. The product was washed with ether and freeze dried in vacuum for 24 h, yield: 63.4%; MS (ESI, m/z): calculated [M-1]\(^+\) for C\(_{24}\)H\(_{32}\)Br\(_2\)O\(_{15}\): 718.01, found 719.13; \(^1\)H NMR (500 MHz, DMSO-\(d_6\)): \(\delta\) 1.75 ppm (CH\(_3\)-N\(_2\)), \(\delta\) 1.91–2.11 ppm (CH\(_3\)-C = O), \(\delta\) 3.67–5.25 ppm (pyranose rings); FT-IR (thin film): \(\nu\) 2105 (C-6-amido), \(\nu\) 1751 (C = O of acetyl).

2.3.5. Preparation of 6-Amino-Lactose\(^{[21]}\)

Hydrazine hydrate (30 mmol) and C (5 mmol) were dissolved in ethanol and stirred at 80°C for 4 h. The solvent was evaporated under reduced pressure. The product was washed by ethanol, and freeze-dried in vacuum for 24 h, yield: 74.2%; MS (ESI, m/z): calculated [M-1]\(^+\) for C\(_{24}\)H\(_{36}\)N\(_2\)O\(_{15}\): 592.21, found 591.11; \(^1\)H NMR (500 MHz, DMSO-\(d_6\)): \(\delta\) 1.75 ppm (CH\(_3\)-N\(_2\)), \(\delta\) 1.91–2.11 ppm (CH\(_3\)-C = O), \(\delta\) 3.67–5.25 ppm (pyranose rings); FT-IR (thin film): \(\nu\) 1751 (C = O of acetyl), \(\nu\) 1590 (C-6-NH\(_2\)).

2.4. Determination of Antioxidant Ability

2.4.1. Hydroxyl-Radical Scavenging Ability Assay

The hydroxyl-radical scavenging ability was measured according to our previous method as follows\(^{[29]}\): a solution of 60 μM H\(_2\)O\(_2\)
was first prepared in phosphate-buffered saline (pH = 7.4). Lactose or lactose derivatives solution at different concentrations (1 mL), was mixed and incubated with safranine O (0.22 mM, 0.5 mL), EDTA-Fe$^{2+}$ (0.22 mM, 0.5 mL), H$_2$O$_2$ (60 μM, 1 mL), and potassium phosphate buffer (0.15 M, pH 7.4, 1 mL) for 30 min at 37 °C. The absorbance was recorded immediately at 520 nm. In the blank, samples were substituted with distilled water. Meanwhile, in the negative control, H$_2$O$_2$ was substituted with potassium phosphate buffer. The hydroxyl-radical scavenging ability of the products was calculated based on the following formula:

\[
\text{Scavenging effect (\%)} = \left[1 - \frac{(A_{\text{control} \ 520 \ \text{nm}} - A_{\text{sample} \ 520 \ \text{nm}})}{(A_{\text{sample} \ 520 \ \text{nm}} - A_{\text{blank} \ 520 \ \text{nm}})}\right] \times 100
\]

where $A_{\text{blank} \ 520 \ \text{nm}}$ is the absorbance of the blank at 520 nm, $A_{\text{control} \ 520 \ \text{nm}}$ is the absorbance of the control at 520 nm, and $A_{\text{sample} \ 520 \ \text{nm}}$ is the absorbance of the sample at 520 nm.

### 2.4.2. DPPH-Radical Scavenging Ability Assay

The DPPH-radical scavenging capacity was evaluated following Tan's method [30]. The test compound (lactose or lactose derivatives) and ethanol solution containing DPPH (180 μM, 2 mL) were stored in complete darkness at 25 °C for 30 min. Then the absorbance of the remaining DPPH radical was recorded at 517 nm. The DPPH-radical scavenging effect was obtained as a decrease in the absorbance based on the following formula:

\[
\text{Scavenging effect (\%)} = \left[1 - \frac{(A_{\text{control} \ 517 \ \text{nm}} - A_{\text{sample} \ 517 \ \text{nm}})}{(A_{\text{sample} \ 517 \ \text{nm}} - A_{\text{blank} \ 517 \ \text{nm}})}\right] \times 100
\]

where $A_{\text{sample} \ 517 \ \text{nm}}$ is the absorbance of the sample at 517 nm, $A_{\text{blank} \ 517 \ \text{nm}}$ is the absorbance of the blank at 517 nm and $A_{\text{control} \ 517 \ \text{nm}}$ represents the absorbance of the control (distilled water instead of DPPH) at 517 nm.

### 2.4.3. Hydrogen Peroxide Scavenging Ability Assay

The hydrogen peroxide scavenging capacity was tested following the method of Ruch, et al. [31]. Briefly, a solution of 10 mM H$_2$O$_2$ was first prepared in phosphate-buffered saline (pH 7.4). Lactose or lactose derivatives were dissolved in the solution above and mixed with 0.1 M phosphate buffer (pH 7.4) subsequently. After incubation at 37 °C for 10 min, the absorbance of final solution was measured at 230 nm. The scavenging effect was calculated according to the following equation:

\[
\text{Scavenging effect (\%)} = \left[1 - \frac{(A_{\text{control} \ 230 \ \text{nm}} - A_{\text{sample} \ 230 \ \text{nm}})}{(A_{\text{sample} \ 230 \ \text{nm}} - A_{\text{control} \ 230 \ \text{nm}})}\right] \times 100
\]

where $A_{\text{sample} \ 230 \ \text{nm}}$ is the absorbance of the sample at 230 nm, and $A_{\text{control} \ 230 \ \text{nm}}$ denotes the absorbance of control (distilled water instead of H$_2$O$_2$) at 230 nm.

The antioxidant activity was expressed as IC$_{50}$, which was defined as the concentration of compound required for inhibition of the radical formation by 50%. Vitamin C was used as the positive control. The IC$_{50}$ was calculated using Excel software. Each experiment was performed in three replicates and the data were expressed as mean and standard deviation (SD). Significant difference analysis was performed using Duncan’s multiple range test. A level of $P < 0.05$ was considered as statistically significant.

### 3. Results and Discussion

#### 3.1. Chemical Analysis of the Starch Derivatives

It was reported that NBS and Ph$_3$P could selectively replace primary hydroxyl groups of polysaccharide with bromine [32]. Therefore 6-bromo-lactose was selected as an intermediate to activate the primary hydroxyls of lactose for azide to replace with. Then we protected the secondary hydroxyls of the 6-bromo-lactose with acetic anhydride to bring hydrophobic groups into lactose backbone. The introduction of hydrophobic groups could effectively reduce the water solubility of lactose derivatives (A, B, and C), which would facilitate the removal of inorganic salt and acetic anhydride by simply washing the products with water. Figure 1 shows the FTIR spectra of lactose, intermediate products (A, B, and C), and 6-amino-lactose (D).

![Figure 1](image_url)
was transformed to amino and a new peak at about 1590 cm⁻¹ appeared. After the deacylation of C, a strong peak at 1600 cm⁻¹ shows up and was assigned to the characteristic absorbance of C-6-NH₂ in the spectrum of 6-amino-lactose. The spectra were identified to be consistent with corresponding molecules shown in Scheme146.1.

The ³¹H NMR spectra of lactose, 6-amino-lactose (D), and intermediate products (A, B, C) are shown in Figure 2 for comparison. The signals at 3.59–5.29 ppm were assigned to the protons of inulin. The new strong signals, which appear at 1.91–2.11 ppm, can be assigned to the protons of CH₃-C=O for intermediate products A, B, and C. When the C-6-azido in C was transformed to amino, a new signal at 1.75 cm⁻¹ appears, which is assigned to N-H.\[21,23\] In the ¹³C NMR spectra (Figure 3) of A, B, and C, methyl carbon of O=C-CH₃ is observed at 20 ppm, and carbonyl carbon of O=C-CH₃ is observed as two peaks at 161 and 170 ppm.\[33\] Meanwhile, compared with A, B, and C, the peaks at 20, 161, and 170 ppm disappear when the O=C-CH₃ was transformed to OH for 6-amino-lactose (D). The ¹³C-NMR spectra of lactose, all the intermediate products, and 6-amino-lactose further confirmed the success of the preparation. The MS spectra of 6-amino-lactose (D) and intermediate products (A, B, C) are provided in supporting information (Figure S1, Supporting Information). All of those spectra data indicated that 6-amino-lactose was synthesized successfully.

### 3.2. Antioxidant Activity

The antioxidant capability of lactose and lactose derivatives were evaluated using different assays and the results are shown in Figure 4 through Figure 6.

![Scheme 1. Synthetic pathway of amino lactose.](image1)

![Figure 2. ¹H NMR spectra of lactose and lactose derivatives.](image2)
Hydrogen peroxide formed by two-electron reduction of O₂ is not a free radical but is an oxidizing agent. In the presence of O₂ and transition metal ions, H₂O₂ can generate OH⁻ via Fenton reaction. In addition, H₂O₂ can easily cross the cell membrane and exerts injurious effects on tissues through a number of different mechanisms, such as perturbing intracellular calcium homeostasis, inducing DNA damage, and inducing apoptosis.⁴⁻⁶ Figure 4 shows the scavenging capacity of lactose, hydrophobic 6-amino-lactose (C), and 6-amino-lactose on hydrogen peroxide. The scavenging ability of lactose is weak, and the maximum inhibitory activity in the test concentration range (0.1–1.6 mg mL⁻¹) is 3.6%. 6-Amino-lactose shows more potent scavenging activity (IC₅₀ <0.1 mg mL⁻¹) than lactose. To explain whether the inhibitory effects of lactose and the amino lactose on the protective effect on H₂O₂-induced cell damage was due to the scavenging of free radicals, it was essential to evaluate whether these compounds were able to scavenge the free radicals using assay systems such as the DPPH radical-scavenging assay and the hydroxyl radical-scavenging assay. Hydroxyl radicals, generated by the reaction of Fe-EDTA complex with H₂O₂ in phosphate buffer, are harmful to the body through reacting with biological molecules such as amino acid and DNA.⁶⁻⁷ Figure 5 shows the *OH scavenging activity of lactose, hydrophobic 6-amino-lactose (C), and 6-amino-lactose at various concentrations. All the compounds show a positive correlation between scavenging effects versus concentrations. 6-Amino-lactose and hydrophobic 6-amino-lactose (C) show stronger antioxidant activity (IC₅₀ 0.55 mg mL⁻¹ and 0.63 mg mL⁻¹, respectively) than lactose (IC₅₀ 0.98 mg mL⁻¹). On the basis of IC₅₀, it was obvious that the amino lactose showed much better antioxidant activity due to the introduction of amino functional group. This observation was in accordance with the conclusion that aminated derivatives of saccharide were more.
potent than natural saccharide as a scavenger of hydroxyl radicals.[8]

The scavenging effect of lactose, hydrophobic 6-amino-lactose (C), and 6-amino-lactose against DPPH radicals are shown in Figure 6. This assay provided information on the reactivity of the compounds with a stable free radical. DPPH shows a strong absorption band at 517 nm in visible spectroscopy because of the odd electron. The absorption would vanish when the odd electron was paired off in the presence of a free radical scavenger.[1] Lactose showed relatively weak scavenging activity against DPPH radical, and the scavenging index is 13% at 1.6 mg/mL. Hydrophobic 6-amino-lactose (38% at 1.6 mg mL\(^{-1}\)) and 6-amino-lactose (IC\(_{50}\) 0.37 mg mL\(^{-1}\)) indicated much stronger DPPH-radical scavenging ability compared with lactose. The bleaching of DPPH absorption is the representative of compound capacity to scavenge free radicals independently from any enzymatic activity. Our results clearly demonstrated that amino lactose was effective in scavenging DPPH radicals. The result was similar to that of scavenging test against hydroxyl radical.

Free radical chain reactions may be inhibited by adding preventive antioxidants that can retard the formation of free radicals or stabilize free radicals. The amino group may play an important role to act as an electron donor to quench free radicals by providing an electron, conceivably via an electron attack on the free radicals.[30,39] The stronger electron-donating groups tend to donate more electrons to quench more reactive free radicals, which may help stabilize the free radicals’ form.[30,39] These may explain why amino-lactose has better free radical scavenging activity than lactose.

On the other hand, the inconsistent relative antioxidant activity of amino lactose against different antioxidants may be related to the different reaction mechanisms in different systems. There are some fundamental differences among the three assays. First, the features of the oxidant such as their redox potentials or stability are not the same. The scavenging effect on DPPH radicals represents direct radical scavenging activity. However, in the hydroxyl radical and hydrogen peroxide scavenging assay, hydroxyl radicals are generated by the Fenton reaction and the inhibition could be attributed to the inhibition of radicals or the Fe\(^{3+}\) chelating effect of the test compounds. Second, other factors such as the surface activity affected by the polymer structures and the different reaction mechanisms in different systems may also affect the ability of test compounds to react with and quench different radicals.[41]

4. Conclusion

In this paper, we reported a five step method to synthesize 6-amino lactose from lactose. The antioxidant activities of lactose and relative derivatives were evaluated by scavenging effects against hydroxyl radicals, DPPH radicals, and hydrogen peroxide. The test results showed that amino lactose had increased antioxidant activities than lactose. These data demonstrated that the chemical modification of lactose with amino group would lead to enhancement of the biological activity against some radicals. We propose that the enhancement is likely resulted from the free electron provided by the amino group, which is an efficient electron donor. The test results showed that the bioactivity of lactose could be improved after appropriate derivatization, which might bring broader usage to lactose.

Abbreviations

\(^1\text{H}\) NMR, \(^1\text{H}\) Nuclear Magnetic Resonance spectrometer; \(^13\text{C}\) NMR, \(^13\text{C}\) Nuclear Magnetic Resonance spectrometer; DPPH, 1,1-diphenyl-2-picylhydrazyl; DMF, N,N-Dimethylformamide; DMSO, dimethyl sulfoxide; EDTA, ethylenediaminetetraacetic acid; FTIR, fourier transform infrared spectroscopy; NBS, N-bromobutanimide; Ph3P, triphenylphosphine.

Supporting Information

Suporting information is available from the Wiley Online Library or from the author.

Acknowledgements

The authors are grateful to the National Natural Science Foundation of China (41376156), Natural Science Foundation of Shandong Province of China (ZR2017BD015), and Yantai Science and Technology Development Plan (2015ZM078), and the Public Science and Technology Research Funds Projects of Ocean (No. 201500022-3).

Conflict of Interest

The authors have declared no conflicts of interest.

Novelty Statement

This work investigated the synthesis and antioxidant activity of amino lactose. In the synthesis process, the introduction of lipophilic groups simplified the purification conditions, and the introduction of amino group improved the biological activity of lactose derivatives. After the chemical modification, amino lactose showed better antioxidant activity than lactose. This synthesis provided a possible method to develop further use of lactose.

Keywords

amino lactose, antioxidant activity, chemical modifications

Supporting Information

Received: November 14, 2017
Revised: January 9, 2018
Published online: March 24, 2018
[10] F. J. Warren, B. Zhang, G. Waltzer, M. J. Gidley, S. Dhital, *Carbohydr. Polym.* 2015, 117, 192.

[11] F. Zeng, F. Chen, F. Kong, Q. Gao, R. M. Aadil, S. J. Yu, *Food Chem.* 2015, 187, 348.

[12] J. Li, F. Ye, J. Liu, G. Zhao, *Food Hydrocolloids* 2015, 46, 226.

[13] W. Tan, Q. Li, H. Wang, Y. Liu, J. J. Zhang, F. Dong, Z. Y. Guo, *Carbohydr. Polym.* 2016, 142, 1.

[14] M. Tupa, L. Maldonado, A. Vazquez, M. L. Foresti, *Carbohydr. Polym.* 2013, 98, 349.

[15] B. Biduski, F. T. Silva, L. Halal, L. Z. Pinto, A. R. G. Dias, E. R. Zavareze, *Food Chem.* 2017, 214, 53.

[16] C. Fuentes, C. Zielke, M. Prakash, P. Kumar, J. M. Penarrieta, A. C. Eliasson, L. Nilsson, *Food Chem.* 2016, 213, 768.

[17] R. Zhang, K. J. Edgar, *Carbohydr. Polym.* 2014, 105, 161.

[18] R. Zhang, S. Liu, K. J. Edgar, *Carbohydr. Polym.* 2016, 136, 474.

[19] J. Ren, J. Liu, F. Dong, Z. Guo, *Carbohydr. Polym.* 2011, 85, 268.

[20] X. Meng, K. J. Edgar, *Prog. Polym. Sci.* 2016, 53, 52.

[21] X. Ma, X. Liu, D. P. Anderson, P. R. Chang, *Food Chem.* 2015, 181, 133.

[22] S. Yang, Z. Guo, F. Miao, Q. Xue, S. Qin, *Carbohydr. Polym.* 2010, 82, 1043.

[23] J. Ren, P. Wang, F. Dong, Y. Feng, D. J. Peng, Z. Y. Guo, *Carbohydr. Polym.* 2012, 87, 1744.

[24] L. Hu, X. Meng, R. Xing, S. Liu, X. L. Chen, Y. K. Qin, H. H. Yu, P. C. Li, *Bioorg. Med. Chem. Lett.* 2016, 26, 4548.

[25] R. Jia, Y. Duan, Q. Fang, X. Wang, J. Huang, *Food Chem.* 2016, 196, 381.

[26] Q. Li, W. Tan, C. Zhang, G. Gu, Z. Guo, *Int. J. Biol. Macromol.* 2016, 91, 623.

[27] Z. Guo, Q. Li, G. Wang, F. Dong, H. Y. Zhou, J. Zhang, *Carbohydr. Polym.* 2014, 99, 469.

[28] Y. Hu, J. Zhang, C. Yu, Q. Li, F. Dong, G. Wang, Z. Y. Guo, *Int. J. Biol. Macromol.* 2014, 70, 44.

[29] W. Tan, Q. Li, W. Li, F. Dong, Z. Guo, *Int. J. Biol. Macromol.* 2016, 82, 404.

[30] J. Ren, Q. Li, F. Dong, Y. Feng, Z. Guo, *Int. J. Biol. Macromol.* 2013, 53, 77.

[31] R. Hampe, T. Heinze, *Starch/Stärke* 2016, 68, 505.

[32] H. Namazi, S. Belali, *Starch/Stärke* 2016, 68, 177.

[33] M. J. Tizzotti, M. C. Sweedman, D. Tang, C. Schaefer, R. G. Gilbert, J. Agric. Food Chem. 2011, 59, 6913.

[34] O. V. Rúnarsson, J. Holappa, C. Malainer, H. Steinssson, M. Hjalmarsdottir, T. Nevalainen, M. Masson, *Eur. Polym. J.* 2010, 46, 1251.

[35] W. Tan, Q. Li, Z. Gao, S. Qiu, F. Dong, Z. Y. Guo, *Carbohydr. Polym.* 2017, 157, 236.

[36] R. Chen, C. Jin, Z. Tong, J. Lu, L. Tan, L. Tian, Q. Q. Chang, *Carbohydr. Polym.* 2016, 136, 187.

[37] Z. Zhong, R. Xing, S. Liu, L. Wang, S. B. Cai, P. C. Li, *Eur. J. Med. Chem.* 2008, 43, 2171.

[38] C. Marteau, R. Guitard, C. Penverne, D. Favier, V. Nardello-Rataj, J. M. Aubry, *Food Chem.* 2016, 196, 418.

[39] P. Mahakunakorn, M. Tonda, Y. Murakami, K. Matsumoto, W. Watanabe, *Biol. Pharm. Bull.* 2004, 27, 38.