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Obtaining non-digestible polysaccharides from distillers' grains of Chinese baijiu after extrusion with enhanced antioxidation capability

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ABSTRACT

Distillers' grains of Chinese Baijiu (DGS) presents a significant challenge to the environmentally-friendly production of the brewing industry. This study utilized screw extrusion to modify the morphological and crystalline characteristics of DGS, resulting in a 316 % increase in the yield of non-digestible polysaccharides extraction. Physiochemical characteristics of extracted polysaccharides were variated, including infrared spectrum, monosaccharide composition, and molecular weight. Polysaccharides extracted from extruded DGS exhibited enhanced inhibitory capacity on α-amylase activity and starch hydrolyzation, as compared to those extracted from unextruded DGS. Additionally, the ABTS, DPPH, and •OH radical scavenging efficiencies took a maximum increase of 1.20, 1.38, and 1.02-fold, correspondingly. Extrusion is a novel approach for the recycling non-digestible polysaccharides from DGS, augmenting the bioactivity of extracts and their potential application in functional food.

1. Introduction

Distillers' grains (DGS) is the solid biomass waste in Chinese Baijiu production, with over 100 million tons generated annually [1]. Grains, such as sorghum, rice husk, and wheat, are the primary raw materials for Chinese Baijiu production. DGS contains various functional components, including peptides, polyphenols, and polysaccharides, which are generated during fermentation and brewing. However, due to its high water concentration and abundant of acid and organic components, DGS can easily become putrid [2]. Therefore, efficient DGS treatment approaches are urgently needed in the brewing industry to achieve cleaner production.

Current normal treatment methods for DGS, such as feeding and landfill, result in resource loss and environmental pollutants (odor, leachate, and greenhouse effect) [3]. Large-scale DGS disposal methods, such as biogas and biochar production, and composting, prioritize high treatment efficiency [4,5]. However, the potential benefits of using DGS to produce healthy products and recycle nutrients are often disregarded. To promote the development of DGS treatment, bioactive components extraction has been identified as a crucial approach [6]. Extracted bioactive components, such as proteins and peptides, have the potential to be developed into various therapeutic agents, including antioxidants, hypolipidemic, and antihypertensive agents [7,8]. Additionally, certain bioactive components, such as phenolic and flavonoid, with physiochemical modification can be utilized as efficient composition for food preservation in packaging [9,10]. Polysaccharides extracted from hydrolyzed cellulose and hemicellulose have the potential to serve as functional food ingredients, augmenting their health-promoting properties. The consumption of polysaccharides can help to maintain a balanced daily nutrient intake and mitigate the risk of several diseases, such as immune disorders, inflammation, and cancer [11]. Especially, the polysaccharides obtained from grains have fewer side effects in

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treating hyperglycemia than synthetic anti-diabetic drugs, and exhibit stronger bioactivity than those derived from fruits and vegetables [12,13].

It should be noticed that rice husk in DGS (contains 30–40 % on dry matter basis) has high concentrations of cellulose, lignin and hemicellulose, which undermines the efficiency of biomass transformation in both large-scale treatment and bioactive component extraction processes [3,14]. Modifying cellulose and hemicellulose for polysaccharides extraction is a potentially effective approach for DGS resourcing and nutrient recycling [15]. The processes of raw materials treatment affect the bioactivities of extracted polysaccharides. Screw extrusion is a promising approach to altering the physicochemical characteristics of biomass through high shear rate and heat transfer between the extruder and raw materials. During extrusion processing, the raw materials undergo rapid heating in the extruder chambers, while the rotating screw generates shear and pressure forces between the materials and the machinery [16]. Water is pumped into chambers and mixed with materials to increase the flow rate and steam pressure [17]. Once the materials are extruded out of the chamber, water transforms into steam due to pressure and thermal variation, and bulging the materials. By optimizing extrusion key factors, such as screw speed, temperature, and moisture content, cellulose and hemicellulose can be efficiently modified into soluble components, making them more accessible for downstream applications [18]. Extrusion has been shown to stimulate the extraction of polysaccharide from high cellulose content biomass waste, such as corn, potato residues, and rice bran, whilst simultaneously augmenting the bioactivity of the extracted polysaccharides [18–20]. In contrast to chemical treatments, extrusion generates no residue of chemical reagents or environmental pollutants. However, limited research exists on the utilization of extrusion for polysaccharide extraction from DGS. Given the advantages of extrusion for biomass waste modification and the urgent need for effective DGS treatment approaches, it is essential to investigate the applicability of extrusion for DGS treatment to promote the polysaccharide extraction and the sustainable DGS utilization.

This study aims to optimize the utilization of DGS by exploring the effects of screw extrusion on the functional properties and yield of extracted polysaccharides from DGS. Structural and physicochemical analyses were conducted to elucidate the factors contributing to the improved functional properties of the polysaccharides. The study indicated that extrusion can be a novel method for recycling functional polysaccharides from DGS, which will promote the cleaner production in brewing industry.

2. Materials and methods

2.1. Materials

DGS is collected from Luzhou, China. Heat-stable α-amylase (40 U/mg), α- amylase (50 U/mg), protease (200 U/mg), cellulase (50 U/mg), pepsin (15 U/mg), 2,2-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS), 1,1-diphenyl-2-picryl-hydrozyl (DPPH), gallic acid, and bovine albumin were purchased from Yuanye Bio-Technology Co., Ltd. (Shanghai, China). NaH_2PO_4, KBr, NaAc, HCl, H_2SO_4, $FeSO_4$, H_2O_2, NaOH, $NaHCO_3$, trifluoroacetic acid (TFA), pancreatin (250 U/mg), and Na_2HPO_4 were purchased from Sinopharm Chemical Reagents Co., Ltd. (Shanghai, China). Monosaccharide standards (mannose, glucuronic acid, galacturonic acid, glucose, galactose, fucose, xylose, ribose, rhamnose and arabinose) were purchased from Tokyo Chemical Industry Co. Ltd. (Tokyo, Japan). BCA protein assay kit were purchased from Beyotime Biotechnology Co. Ltd. (Shanghai, China).

2.2. DGS extrusion and structure characteristic analysis

2.2.1. DGS extrusion with variated conditions

To modify the structure and components of DGS, a twin-screw extruder was utilized to process the DGS. The dried DGS was initially milled and passed through a 0.25 mm sieve. The powders were then placed into the extruder after adjusting the screw speed and temperature of the heating chambers to the desired conditions. Water was simultaneously pumped into the chambers, and the screw pushed the wet powders forward to the end of the screw until the outside of the chambers. Finally, the extruded samples were collected after the extruder operated in stable state. This process was repeated with different aim conditions to obtain different extruded DGS for analysis.

The initial extrusion experiment (G0) was performed under controlled conditions of 135 °C temperature, 180 rpm screw speed, and 35 % moisture content. The subsequent extrusion experiments were categorized into three groups: temperature variation group (G1 and G2, at 120 °C and 150 °C, respectively), screw speed variation group (G3 and G4, at 150 rpm and 210 rpm, respectively), and moisture content variation group (G5 and G6, at 30 % and 40 %, respectively). The unextruded sample was labeled as UE. Table 1 provides an overview of the DGS extrusion conditions.

2.2.2. Scanning electron microscopy of DGS

Morphological characterization of the extruded and unextruded DGS was conducted using a scanning electron microscope (SEM, SU8100 Hitachi, Tokyo, Japan) at 15 kV. In short, DGS powders were dispersed to from uniform thin layers and fixed on an aluminum mounting stub. Gold was sputtered on the layers to enhance the electrical conductivity of samples. Subsequently, samples were observed by SEM at 300 and 1000 magnifications [7].

2.2.3. X-ray diffraction analysis of DGS

The X-ray diffraction patterns of DGS were obtained using a diffractometer (D2 PHASER; BRUKER AXS GMBH, Karlsruhe, Germany) with CuKα-radiation (Ka1 = 0.1542 nm, 30 kV, 10 mA) in the 2θ range of 5–70°, with an increment of 0.05° and data collection of 0.5 s per step [7].

2.3. Polysaccharides extraction and physicochemical analysis

2.3.1. Polysaccharides extraction from DGS

The polysaccharide was extracted using enzymatic method with some modifications [21]. Briefly, 10 g of extruded and dried DGS was dispersed in phosphate buffered solution (120 mL, 0.1 mol/L, and pH = 8.0) and subjected to successive incubation with 0.1 g heat-stable α-amylase (at 90 °C, 1 h) followed by 0.1 g protease (at 45 °C, 1 h). Subsequently, the pH was adjusted to 5.5, and 0.05 g cellulase was added for cellulose hydrolysis at 45 °C for 1 h. The enzymatic hydrolysate was then centrifuged at 3200 ×*g* and 4 °C for 15 min. The supernatant was boiled for 5 min to inactivate the enzyme. Subsequently, the hydrolysate underwent a 48-h desalination process using a dialysis bag with a 500 Da cutoff, which was maintained at 4 °C. To precipitate the polysaccharide, four-fold volume of 95 % ethanol was added into the desalinated solution at 4 °C and left for 12 h. The mixture was then centrifuged at 3200 ×*g* for 20 min and the precipitation (polysaccharide) was stored in a desiccator after freeze-drying. Unextruded DGS (UE) was used as the control group. The yield of polysaccharide was calculated with Eq. (1) [21].

$$\text{Yield}\ (\%) = m_s/m_0 \times 100 \quad (1)$$

where m_0 is the dry weight of DGS with or without extrusion, and m_s is the weight of lyophilized polysaccharide.

2.3.2. Chemical component analysis of DGS and polysaccharides

All of the extracted polysaccharides were dissolved into deionized water at same concentration. According to the Xiong, et al. [22], the total sugar contents of polysaccharides were analyzed using the phenol-sulfuric acid colorimetry method at 490 nm, with glucose as the standard. The content of uronic acid was determined using the *M*-

Table 1
Extrusion conditions and chemical component analysis of polysaccharide extracted from Chinese Baijiu distillers' grains.

Samples	Extrusion temperature (°C)	Extrusion speed (rpm)	Moisture content (%)	Polysaccharide yield (%)	Total sugar content (%)	Uronic acid content (%)	Total phenol contents (%)	Protein (%)
UE	0	0	0	1.64 ± 0.02^{f}	13.86 ± 0.05^{c}	5.66 ± 0.01^{c}	0.16 ± 0.001 ^{b}	ND
G0	135	180	35	5.81 ± 0.01^{a}	14.24 ± 0.05^{c}	5.61 ± 0.01^{c}	0.13 ± 0.001 ^{c}	ND
G1	120	180	35	3.21 ± 0.01^{e}	11.55 ± 0.03^{e}	2.28 ± 0.02^{e}	0.09 ± 0.001 ^{b}	ND
G2	150	180	35	5.66 ± 0.04^{b}	11.94 ± 0.07^{d}	2.17 ± 0.03^{e}	0.03 ± 0.001 ^{e}	ND
G3	135	150	35	1.70 ± 0.07^{f}	19.59 ± 0.02^{b}	2.97 ± 0.01^{d}	0.11 ± 0.001 ^{d}	ND
G4	135	210	35	5.15 ± 0.01^{c}	17.68 ± 0.01^{c}	2.21 ± 0.01^{d}	0.09 ± 0.009 ^{b}	ND
G5	135	180	30	4.15 ± 0.19^{d}	25.64 ± 0.03^{a}	12.55 ± 0.09^{a}	0.19 ± 0.001 ^{a}	ND
G6	135	180	40	1.80 ± 0.01^{f}	21.86 ± 0.04^{b}	10.30 ± 0.03^{b}	0.13 ± 0.008 ^{c}	ND

ND: None detected; G0 to G6: polysaccharide samples from extruded Chinese Baijiu distillers' grains (in different extrusion conditions); UE: polysaccharide sample from unextruded Chinese Baijiu distillers' grains. Values in the same column with different letters indicate significant differences, $p < 0.05$.

hydroxybiphenyl method, with galacturonic acid being utilized to establish a standard curve at 560 nm. The BCA method was employed to analyze protein at 520 nm, with bovine albumin serving as the standard. The total phenol concentration was determined at 760 nm *via* the Folin-Ciocalteu method, with gallic acid as the standard. The chemical component results of all samples are shown in Table 1.

2.3.3. UV–visible absorption and FT-IR spectra measurements of polysaccharides

The polysaccharide were dissolved in deionized water at a concentration of 1 mg/mL and the ultraviolet (UV)-visible absorptions were detected using a UV spectrophotometer (Spectra Max 190, Molecular Devices, California, USA) in the range of 200–850 nm [22].

The 2 mg polysaccharides and 200 mg KBr were mixed then tableted. The mixed samples were recorded on a FT-IR spectrometer (Spectrun100; Thermo Fisher Scientific, Massachusetts, USA) in the frequency range from 400 cm^{-1} to 4000 cm^{-1} using 32 scans with a resolution of 4 cm^{-1} [10].

2.3.4. Molecular weight distribution determination of polysaccharides

The molecular weight distributions of polysaccharides were detected using a high-performance gel permeation chromatography method. The method involved utilizing a Waters2695 system (Waters, Massachusetts, USA) with a refractive index detector (Waters 2414, Waters, Massachusetts, USA), which was equipped with Ultrahydrogel™ column (7.8 × 300 mm, Waters, Massachusetts, USA). A mobile phase consisting of 0.1 mol/L $NaNO_3$ with a flow rate of 0.8 mL/min was employed. A series of dextran standards (2.7 kDa, 9.7 kDa,135 kDa, 300 kDa, and 2000 kDa) and glucose were utilized to construct a standard curve to calculate the molecular weight [23].

2.3.5. Monosaccharide composition of polysaccharides

Referring to the method reported by Xiong, et al. [22] with some modifications, 5 mg polysaccharide samples were hydrolyzed with 1.0 mL 4.0 mol/L TFA at 120 °C for 3 h. The hydrolyzed samples were analyzed by ICS-5000 + SP-5 (Thermo Fisher Scientific, Massachusetts, USA) equipped with pulsed amperometric detector (PAD). A CarboPac™ PA20 guard column (3 mm × 50 mm) and a CarboPac™ PA20 analytical column (3 mm × 150 mm) were connected in series for the analysis. Sample loading volume was 20 μL and column temperature was 30 °C. The mobile phase was deionized water (solvent A), 250 mmol/L NaAc (solvent B) and 250 mmol/L NaOH (solvent C). The elution procedure was as 99.1 % A and 0.9 % C in 20 min, 19.1 % A, 80 % B and 0.9 % C in 20.1–27 min, and 20 % A, 0 % B and 80 % C in 27.1–38 min. The flow rate was 0.5 mL/min.

2.4. Polysaccharides functional properties assays

2.4.1. Estimation of inhibitory effect on α-amylase activity

The inhibitory effect on α-amylase activity of polysaccharides was estimated according to the method reported by Tang, et al. [24] with slight modifications. 50 mg polysaccharide mixed with 5 mL of α-amylase (20 μg/mL), and incubated at 37 °C for 15 min. The mixture was boiled for 5 min for enzyme inactivation followed by centrifugation (3200 ×*g*, 10 min). Acarbose was used as the positive control. The glucose concentration in the supernatant was measured using the DNS method. The inhibition rate was calculated using the Eq. (2) [24]:

$$\text{Inhibition rate } (\%) = (A_1 - A_2)/A_1 \times 100 \quad (2)$$

where, A_1 and A_2 are the absorbances of supernatant with and without polysaccharide at 490 nm, respectively.

2.4.2. Determination of degree of starch digestion

The method for starch *in vitro* digestion was adapted from Zhang, et al. [25] and Wang, et al. [26] with some modifications. The mixtures of soluble starch (SS, 100 mg) with polysaccharides (25 mg) were dissolved in distilled water (10 mL). The suspensions were heated at 95 °C for 20 min with constant stirring for gelatinization. The cooled suspensions were infused with 5 mL of simulated saliva fluid, containing α-amylase (0.2 mg/mL). The mixture was then agitated at 60 rpm, 37 °C for 5 min. After this, 1 mol/L HCl was used to bring the pH of mixture down to 1.8. Subsequently, 25 mL of simulated gastric fluid containing pepsin (0.3 mg/mL) was added to the digested mixture. The mixture was further stirred at 175 rpm and maintained at 37 °C for 30 min. Finally, the digested juices were combined with 40 mL of simulated intestinal fluid containing pancreatin (0.6 mg/mL), while the pH of mixture was adjusted to 6.8 using 1 mol/L $NaHCO_3$. The sample were incubated at 37 °C with stirring at 175 rpm. Consecutively, digestive juices (0.5 mL) were withdrawn at 0, 10, 20, 30, 45, 60, 90, 120, 150 and 180 min of simulated intestinal digestion, respectively. Ethanol (2 mL) was added into the samples to inactivate enzyme. After centrifugation (8000 ×*g*, 20 min, and 4 °C), The content of reducing sugar in the supernatant was determined using DNS method. The degree of starch digestion (HI), rapid digestion starch (RDS), slowly digestion starch (SDS), and resistant starch (RS) were calculated using the Eqs. (3), (4), (5) and (6), respectively [25]:

$$\text{HI } (\%) = (R_t - R_0) \times 0.9/TS \times 100 \quad (3)$$

$$\text{RDS } (\%) = (R_{20} - R_0) \times 0.9/TS \times 100 \quad (4)$$

$$\text{SDS } (\%) = (R_{180} - R_{20}) \times 0.9/TS \times 100 \quad (5)$$

$$\text{RS } (\%) = (TS - RDS - SDS)/TS \times 100 \quad (6)$$

where R_t is the reducing sugar content at the time of t min during the intestinal digestion. R_0 is the reducing sugar at the time of 0 min during the intestinal digestion. TS is the total dry weight of starch in samples.

2.4.3. ABTS radical scavenging activity determination

The ABTS radical scavenging activities of the obtained polysaccharides were analyzed based on the method of Li, et al. [27] with some modifications. The ABTS radical stock solution was prepared by

combining 20 mL of 7 mmol/L ABTS with 352 μL of 140 mmol/L potassium peroxodisulfate. The stock solution was then diluted using 95 % ethanol until an absorbance of 0.70 ± 0.02 was achieved at 734 nm before the test. The 180 μL of diluted ABTS mixed with 20 μL of polysaccharides water solutions (at different concentration 0.2, 0.4, 0.6, 0.8, 1, and 2 mg/mL) in 96-well plates in the dark for 5 min reaction. Then, the absorbance was tested at 734 nm. In this study, A-Tocopherol (Ve) was used as the positive control in all antioxidation capability assays. The ABTS radical scavenging activity of the polysaccharides was calculated according to Eq. (7) [27]:

$$\text{Scavenging percentage}\ (\%) = (A_2 - A_1)/A_2 \times 100 \quad (7)$$

where A_1 is the absorbance of the polysaccharide solutions mixed with ABTS, and A_2 is the absorbance of the water mixed with ABTS solution.

2.4.4. DPPH radical scavenging activity determination

The scavenging activities of samples on DPPH were determined by the reported method with some modifications [22]. Each of 100 μL of polysaccharides water solution (at different concentration 0.2, 0.4, 0.6, 0.8, 1, and 2 mg/mL) were mixed with 100 μL of DPPH ethanol solution (0.25 mmol/L) in 96-well plates. Then the absorbance was measured at 517 nm after 30 min of reaction at 37 °C in darkness. DPPH radical scavenging activity was calculated using the Eq. (8) [22]:

$$\text{Scavenging percentage}\ (\%) = [A_0 - (A_1 + A_2)]/A_0 \times 100 \quad (8)$$

where A_0 is the absorbance of the water and DPPH ethanol solution, A_1 is the absorbance in the presence of the polysaccharides water solution and DPPH ethanol solution, and A_2 is the absorbance of the polysaccharides water solution without DPPH ethanol solution.

2.4.5. Hydroxyl radical scavenging activity determination

Hydroxyl radicals (•OH) scavenging activity was determined by previously reported method with some modifications [22]. The 1 mL of 3 mmol/L $FeSO_4$, 3 mmol/L sodium salicylate, 6 mmol/L H_2O_2, and varying concentrations of polysaccharides water solutions (0.2, 0.4, 0.6, 0.8, 1, and 2 mg/mL), were incubated at 37 °C for 30 min in darkness. The absorbance was detected at 510 nm. •OH radical scavenging activity was calculated using the Eq. (9) [22]:

$$\text{Scavenging percentage}\ (\%) = [A_0 - (A_1 + A_2)]/A_0 \times 100 \quad (9)$$

where A_0 is the absorbance of the water instead of test polysaccharide solutions, A_1 is the absorbance of the test polysaccharide solutions, and A_2 is the absorbance of the polysaccharides solutions mixed with water instead of H_2O_2 in the hydroxyl radicals generating system.

2.5. Statistical analysis

All the data were calculated with three replications and shown in mean ± standard deviation. Statistical analysis was conducted using SPSS 20.0 software (SPSS Incorporated, Illinois, USA). The data were analyzed by means of the analysis of variance (ANOVA), and Student Newman-Keuls test was used for significant differences evaluation (the differences were considered as significant at $p < 0.05$). The analyses were performed using Origin software, version 8.0 (OriginLab Corporation, Massachusetts, USA).

3. Results and discussion

3.1. Structural modification in extruded DGS

3.1.1. Scanning electron microscopy of DGS

SEM images of distiller's grains were analyzed and presented in Fig. 1. The SEM images revealed that the surface of DGS-UE (Fig. 1A and a) was smooth and rigid with clearly visible veins (rice husk). However,

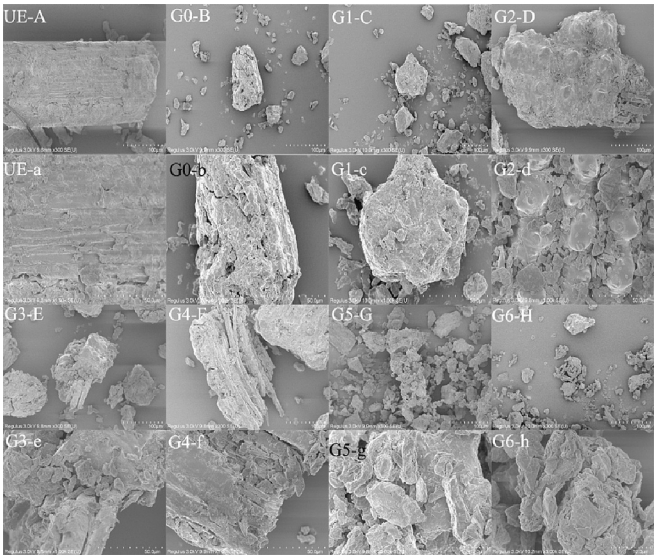

Fig. 1. Scanning electron microscopy (SEM) of extruded and unextruded Chinese Baijiu distillers' grains from SPRs × 300 magnification (capital letters) and ×1000 magnification (lower case).

remarkable differences were observed on the surface of the extruded DGS samples. Fissures and holes were shown on the surface of DGS-G0 (Fig. 1B and b), which could enhance the efficiency of further hydrolyzation by exposing the components to the solution [7]. Higher temperature resulted in the formation of distinctive bulges on the surface of DGS-G2 (Fig. 1C), which could be attributed to the swelling of starch and cellulose under high temperature. Comparable materials surface transformations were noted in another study about DGS extrusion [28]. Higher extrusion speed generated stronger force on the materials, leading to more pronounced shear marks in DGS-G4 (Fig. 1f) compared to DGS-G3 (Fig. 1e). Fault lines were also discerned in the granules of DGS-G4, while the intact branches were retained in DGS-G3, indicating that higher extrusion speed caused a more severe destruction of the DGS structure. Higher water concentration reduced the shear force on granules, resulting in a relatively weak compactness in the higher moisture content sample (DGS-G6) compared to the lower moisture content sample (DGS-G5).

3.1.2. X-ray diffraction analysis of DGS

The crystallinity of the samples was investigated using an X-ray (Fig. 2). Results revealed the presence of a crystalline structure of cellulose in both extruded and unextruded DGS, as evidenced by peaks around $2\theta = 22°$, along with a small peak at $2\theta = 34°$, and the intensity of peaks was increased in extruded samples [29]. However, compared to the unextruded sample, the extruded DGS showed a roader peak at 17° and a sharper peak at 18°, suggesting a loss of B-type starch crystallinity and an increase in V-type starch crystallinity [30]. The decreased intensity of peak at 15° could indicate the hydrolyzation of starch [31]. The intensity of cellulose peak increased in extruded samples, especially in DGS-G5, which could be related to the lignin and hemicellulose elimination [32]. The increased cellulose crystallinity improves the glucose adsorption capacity of polysaccharide, which could promote their potential application in food digestion and fermentation [31].

3.2. Yield and physicochemical analysis of polysaccharides

3.2.1. The effect of extrusion conditions variation on polysaccharide yield

The yield of polysaccharide took significant variation in different extrusion conditions. DGS with extruder treatment (G0-G6) processed higher polysaccharide yield (from 1.70 ± 0.07 % to 5.81 ± 0.04 %) than unextruded DGS (UE 1.64 ± 0.02 %, Table 1). DGS-G0 took the highest

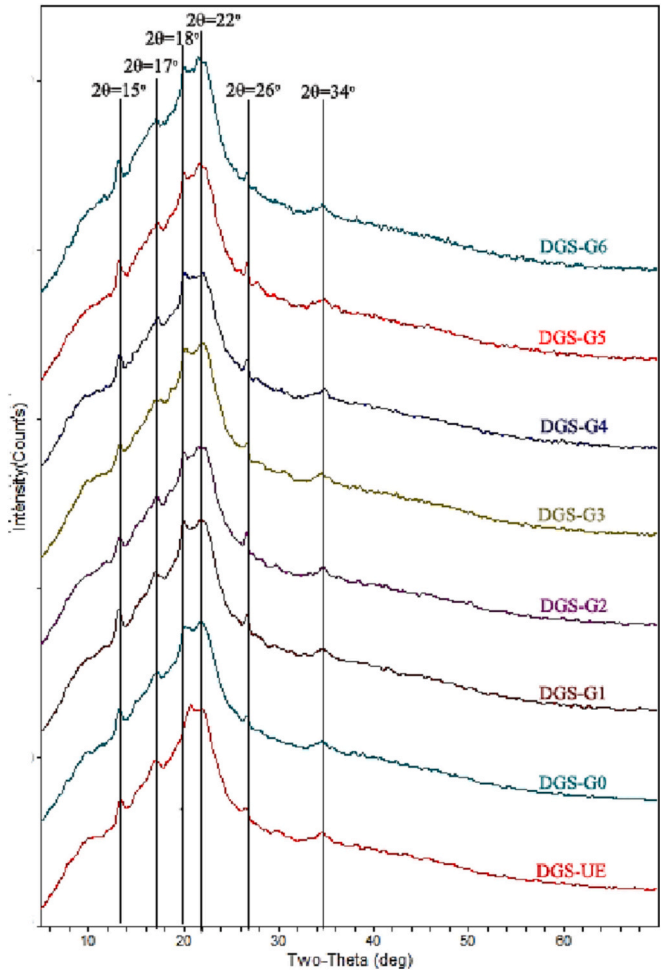

Fig. 2. X-ray diffractograms of Chinese Baijiu distillers' grains. DGS: Chinese Baijiu distillers' grains.

yield (5.81 ± 0.01 %) which led to 3.16-folds increase in the yield of UE.

Extrusion screw speed and temperature affected polysaccharides yield. Higher screw speed generated stronger share force (made deeper shear marks on DGS), which promoted the release of components. However, lower speed was not as efficient for polysaccharide extraction. A similar result was observed in rice husk extrusion, where the polysaccharide yield decreased significantly with decreased screw speed and increased temperature [33].

In the material moisture group, the yield of polysaccharide in G5 was higher than that in G6. The stimulated yield could associated with the promoted cell-wall structure modification in lower moisture conditions [18]. However, higher moisture content resulted in a smoother material surface and dissipated the mechanical energy and extrusion efficiency [34].

3.2.2. *UV–visible absorption and FT-IR spectra of polysaccharides*

In the result of UV–visible analysis, only an absorption peak around 260 nm was observed in the UV–visible spectra of all polysaccharides (Fig. 3A), indicating the existence of nucleic acid (Rib was detected in later monosaccharide composition analysis) rather than protein (absorption peak around 280 nm) [22].

FTIR spectra for polysaccharides was shown in Fig. 3B. Characteristic absorption peaks of all polysaccharides were observed at 3390–3421 cm^{-1}, and were associated with stretching vibration of O—H of hydroxyl groups and C—H in methyl and methylene groups, respectively [20]. The peak at 2930 cm^{-1} was covered by the broad peak of C—H bond confirmed the existence of cellulose and hemicellulose in all samples. The peak at around 1663 cm^{-1} was probably associated with C═O or COO— bond of the uronic acids (carboxylic acid functionality), which was also shown in all samples [18]. It is important that extrusion processed the spectrum intensity variation and redshift. Firstly, polysaccharide sample G5 and G0 took stronger peak intensity of the hemicellulose spectrum peak (close to 1000 cm^{-1}) than UE, which could indicate the decomposition of insoluble components to polysaccharide [35]. Secondly, the intensity of peak around 1663 cm^{-1} was increased in extruded samples, especially in G5, which could attribute to higher uronic acids content [18]. Furthermore, the spectral band at 580 cm^{-1} represents the C—H bond in β- pyranose (soluble hemicellulose) was increase in samples from extruded DGS [36]. This variation was remarkable in samples from lower moisture content condition (G5), and higher speed (G4) treatments, which indicated that improved yield could associated with stronger share force on materials. On the other hand, characteristic peak of cellulose (1070–1170 cm^{-1}) separated to two peaks in some extruded samples (such as G0) and the intensity of peak with remarkable modification (such as G5 and G6). It indicating that extrusion would degrade the cellulose or modify the structure characteristic of cellulose [15]. These results indicated that extrusion could promote the decomposition of cellulose and hemicellulose. With the decomposition and modification, the yield and functional characteristic of polysaccharides from extruded DGS was increased and enhanced, respectively.

3.2.3. *Molecular weight determination of polysaccharides*

The average molecular weight distribution of the polysaccharide from extruded DGS was higher than that of UE (Table 2). This could be attributed to the efficiency of DGS component hydrolyzation after structure modification by extrusion. Extrusion with higher screw speed (G4) and lower moisture content (G5) modified the DGS structure remarkable, fragmented the molecular chains of polysaccharide. The extrusion process induced shear force and heat that caused the rupture of the DGS structure. This promoted the modifications of the cellulose and hemicellulose chains in further enzymatic hydrolysis, leading to the transformation of components and an increase in the dissolution ratio of macromolecular polysaccharides [27].

3.2.4. *Monosaccharide composition of polysaccharides*

Monosaccharide composition of polysaccharides is shown in Table 2. According to the standard, neutral (Fuc, Rha, Ara, Gal, Glc, Xyl, Man and Rib) and acidic (Gal-UA and Glc-UA) sugars were found in all obtained polysaccharides with different molar ratios, which further confirms that all extracted polysaccharides were acidic heterogeneous polymers. The predominant monosaccharide in the polysaccharide of differently treated DGSs were Glc, Man, Gal, and Ara, with a higher molar ratios of Ara and Gal in G5 and G6. Moisture content variation during extrusion affected the ratio of Gal-UA in G5 and G6, which could be attributed to increased pectin explosion from the cell wall after extrusion [35]. The monosaccharide composition variation affects the biological activity of polysaccharides. Previous studies have demonstrated that polysaccharides with higher contents of Gal-UA exhibit stronger bioactivities [22], while polysaccharides rich in Gal and Ara have the potential for strong antioxidant activity [37]. These findings suggest that the biological activities of G5 and G6 would be more potent than other polysaccharides. Furthermore, the compositions of polysaccharides form DGS different to those from pure rice husk. Rivas, et al. [38] reported that polysaccharides from rice husk mainly consisted of Glu, Xyl and Ara without Gal or Man. The similar neutral monosaccharides between rice husk polysaccharides and DGS polysaccharides could hydrolyze from hemicellulose, but the distinction could associate with the modification in the process of solid-state fermentation and the extrusion [39].

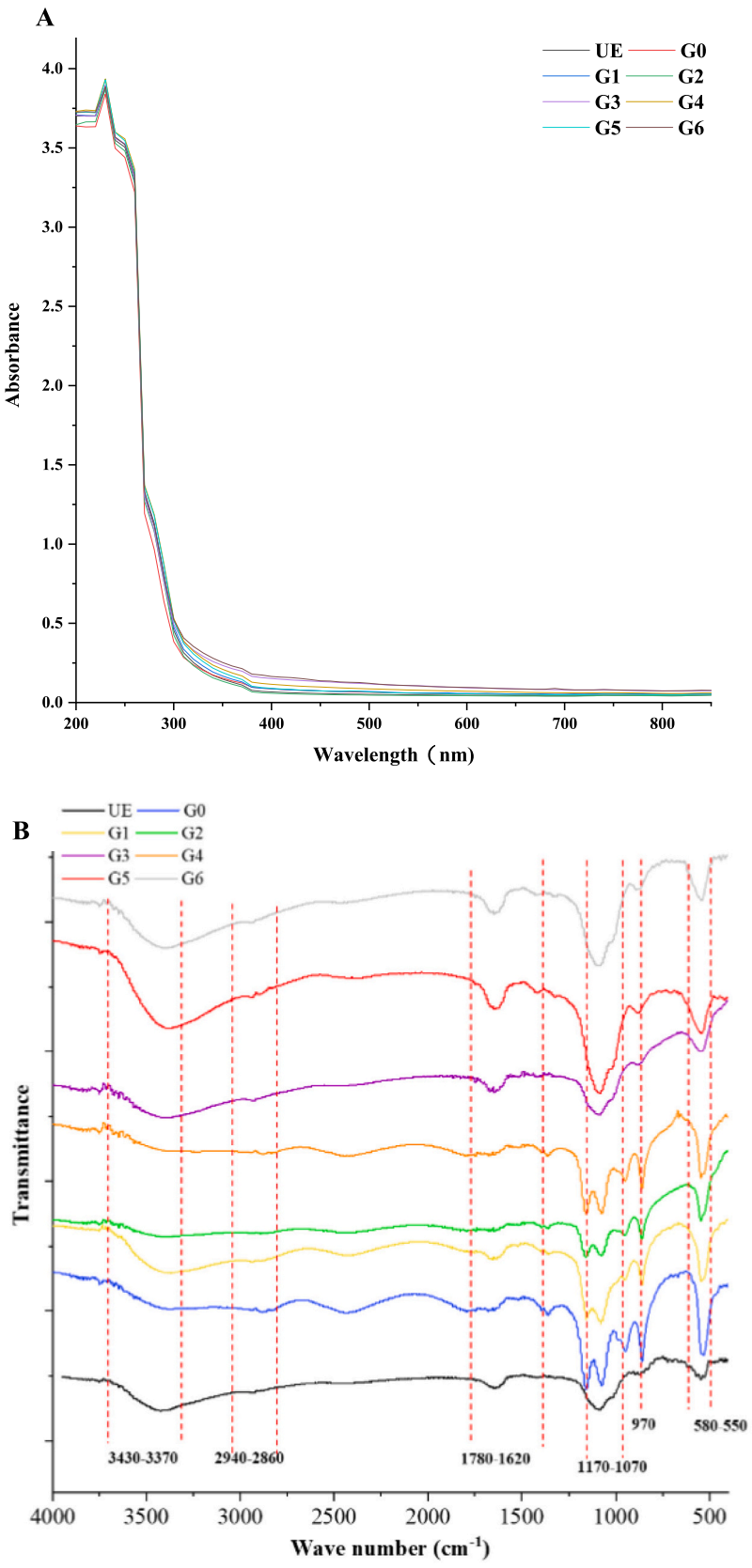

Fig. 3. UV–visible absorption (A) and FT-IR spectra measurements (B) of polysaccharide samples from Chinese Baijiu distillers' grains.

Table 2
Monosaccharide composition and molecular weight of polysaccharide from extruded and unextruded Chinese Baijiu distillers' grains.

	Molecular weight (kDa)	Fuc (%)	Rha (%)	Ara (%)	Gal (%)	Glc (%)	Xyl (%)	Man (%)	Rib (%)	Gal-UA (%)	Glc-UA (%)
UE	1.48	12.39	6.30	8.52	8.37	35.98	5.05	16.39	0.68	4.36	1.96
G0	19.35	3.70	4.54	5.34	9.43	57.14	5.64	7.10	ND	4.86	2.25
G1	7.60	5.01	1.78	5.80	13.90	49.75	3.88	9.47	ND	4.39	6.02
G2	10.21	5.65	6.25	6.03	8.79	54.41	ND	9.94	ND	3.24	5.69
G3	5.81	7.79	4.50	8.32	10.47	43.58	5.74	12.11	ND	3.50	3.99
G4	24.06	6.66	8.20	7.80	10.01	40.73	1.36	16.82	0.54	4.27	3.61
G5	25.70	7.20	7.18	10.66	8.21	36.96	6.68	11.90	0.76	6.40	4.05
G6	21.35	7.45	6.95	9.98	9.34	34.57	6.25	13.09	1.32	7.16	3.89

Fuc: fucose, Rha: rhamnose; Ara: arabinose; Gal: galactose; Glc: glucose; Xyl: xylose; Man: mannose; Rib: ribose; Gal-UA: galacturonic acid; Glc-UA glucuronic acid.

3.3. Functional properties assay of polysaccharides

3.3.1. The α-amylase inhibitory activity of polysaccharides

Inhibiting activities of starch digestive enzymes, such as α-amylase, is an effective approach to delaying the rise of blood glucose level [40]. The obtained results (Fig. 4) demonstrate that, with the exception of UE, all polysaccharides were found to be resistant to hydrolysis by α-amylase, implying the ability of polysaccharides from DGS to maintain stability in an α-amylase mixed solution. Meanwhile, the degree of hydrolysis of G0-G6 by α-amylase was lower than that of UE, indicating that the proportion of α-starch in extruded samples was decreased, and non-starch polysaccharides and other structure starch were released by extrusion [41]. Considering with the intensity variation of starch crystallinity in DGS (Fig. 2) and spectrum peak in polysaccharide (Fig. 3B), enhanced α-amylase inhibitory activity of extruded polysaccharide indicates that extrusion significantly modified the composition of polysaccharide, and boosted its functional bioactivity. However, further study is needed to gain more information in this regard.

As shown in Table 3, the α-amylase inhibition abilities of all polysaccharides were concentration-dependent in tested range from 0.2 to 5.0 mg/mL. Although G5 exhibited a relatively stronger α-amylase inhibition ability compared to other samples, its inhibitory potential was still much weaker than that of acarbose. It was not until the concentration was higher than 5 mg/mL, the inhibition abilities of polysaccharide were over 10 %. The inhibition of enzyme activity can be attributed to the encapsulation of the enzyme within polysaccharides, at a sufficient concentration, thereby restricting the interaction between the enzyme and starch [42]. Thus, inefficient inhibition at low polysaccharide concentrations (<5 mg/mL) could be due to the lower sugar content (with insufficient components) of the polysaccharide. Samples with larger molecular weight could take advance to form the barriers, as seen the difference between G5 *versus* G6 and G4. Similar results were demonstrated in the study conducted by Amamou, et al. [43], where variations in α-amylase inhibitory capabilities were detected among samples with varying molecular weights. Additionally, polysaccharide can increases the viscosity of the reaction system and effectively bind glucose, thereby limiting the diffusion of glucose in enzymatic hydrolysis [44].

3.3.2. Effect of polysaccharides on starch digestion in vitro

The reductions in reducing sugar release were achieved by incorporating different polysaccharides into SS solutions during the digestion process. The starch hydrolysis rate of pure SS increased rapidly in the first 60 min of intestinal digestion, which rose from 12.02 ± 0.02 % to 50.20 ± 0.61 % (Fig. 4B). However, the hydrolysis rate of SS mixed with polysaccharides rose slower than that of pure SS in the first 60 min, especially the hydrolysis rate of SS mixed with G5 only rose to 39.99 ± 0.56 %. G0 and UE also inhibited the SS hydrolysis which rate was 43.66 ± 0.97 % and 42.96 ± 0.60 % at 60 min, respectively. Compared to pure SS, the hydrolysis rates were decreased by 6.91 % (G0) to 12.18 % (G5) at the initial 20 min, and by 1.25 % (G1) to 8.07 % (G5) at 120 min of digestion, respectively. It indicated that the presence of polysaccharides in the digestant leads to a decrease in SS hydrolysis rates, and an enhanced inhibition capacity was observed in G5 and G6. Furthermore, the fragments of RDS, SDS, and RS in digestive juice were calculated in Table 4. The results demonstrated that the percentage of RDS fragments declined followed by the rose of SDS and RS fragments in SS and polysaccharide hybrid system, which then attenuated the digestion process of SS. According to reported studies, polysaccharides took good performance in postprandial glycaemia and starch viscosity reduction [45]. Carbohydrate interactions could also modify the digestion of starch by limit the availability of water for enzyme substrate reactions [46,47]. Polysaccharides derived from extruded DGS exhibited efficient inhibition of the starch-enzyme reaction.

3.3.3. ABTS radical scavenging activity of polysaccharides

ABTS radical scavenging activity was used as comprehensive indicator to evaluate the antioxidant capacity of samples. The ABTS radical scavenging activity of polysaccharides G0-G6 and UE strengthened with the concentration (Fig. 5A). Significant differences ($p < 0.05$) between all groups were only observed in the higher concentrations range (0.8–2 mg/mL). UE exhibited a slightly higher scavenging percentage (approaching 20 %) in the higher concentration range, compared to its scavenging percentage in the lower concentration range (0.2–0.6 mg/mL). G0 showed a similar trend to UE, but the difference in percentage between the lower and higher concentration range was not significant. In all tested concentrations, samples with higher moisture content (G5 and G6) performed stronger capacity than other samples. Among all tested samples, G5 exhibited the highest percentage of ABTS radical scavenging activity at all concentrations, ranging from 18.06 ± 0.21 % to 25.80 ± 0.79 %. However, other extruded samples did not take better performances than the unextruded sample (UE). Samples from the screw speed variation group (G3 and G4) took weaker ABTS radical scavenging activity than G0 and UE. Meanwhile, samples from temperature variation group (G1 and G2) took the lowest scavenging percentage in the lower concentration range, especially at 0.2 mg/mL. However, some studies have shown that the polysaccharides with lower molecular weight took stronger ABTS radical scavenging activity than that of high molecular weight polysaccharides [22,48,49]. The results suggest that the ABTS radical scavenging activities of the DGS polysaccharides were primarily influenced by the total sugar and acid sugar content (Table 1), with samples having higher total sugar content exhibiting better activity. This could be the main factor for G5 achieving the strongest ABTS radical scavenging activity among all samples.

3.3.4. The DPPH radical scavenging capacity of polysaccharides

DPPH radical scavenging capacity assays is a convenient *in vitro* approach for evaluating the antioxidative activity of samples. It could be seen from Fig. 5B that the scavenging capacity of polysaccharides to DPPH was concentration dependent. DPPH radical scavenging activity shown significant differences ($p < 0.05$) between all groups at all tested concentrations. UE took a relative higher scavenging percentage (22.17 ± 0.42 %) at 2 mg/mL, but this rate gradually dropped to a lower scavenging percentage (5.46 ± 0.22 %) at 0.2 mg/mL. G0 took a higher scavenging percentage than UE in all tested concentration range. At 0.2 mg/mL, the scavenging percentage of G0 was 9.93 ± 0.31 %, which was

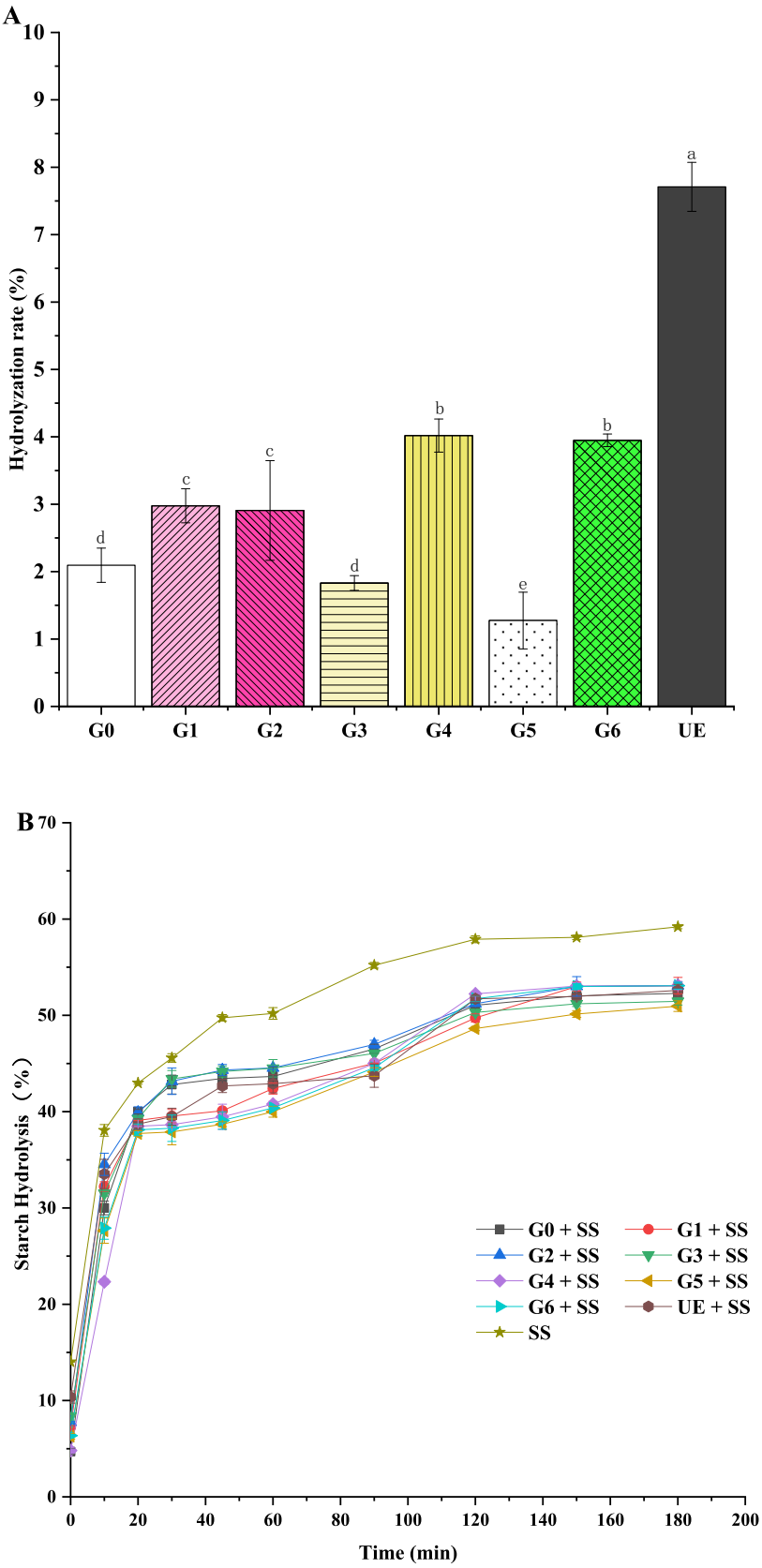

Fig. 4. Hydrolyzation rate of polysaccharides from Chinese Baijiu distillers' grains (at concentration of 5 mg/mL) (A); Starch hydrolysis in a simulated digestion process during 180 min *in vitro* analysis of different polysaccharides from Chinese Baijiu distillers' grains mixed solution (B); SS: Soluble starch. Different letters indicated significant differences of the different samples, $p < 0.05$.

Table 3
Inhibitory rate of polysaccharides from Chinese Baijiu distillers' grains on α-amylase.

Samples	G0 (%)	G1 (%)	G2 (%)	G3 (%)	G4 (%)	G5 (%)	G6 (%)	UE (%)	Acrbose (%)
0.2 mg/mL	1.95 ± 0.02^{d}	2.49 ± 0.65^{b}	2.03 ± 0.20^{b}	2.09 ± 0.16^{c}	2.32 ± 0.19^{c}	2.07 ± 0.46^{c}	2.6 ± 0.17^{b}	2.52 ± 0.02^{b}	13.12 ± 1.55^{a}
0.8 mg/mL	2.68 ± 0.89^{e}	4.39 ± 0.75^{c}	3.01 ± 0.45^{d}	1.76 ± 0.36^{f}	3.27 ± 0.36^{d}	6.07 ± 0.99^{b}	4.87 ± 0.68^{c}	2.42 ± 0.03^{e}	30.89 ± 0.39^{a}
1 mg/mL	4.48 ± 1.25^{c}	5.08 ± 0.44^{c}	3.97 ± 0.51^{d}	2.71 ± 0.53^{e}	3.6 ± 0.53^{d}	4.97 ± 2.56^{c}	6.29 ± 0.66^{b}	2.91 ± 1.08^{e}	35.72 ± 0.09^{a}
2 mg/mL	7.18 ± 0.48^{c}	7.24 ± 0.43^{c}	5.93 ± 0.24^{d}	4.24 ± 1.45^{f}	5.03 ± 1.19^{e}	5.89 ± 0.30^{d}	9.34 ± 0.34^{b}	4.21 ± 0.20^{f}	40.2 ± 0.09^{a}
5 mg/mL	14.49 ± 0.28^{c}	14.36 ± 0.43^{c}	8.48 ± 0.52^{f}	10.05 ± 0.50^{e}	11.79 ± 0.78^{d}	18.67 ± 2.07^{b}	11.86 ± 0.63^{d}	8.43 ± 0.81^{f}	89.93 ± 0.28^{a}

Values in the same row with different letters indicate significant differences, $p < 0.05$.

Table 4
Parametric analysis of polysaccharides from Chinese Baijiu distillers' grains on soluble starch properties based on simulated digestion process.

	G0	G1	G2	G3	G4	G5	G6	UE	Soluble starch
HI-180 (%)	52.29 ± 0.44	50.96 ± 0.86	53.09 ± 0.36	51.45 ± 0.37	53.10 ± 0.43	53.08 ± 0.55	53.08 ± 0.36	52.60 ± 0.62	59.22 ± 0.21
HI-20 (%)	40.00 ± 0.23	39.09 ± 0.02	39.91 ± 0.61	39.28 ± 0.06	38.49 ± 0.05	37.74 ± 0.28	38.11 ± 0.60	38.71 ± 0.55	42.97 ± 0.13
HI-60 (%)	43.66 ± 0.97	40.41 ± 0.58	44.54 ± 0.14	44.50 ± 0.90	40.80 ± 0.28	39.99 ± 0.56	40.39 ± 0.08	42.91 ± 0.95	50.20 ± 0.61
RDS (%)	31.77 ± 0.03	28.69 ± 0.19	28.93 ± 0.13	27.79 ± 0.16	30.31 ± 0.05	28.30 ± 0.54	28.58 ± 0.23	20.10 ± 0.27	35.05 ± 0.13
SDS (%)	11.06 ± 0.37	10.68 ± 0.87	11.86 ± 0.33	10.95 ± 0.41	13.14 ± 0.33	13.80 ± 0.91	13.47 ± 1.16	12.50 ± 0.16	14.60 ± 0.33
RS (%)	57.18 ± 0.18	60.65 ± 1.05	59.21 ± 1.19	61.26 ± 0.37	56.55 ± 0.82	57.90 ± 0.44	57.95 ± 1.19	67.40 ± 0.16	50.34 ± 0.20

HI-180/60/20: Degree of starch hydrolysis at 180, 60 and 20 min at the time of the intestinal digestion, respectively; RDS: Percentage of rapid digestion starch; SDS: Percentage of slowly digestion starch; RS: Percentage of resistant starch.

almost twice that of UE. Interestingly, there was no significant difference in the total sugar content of these 2 polysaccharides, but their molecular weight took significant variation. It could indicate that higher molecular weight components enhance the activity of polysaccharides on DPPH radical scavenging. G5 and G6 showed higher and more stable scavenging activities than other polysaccharides. G5 kept good performance both at the highest (30.68 ± 0.51 % at 2 mg/mL) and the lowest tested concentration (14.54 ± 0.23 % at 0.2 mg/mL). A similar characteristic was observed in G6, which could associate with the similar molecular weight and monosaccharide composition between G5 and G6 (Table 2) [50]. It should be noticed that even though G6 had higher ratio of Gal-UA and Gal in monosaccharide composition than G5, its capacity of DPPH radical scavenging was still lower than G5. This could be due to the higher total sugar content of G5. A noticeable decline in the rate of clearance of G1 and G2 to DPPH was observed as the concentration of polysaccharides decreased, with data decreasing by 84.44 % and 73.71 %, respectively. In contrast, screw speed variation did not promote the antioxidation capacity of samples. The scavenging percentage of G4 even dropped to 2.9 ± 0.11 %, which was the lowest among all samples at 0.2 mg/mL. Shortly, G0 took a higher DPPH radical scavenging capacity than UE, and extrusion with lower moisture content can further improve the capacity.

3.3.5. The •OH radical scavenging capacity of polysaccharides

The scavenging of hydroxyl free radical may avoid biological organisms injury. As showed in Fig. 5C, both of the polysaccharides took good performance in the high concentration range (0.8–2 mg/mL), with the •OH radical scavenging percentage exceeding 85 %, and UE even reached 87.42 ± 0.51 % at 0.8 mg/mL. Compared to the Ve, all polysaccharides exhibited much stronger activity in •OH radical scavenging capacity. The IC50 of Ve (0.59 mg/mL) was much higher than that of all polysaccharides (0.11–0.32 mg/mL). On the other hand, at lower concentration (0.2–0.6 mg/mL), the scavenging capability of polysaccharides were varied. For instance, the scavenging percentage of G1, G2 and G6 dropped sharply by 48.07 %, 55.87 % and 47.41 %, respectively. There was no significant variation in the scavenging percentage of G3 and G4, but the rate of both two samples drop to <70 % at 0.2 mg/mL. Polysaccharides with stable performance in the tested concentration range could associated with higher total phenol contents (G5). Meanwhile, polysaccharides with similar level of total phenol content took the same trend of variation (G5 and G6, Table 1). It also could be speculated that the •OH radical scavenging activities of polysaccharides from DGS correlated closely with their total phenol contents [22].

Compared with the ATBS and DPPH scavenging ability, all polysaccharide samples possessed more effective scavenging ability to •OH radical. It was revealed that the IC50 values for the scavenging effects of Ve on ABTS and DPPH were 0.07 and 0.02 mg/mL, respectively, which were found to be lower than those of all polysaccharides. In terms of the scavenging effects on the •OH radical, the IC50 comparison between Ve and all polysaccharides demonstrated the opposite trend (Table 5). The IC50 of polysaccharides from extruded DGS on ABTS, DPPH and •OH was much less than that of the unextruded sample. It indicated that extrusion, especially treated with lower moisture content, improve the polysaccharide functional properties. Comparing with the rice bran polysaccharide extracted by Qiao, et al. [18], polysaccharides from DGS took stronger radical scavenging capacity on DPPH and similar capacity on ABTS. Meanwhile, the radical scavenging capacity on •OH and DPPH was similar to the polysaccharides from glutinous sorghum [51], which could indicate that sorghum components play an important role in the polysaccharide from DGS.

3.4. Discussion of polysaccharides structure modification and functional properties improvement by extrusion

Extrusion modifies sample structure efficiently by generating energy through shear forces, steam pressures, and heating. The aforementioned energy destabilizes the composition of DGS, leading to modifications in its crystalline structure, reduction in hardness, and liberation of soluble components [18]. These alterations effectively promote the hydrolysis of DGS for polysaccharides extraction. Increasing the extrusion temperature (G2) and screw speed (G4) resulted in a higher yield of polysaccharide compared to unprocessed DGS. However, extrusion without sufficient heat and shear force leading to inefficient hydrolyzation of cellulose and hemicellulose, and did not significantly alter the characteristics of DGS and polysaccharide [34]. Therefore, the yield and molecular weight of polysaccharide dropped significantly in some cases (such as G1 and G3). Meanwhile, excessive water concentration (G6) decreased the temperature in chamber the and undermined the force and energy between screws and DGS, but could impel substance expansion and dissolution [18].

Extrusion also improved the functional properties of polysaccharide, including its α-amylase and starch digestion inhibition capacity, and antioxidation capacity. Polysaccharides with higher galacturonic acid

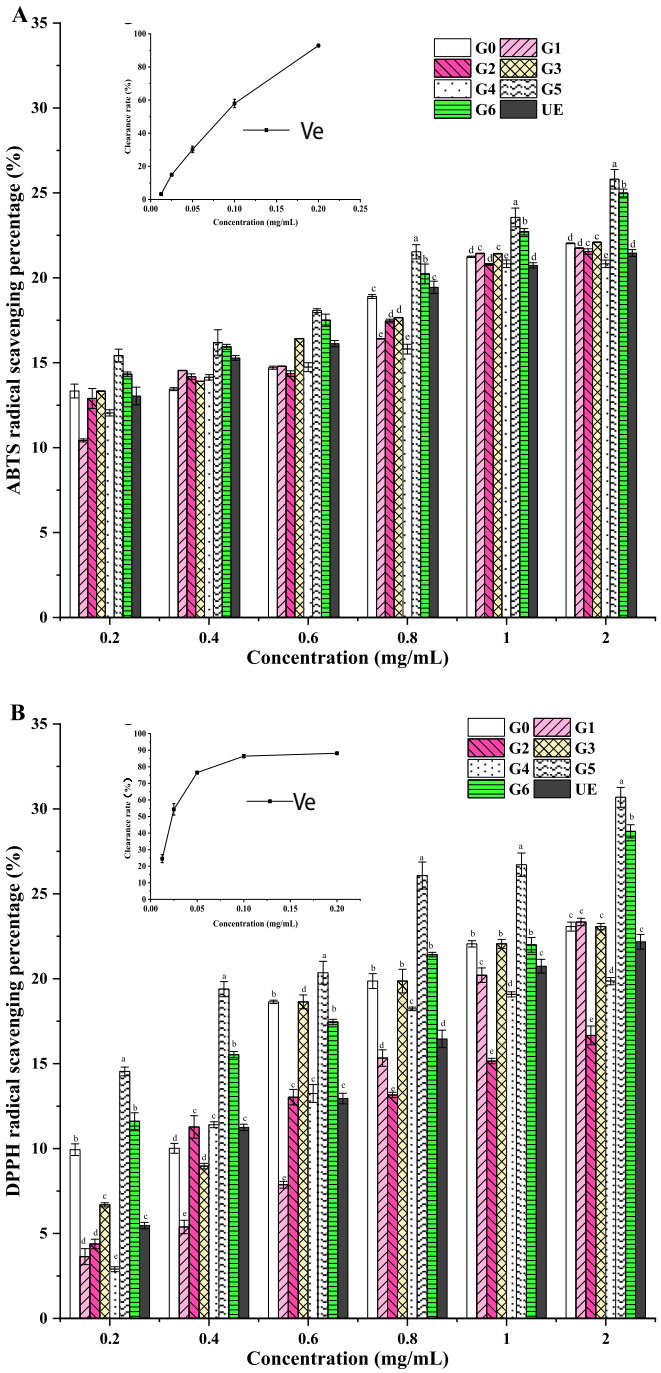

Fig. 5. Antioxidant activity of the polysaccharides from Chinese Baijiu distillers' grains at variated concentrations. The antioxidant activity was detected by testing the scavenging percentage of ABTS (A), DPPH (B) and •OH(C). Different letters indicated significant differences of the different samples at the same concentration, $p < 0.05$.

and molecular weight have been shown to take stronger polysaccharide-enzyme binding action and starch digestion inhibitory activities [22,52]. Extrusion with lower moisture content attributed to polysaccharides took higher concentration of galacturonic acid and phenol contents (both are key factors of polysaccharide bioactivity) and higher molecular weight [20]. Therefore, the functional properties of polysaccharides (G5) can be improved. The increased β-glucan extractability in extruded DGS might be another potential reason of enhanced properties. Lower moisture content extrusion support the sufficient heat and energy generation for higher β-glucan extractability (the ratio of Gal was increased in extruded polysaccharides) [34]. Polysaccharides with higher β-glucan can induce high viscosity in starch solutions and decrease the starch digestion [25]. The results suggested that polysaccharides from extruded DGS have potential values in the production of functional foods with anti-aging and weight reduction benefits.

Comparing in all samples, it can be concluded that variation in extrusion speed and temperature took significant impacts on the physicochemical characteristics of polysaccharides. These modifications resulted in variation in molecular weight, total sugar content, and the ratio of Gal-UA, which in turn affected the functional activity of polysaccharides. Therefore, it is important to carefully control the extrusion speed and temperature in extrusion process to obtain the desired polysaccharides with enhanced functional properties.

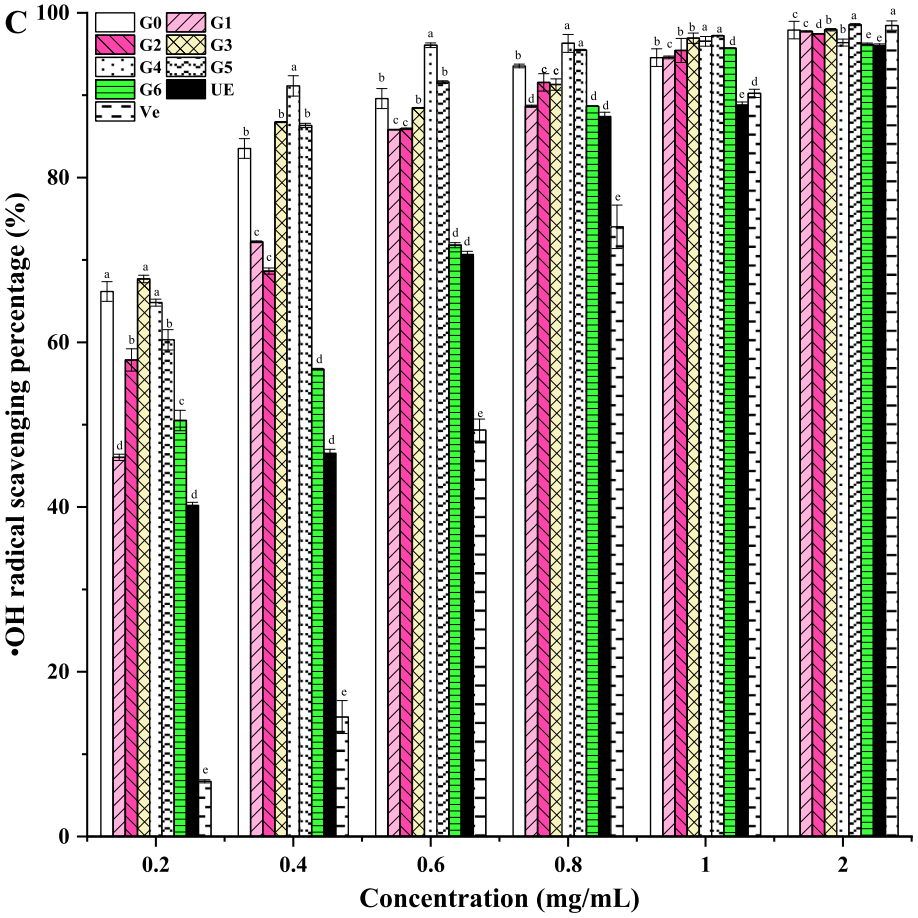

Fig. 5. (*continued*).

Table 5
The IC50 for the scavenging effects of polysaccharides from Chinese Baijiu distillers' grains and Ve on ABTS, DPPH, and •OH radical.

	ABTS (mg/mL)	DPPH (mg/mL)	•OH (mg/mL)
UE	156.21	11.12	0.32
G0	89.86	17.24	0.12
G1	46.31	5.71	0.22
G2	112.44	38.04	0.18
G3	106.43	9.82	0.11
G4	128.24	10.85	0.12
G5	51.39	9.80	0.16
G6	53.55	12.06	0.29
Ve	0.07	0.02	0.59

4. Conclusion

In summary, this study proposed a novel approach to enhance the functional properties and resource value of DGS by extracting non-digestible polysaccharides using an extruder. The extrusion process modified the morphological and crystalline characteristics of DGS, resulting in increased polysaccharide yield (the highest was 5.18 %). Polysaccharides extracted from lower moisture content extruded DGS showed improved functional properties such as α-amylase inhibitory activity, starch hydrolyzation inhibition, and antioxidation compared to polysaccharides from unextruded DGS. These improvements could be mainly associated with modified crystal structure, enriched content of total sugar, uronic acid, total phenol, and Gal-UA, and increased molecular weight. Our results demonstrated that extrusion can efficiently improve the resourcing of non-digestible polysaccharides from DGS for health food production and enhance their functional properties. Nevertheless, further researches are proposed to investigate the relationship between alterations in polysaccharide structure after extrusion and their effects on bioactivity *in vivo*.

CRediT authorship contribution statement

Conceptualization– Yizhou Liu, Suyi Zhang, Beidou Xi and Jian Mao; Visualization– Yizhou Liu, Xiong Li and Rui Chang; Methodology– Yizhou Liu, Xiong Li, and Rui Chang; Validation– Yizhou Liu, Hui Qin, Mengyang Huang; Formal draft preparation– Yizhou Liu; Data curation– Yizhou Liu; Writing– original draft preparation- Yizhou Liu; Writing-review and editing– Shuangping Liu, Xiong Li, Hui Qin, Mengyang Huang and Jian Mao; Supervision –Beidou Xi and Suyi Zhang. All authors have read and agreed to the published version of the manuscript.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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