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# Structural, pasting and thermal properties of common buckwheat (Fagopyrum esculentum Moench) starches affected by molecular structure



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#### ABSTRACT

Common buckwheat starch (CBS) has extensive using value in the human diet. In this study, the molecular structure and physicochemical properties of CBS isolated from five cultivars collected from three regions of China were studied. Variations in molecular structure, crystalline structure, complexity, water solubility (WS), swelling power (SP), pasting properties, and thermal characteristics were recorded among the starches. The CBS had both similarities and differences in its properties by comparison with maize starch (MS) and potato starch (PS). The average molecular weight (M $_{\rm W}$ ) and amylopectin average chain length (ACL) of CBS ranged from 3.86  $\times$  10 $^{7}$  g/mol to 4.68  $\times$  10 $^{7}$  g/mol and from 21.29% to 22.68%, respectively. CBS and MS were divided into one subgroup and showed typical A diffraction patterns, while PS was divided into two subgroups and exhibited a typical B polymorphic pattern. The WS and SP of all the starches significantly increased with increasing temperature and had great variation at 70  $^{\circ}$ C and 90  $^{\circ}$ C. Pearson's correlation analysis showed that the molecular structure of starches greatly affected the physicochemical properties. This study revealed that the physicochemical properties of CBS could be affected by the molecular structures.

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# 1. Introduction

Starch, a main nutrient component in the human diet, has been widely used as an ingredient in many food and non-food applications [1,2]. Starch consists of amylose (largely linear) and amylopectin (branched), the former is a basically linear polymer with  $\alpha\text{-}(1\text{-}4)\text{-linked}$  glucopyranosyl units, while the latter is a highly branched molecule with  $\alpha\text{-}(1\text{-}4)\text{-linked}$  glucopyranosyl units in a chain connected by  $\alpha\text{-}1,6$  linkages [3]. Currently, the relationship between the structure and properties of starch components is still a research hotspot [4]. In particular, the importance of the internal molecular structure of amylopectin in influencing the functional properties of starch has been increasingly understood [2].

Common buckwheat, *Fagopyrum esculentum*, originates from China [5]. It is widely grown in Asia, Europe, and the Americas, and has proven to be a healthy food containing large amounts of starch, protein, and minerals, in addition to other healthful ingredients [6]. Recently,

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common buckwheat has received more attention as a potential material for functional food development and production, and many functional foods derived from common buckwheat have been commercialized, including breads, noodles, and honey [7]. Common buckwheat starch (CBS) accounts for about 70% of the grain, its structural characteristics determine its quality. Thus, understanding the starch molecules of common buckwheat is critical for the food and non-food applications.

Recent studies have shown that the physicochemical properties of starches could be affected by the molecular structures. Lee et al. [8] have found that rice starches with larger amylopectin chains (DP  $\geq$ 37) have a higher gelatinization temperature, whereas Huang et al. [9] determined that there was a negative correlation of swelling power (SP) with amylopectin long branch-chain content of maize starch. Similarly, Zhu and Hao [4] have reported that there are differences in the molecular weight of various potato starches, manifested by significant differences in their crystal structure, thermal properties, and pasting characteristics. However, how the molecular structure of CBS affects its physicochemical properties has not been reported.

In this study, five common buckwheat cultivars collected from three regions were selected to determine how the molecular structure affects the crystalline structure, complexity, pasting properties, and thermal

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characteristics of CBS. Maize starch (MS) and normal potato starch (PS) samples were employed as references. The results of this work will provide important basis for its development.

## 2. Materials and methods

# 2.1. Materials

Five common buckwheat cultivars were collected from three regions in China: Chitian128 (CT), Meng1308 (ME), and Tongqiao4 (TQ) from Inner Mongolia; Guqiao1 (GQ) from Ningxia Hui Autonomous Region; and Xinong9976 (XN) from Shaanxi Province. The temperature and precipitation of the three locations were similar. A normal maize (Shandan683) from the Northwest A&F University and potato (Xiabodi) provided by Yulin Modern Agriculture Demonstration Garden were used for comparison.

#### 2.2. Starch isolation

CBS was isolated from five cultivars through the method by Gao et al. [10]. MS and PS were isolated followed by Zhu and Cui [11].

#### 2.3. Methods

# 2.3.1. Average molecular weight

The GPC-RI-MALS (gel chromatography - differential - multi - Angle laser light scattering) analysis method was used to detect the molecular weight distribution of samples. The main analysis conditions were as follows: RI: Optilab T-rEX (Wyatt technology, CA, USA), MALS: DAWN HELEOS II (Wyatt technology, CA, USA), and Pump: Series 1500 Pump, waters.

#### 2.3.2. Amylopectin chain length distribution

The branched chain distribution of amylopectin was analyzed using the method of Yang et al. [12]. An electrochemical detector and Dionex CarboPac PA200 (3.0  $\times$  250 mm, 062895) ion column were used. Mobile phase A (aqueous solution), mobile phase B (100 mM NaOH and 1 M NaAC), mobile phase C (100 mM NaOH). The flow rate control was 0.4 mL/min, and column temperature control was 30 °C.

# 2.3.3. X-ray diffraction (XRD)

X-ray diffraction was analyzed by employing an X-ray diffractometer (D/Max 2550PC, Rigaku, Japan). The diffraction angle ( $2\theta$ ) was in the range of 5°-50° with a scanning rate of  $10^\circ$ /min. The diffraction peaks at  $2\theta$  were calculated using MDI Jade 6 software.

# 2.3.4. Fourier transformed infrared spectrometry (FTIR) analysis

The short-range ordered structures of starches were analyzed using a Fourier transform infrared spectrometer (7000, Varian, USA) following the method of Guo et al. [13]. The spectra was set from 700  $\rm cm^{-1}$  to 1200  $\rm cm^{-1}$ , and the resolution was 4  $\rm cm^{-1}$ .

# 2.3.5. Bivariate flow cytometric analysis

2 mg of starch, 3  $\mu$ L APTS (20 mM) and 3  $\mu$ L sodium cyanoborohydride (1 M) were mixed in the tubes, and stained at 30 °C in the dark for 15 h. The samples were washed with ddH<sub>2</sub>O for 5 times to make starch suspension. Then the starch suspensions were observed under with an inverted fluorescence microscope (Imager M2, Carl Zeiss, Germany) for the staining to occur. The stained starch suspension was analyzed by flow cytometry [14].

# 2.3.6. Water solubility (WS) and swelling power (SP)

WS and SP were determined following the method of Liu et al. [15] with some modifications. 0.15 g starch ( $m_1$ ) and 5 mL distilled water were mixed and transferred to centrifuge tubes, and heated from

 $50\,^{\circ}\text{C}$  to  $90\,^{\circ}\text{C}$  for 30 min at water bath. Then the samples were cooled to room temperature and centrifuged (3000 rpm, 15 min). The supernatant was poured into a beaker and dried to stable weight ( $m_2$ ) at  $105\,^{\circ}\text{C}$ , and the remaining starch paste was weighed ( $m_3$ ). The WS and SP were calculated as follows:

WS (%) = 
$$(m_2/m_1) \times 100$$
 (1)

$$SP(g/g) = [m_3/(m_1 - m_2)] \times 100$$
 (2)

#### 2.3.7. Thermal properties

The gelatinization properties of starches were studied following the modified method of Zhang et al. [16]. Starch (3 mg) and distilled water (6  $\mu$ L) were added into an aluminum pan, and kept at 4 °C refrigerator for 24 h. Then the samples were heated from 30 °C to 100 °C at a heating rate of 10 °C/min using a differential scanning calorimetry (DSC) (Waters, Q2000, USA).

## 2.3.8. Pasting properties

The pasting behavior of starches were measured with a rapid viscosity analyzer (Perten, Stockholm, Sweden) through the method of Zhang et al. [16]. The main viscosity parameters were obtained from the pasting curves.

## 2.4. Statistical analysis

All samples were repeated three times. Results were expressed as mean  $\pm$  standard deviations. Statistical analyses with Duncan's multiple test (p < .05) were performed with SPSS. 17.0 software.

#### 3. Results and discussion

## 3.1. Molecular weight distribution

A degree of diversity in average molecular weight (M<sub>W</sub>), radius of gyration (R<sub>Z</sub>), and polydispersity index (PDI) of CBS, MS, and PS was recorded in Table 1. For example, the  $M_W$  of CBS ranged from  $3.86 \times 10^7$  g/ mol (XN) to  $4.68 \times 10^7$  g/mol (TQ). Zeng et al. [17] found that the M<sub>W</sub> of waxy rice starch was  $10.38 \times 10^7$  g/mol, and the M<sub>W</sub> of waxy potato starch and normal rice starch were  $5.17 \times 10^7$  g/mol,  $20.17 \times 10^7$  g/ mol, respectively [18,19]. Yang et al. [20] reported that the  $M_W$  of CBS was  $10.7 \times 10^7$  g/mol, which was higher than our results. The difference may be related to the genotypes of common buckwheat varieties and methods of starch isolation. Lee et al. [8] have reported that differences in relative molecular weight would affect the physicochemical properties of rice starch. Therefore, the differences in M<sub>W</sub> of CBS might influence their physicochemical properties. Compared with MS and PS, the M<sub>W</sub> of CBS tended to be the lowest, while PS had the highest M<sub>W</sub>  $(8.35 \times 10^7 \text{ g/mol})$  among the starches. The low M<sub>W</sub> of CBS indicated that it was composed of low-polymerized amylopectin.

Amylopectin with a larger ratio of long branched chains might result in higher  $R_Z$  [21], so it can occupy a larger volume in the solution due to the rotation period of the linear structure [22]. In this study, the  $R_Z$  values of CBS (115.90–158.10 nm) were lower than that of MS and PS, which agreed with the amylopectin chain lengths (Table 1).

PDI was related to the diversity of molecular shapes, and the PDI of typical polymers was usually >1.0 [23]. Similarly, CBS also had the lowest PDI compared with MS and PS, and the results suggested that PS had more heterogeneous molecular weight distribution and included various chains with different DPs.

# 3.2. Chain length distribution of amylopectin

Normalized chromatograms and peak area ratios of chain length distributions in CBS, along with those of MS and PS, are shown in Table 1

 Table 1

 Weight-average molar mass  $(M_W)$ , radius of gyration  $(R_Z)$ , polydispersity index (PDI), chain length distribution, and average chain length (ACL) of amylopectin of starches.

Samples	$M_W$	$R_Z$	PDI	Chain length distr	Average chain length			
	$(\times 10^7, g/mol)$	(nm)		DP 6-12	DP 13-24	DP 25-36	DP ≥ 37	of amylopectin (%)
CT	4.43 ± 0.25 cd	125.10 ± 0.19e	2.72 ± 0.12a	26.85 ± 0.87c	44.24 ± 0.32bc	11.90 ± 0.27c	17.01 ± 0.28a	22.60 ± 0.44a
GQ	$4.16 \pm 0.30 \text{ cd}$	$127.70 \pm 0.20d$	$2.67 \pm 0.18a$	$28.51 \pm 0.52b$	$43.17 \pm 0.28c$	$12.05 \pm 0.31c$	$16.27 \pm 0.19b$	$22.18 \pm 0.37ab$
ME	$4.36 \pm 0.38 \text{ cd}$	$158.10 \pm 0.18b$	$2.70 \pm 0.17a$	$27.65 \pm 0.56$ bc	$43.63 \pm 0.41$ bc	$11.99 \pm 0.16c$	$16.73 \pm 0.17ab$	$22.44 \pm 0.51a$
TQ	$4.68 \pm 0.23c$	$124.90 \pm 0.18e$	$2.68 \pm 0.11a$	$26.66 \pm 0.48c$	$44.36 \pm 0.53b$	$11.95 \pm 0.19c$	$17.00 \pm 0.24a$	$22.68 \pm 0.30a$
XN	$3.86 \pm 0.22d$	$115.90 \pm 0.19f$	$2.70 \pm 0.11a$	$32.04 \pm 0.51a$	$41.89 \pm 0.48d$	$11.77 \pm 0.33c$	$14.30 \pm 0.13c$	$21.29 \pm 0.41b$
Mean	4.30	130.34	2.69	29.35	43.13	11.86	16.27	22.24
MS	$6.21 \pm 0.37b$	$148.25 \pm 0.20c$	$2.75\pm0.14a$	$26.76 \pm 0.61c$	$45.94 \pm 0.29a$	$13.58 \pm 0.39b$	$16.72 \pm 0.33ab$	$22.10 \pm 0.29ab$
PS	$8.35\pm0.43a$	$172.55 \pm 0.18a$	$2.90\pm0.22a$	$22.46 \pm 0.32d$	$44.23\pm0.38bc$	$16.31 \pm 0.22a$	$17.03\pm0.26a$	$23.19 \pm 0.40a$

Different letters within a column indicate significant difference among mean values at p < .05.

and Fig. 1. All of the starches had two obvious peaks. However, some distinct differences were observed among the starches. For CBS and MS, the first peak appeared at DP 12, and the other one peaked at approximately DP 45, while the two larger peaks for PS peaked at DP 13 and DP 49, respectively. Hanashiro et al., [24] reported that the amylopectin branch chains were divided into four parts by degree of polymerization (DP): DP 6–12, 13–24, 25–36, and  $\geq$  37, corresponding to A, B1, B2, and B3+ chains. In our study, the A, B1, B2, and B3+ chains of the starches were 22.46-32.04%, 41.89-45.94%, 11.77-16.31%, and 14.30–17.03%, respectively. The amylopectin in CBS had a higher percentage of A chains but a lower percentage of B3+ chains than MS and PS, and the chain length distributions of CBS were significantly different among five cultivars, which might be attributed to their genotype. Lin and Chang [25] have reported that the amylopectin branch chains (DP 6-24) would form a double helix structure, while the A chain could form a sub-crystalline structure. The average chain length (ACL) of CBS varied from 21.29% (XN) to 22.68% (TQ), which was higher than that of MS but lower than that of PS. Kim et al. [26] believed that amylopectin chain length distribution greatly affected the physicochemical properties, therefore, it could be speculated that the physicochemical properties of CBS would also be influnced by the molecular structure.

# 3.3. Crystalline structure

Generally, natural starches are divided into type A, B, and C in line with their X-ray diffraction (XRD) spectra [27]. Grain crystallinity is thought to be caused by the AP chain clusters of DP 13-15 [28]. XRD patterns and relative crystallinity were presented in Fig. 1 and Table 2, respectively. In this study, all the CBS varieties showed the A-type diffraction patterns, and the strong peaks appeared near 15° and 23°, while the unresolved peaks appeared at 17° and 18° (Fig. 2A). There was no significant difference in peak positions among different cultivars, and MS displayed similar results with CBS, which were common to cereal starches. However, PS exhibited a typical B polymorphic pattern, which agreed with the results studied by Gao et al. [10]. Therefore, the polymorph type was directly related to the internal unit chain composition [29]. Amylopectin is the main crystalline component of starch granules with short chains forming local organizations [27]. The relative crystallinity of CBS ranged from 24.74% to 27.19%, which was similar to MS but higher than PS. Raghunathan et al. [28] reported that the differences in relative crystallinity among different starches could be influnced by the degree of double helix accumulation in the crystalline layer, and the higher the degree of AP branching, the lower the relative crystallinity. As shown in Table 1, CBS and MS with lower  $M_W$  and lower proportion of long chains of amylopectin had higher relative crystallinity, while PS with higher average chain length of amylopectin had lower relative crystallinity. These results indicated that the AP branching degree of CBS was lower than PS. Longer chains could form more ferfect crystalline structures [8]. In this study, PS had the most stable crystal structure as compared with CBS and MS, this stability may be related to the crop variety, growing conditions, and starch granule sizes [30].

#### 3.4. Short-range ordered structure

The short-range ordered structure of starch granules can be well reflected by the FTIR spectra. The order degree and ratio of disordered carbohydrate structure could be measured by the absorbance ratio of 1045/1022 cm<sup>-1</sup> and 1022/995 cm<sup>-1</sup>, respectively [31]. The deconvoluted FTIR spectra of CBS, MS, and PS are reflected in Fig. 2B, and their FTIR ratios are summarized in Table 2. The FTIR spectra of five common buckwheat cultivars displayed no significant differences, but the absorbance ratios of 1045/1022 cm<sup>-1</sup> and 1022/995 cm<sup>-1</sup> were different, and ranged from 0.688 to 0.728 (CT < XN < ME < GQ < TQ) and from 0.768 (XN) to 0.852 (TQ), respectively. The ratio of 1045/ 1022 cm<sup>-1</sup> of MS was higher than that of CBS but lower than PS, while there was the lowest ratio of  $1022/995 \, \mathrm{cm}^{-1}$  in PS but the highest in MS. Zhang, et al. [32] found that the  $1045/1022 \, \mathrm{cm}^{-1}$  ratios of purple sweet potato varied from 0.631 to 0.671, and those of five fruits kernel starches ranged from 0.576 to 0.654 by Guo et al. [31]. Differences in FTIR spectra among various starches may be related to biological origin, amylose content, and molecular structure.

#### 3.5. Bivariate flow cytometric analysis

Flow cytometry is widely used for particle classification, and the starch suspension is divided into subgroups of different sizes [20]. The histograms of SSC versus FSC reflect the overall structural complexity of starch granules, while FSC versus FITC represent the size and internal structure of starch granules [16]. As shown in Fig. 3, PS was divided into two subgroups (P1 and P2) on the flow biparameter diagram. The P1 subgroup was located in the upper right corner of the diagram with a proportion of 52.8%, which was the largest population in terms of particle size and complexity. However, the P2 subgroup, accounting for 6.8%, was smaller than the P1 group in terms of volume and complexity. Unlike PS, CBS and MS were only divided into one group (P1) by flow

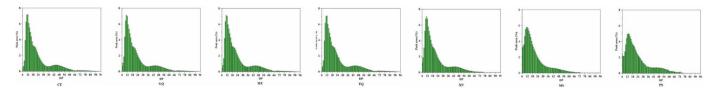


Fig. 1. Peak area ratios of chain length distributions in common buckwheat, maize, and potato starches.

**Table 2**Water solubility, swelling power, relative crystallinity, and FTIR ratios of starches.

Samples	Water solubili	ty (%)			Swelling power (g/g)		Relative	FTIR ratios	
	50 °C	70 °C	90 °C	50 °C	70 °C	90 °C	crystallinity (%)	1045/1022 cm <sup>-1</sup>	1022/995 cm <sup>-1</sup>
CT	2.04 ± 0.11c	10.90 ± 0.07e	13.95 ± 0.06e	3.13 ± 0.04e	15.22 ± 0.25e	24.45 ± 0.15f	26.73 ± 0.15ab	0.688 ± 0.02e	0.796 ± 0.05c
GQ	$2.30 \pm 0.14c$	$11.32 \pm 0.10c$	$15.15 \pm 0.10c$	$3.39 \pm 0.03c$	$16.06 \pm 0.14c$	$27.12 \pm 0.25d$	$24.74 \pm 0.41c$	$0.713 \pm 0.01d$	$0.832 \pm 0.04b$
ME	$2.19 \pm 0.09c$	$11.06 \pm 0.05$ de	$14.47 \pm 0.04d$	$3.17 \pm 0.05$ de	15.44 ± 0.16de	$24.86 \pm 0.04ef$	$25.35 \pm 0.25c$	$0.712 \pm 0.02d$	$0.851 \pm 0.04a$
TQ	$2.21 \pm 0.10c$	$11.20 \pm 0.06$ cd	$14.60 \pm 0.03d$	$3.27 \pm 0.04$ cde	15.67 ± 0.07cde	$25.12 \pm 0.17e$	$27.19 \pm 0.12a$	$0.728 \pm 0.03b$	$0.852\pm0.04$ a
XN	$2.28 \pm 0.11c$	$11.31 \pm 0.05c$	$15.09 \pm 0.08c$	$3.35 \pm 0.06$ cd	$15.79 \pm 0.15$ cd	$28.38 \pm 0.20c$	$26.69 \pm 0.24ab$	$0.694 \pm 0.01e$	$0.768 \pm 0.03d$
Mean	2.20	11.16	14.65	3.26	15.64	25.99	26.14	0.71	0.82
MS	$3.64 \pm 0.12b$	$14.52 \pm 0.06b$	$24.64 \pm 0.13b$	$4.31 \pm 0.10b$	$19.47 \pm 0.12b$	$32.41 \pm 0.14b$	$26.48 \pm 0.12b$	$0.721 \pm 0.01c$	$0.836 \pm 0.05b$
PS	$4.97\pm0.08a$	$18.76 \pm 0.09a$	$32.97 \pm 0.18a$	$6.16 \pm 0.09a$	$34.79 \pm 0.26a$	$48.77 \pm 0.21a$	$17.31 \pm 0.11d$	$0.740\pm0.02a$	$0.802\pm0.02c$

Different letters within a column indicate significant difference among mean values at p < .05.

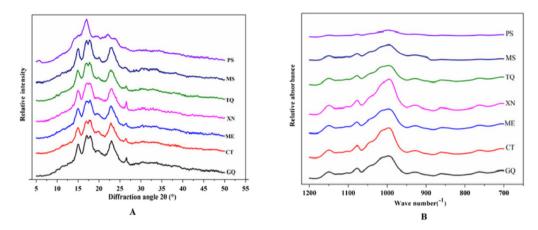


Fig. 2. XRD pattern (A), and FITR spectrum (B) of common buckwheat, maize, and potato starches.

cytometry, which indicated that the CBS and MS groups were a homogeneous group with relatively uniform particle size and complexity. Zhang et al. [16] reported that Tartary buckwheat starches were composed of three subgroups, with the difference possibly being related to the starch molecular weight, amylopectin chain length distribution, and genotype.

# 3.6. WS and SP

Diversity in WS and SP of starches are recorded in Table 2. The WS and SP of all starches significantly increased with increasing temperature, and great variations were observed at 70 °C and 90 °C, respectively. PS showed the highest WS and SP, followed by MS, while CBS had the

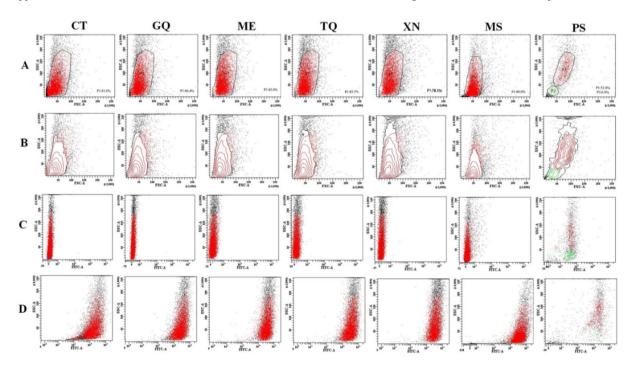


Fig. 3. Bivariate flow cytometric histograms of common buckwheat, maize, and potato starches. A: forward scattered-side scattered image; B: fluorescence image; C: image of unstained starch (negative control); D: image of 1-aminopyrene-3,6,8-trisulfonic acid (APTS) stained starch.

**Table 3** Pasting properties and thermal properties of starches.

Samples	Pasting properties					Thermal properties				
	PV (cP)	TV(cP)	BD (cP)	FV(cP)	SB (cP)	PT (°C)	To (°C)	Tp (°C)	Tc (°C)	ΔH (J/g)
CT	2289 ± 64b	1531 ± 38 cd	758 ± 9b	2465 ± 28d	934 ± 9d	77.63 ± 0.45abc	66.52 ± 0.49a	71.50 ± 0.03a	77.08 ± 0.15a	8.30 ± 0.52bc
GQ	$2423 \pm 55b$	$1635 \pm 63c$	$788 \pm 8b$	$3429 \pm 58ab$	$1794 \pm 7b$	$75.53 \pm 0.25d$	$64.95 \pm 0.05ab$	$69.04 \pm 0.03c$	$76.80 \pm 0.06e$	$7.75 \pm 0.13 \text{ cd}$
ME	$2304\pm28b$	$1336\pm66d$	$968 \pm 38a$	$3602 \pm 8ab$	$2267\pm73a$	$79.60 \pm 0.64a$	$65.02\pm0.26ab$	$70.86 \pm 0.05b$	$71.66 \pm 0.06d$	$7.34\pm0.02$ cd
TQ	$2376\pm56b$	$1903\pm24b$	$473\pm32c$	$3061 \pm 80c$	$1159\pm56d$	$75.63 \pm 0.71 \text{ cd}$	$65.56 \pm 0.41ab$	$69.12 \pm 0.16c$	$75.08 \pm 0.21b$	$6.83 \pm 0.08d$
XN	$2390\pm33b$	$2268\pm36a$	$122 \pm 3d$	$2919 \pm 66c$	$651 \pm 30e$	$78.78\pm0.22ab$	$62.59 \pm 0.36c$	$65.68 \pm 0.28d$	$70.19 \pm 0.02e$	7.77 $\pm$ 0.02 cd
Mean	2356	1735	616	3095	1361	77.43	64.93	69.24	74.16	7.60
MS	$1980 \pm 69c$	$1514 \pm 41$ cd	$467 \pm 27c$	$3381 \pm 86b$	$1867\pm45b$	$76.78\pm0.01bcd$	$63.88 \pm 0.17$ bc	$69.41 \pm 0.01c$	$73.63 \pm 0.70c$	$9.32 \pm 0.14b$
PS	$3038\pm79a$	$2210\pm14a$	$828\pm65ab$	$3679\pm47a$	$1470\pm61c$	$69.80 \pm 0.26e$	$57.82 \pm 0.83d$	$63.77 \pm 0.01e$	72.95 $\pm$ 0.18 cd	$15.07\pm0.36a$

Different letters within a column indicate significant difference among mean values at p < .05.

PV: peak viscosity; TV: trough viscosity; BD: breakdown; FV: final viscosity; SB: setback; PT: pasting temperature; To: onset temperature; Tp: peak temperature; Tc: conclusion temperature; ΔH: gelatinization enthalpy.

lowest WS and SP, and the mean of WS and SP ranged from 2.20 to 14.65%, and from 3.26 to 25.99 g/g, respectively. WS and SP can reflect the magnitude of the interaction between starch and water [33]. Lii et al. [34] reported that the SP was inversely proportional to amylose content, and the more amylopectin short branched chains, the higher the SP, while the more long branched chains, the lower the SP [35]. In this study, different varieties of starch amylopectin length distribution led to different WS and SP.

#### 3.7. Pasting properties

Pasting properties of starches showed significant variation (p < .05) among the cultivars (Table 3). For example, the PV, TV, BD, FV, SB, and PT of CBS ranged from 2289 to 2423 cP, from 1903 to 2268 cP, from 122 to 969 cP, from 2465 to 3429 cP, and from 75.53 °C to 79.60 °C, respectively. PV is caused by the friction of starch granules after full water absorption and expansion, and can reflect starch swelling capacity [36]. FV is due to the decreased movement of the water molecules surrounded by amylose and amylopectin after the temperature drops, causing the viscosity to rise again [37]. Among the tested CBS, CT

showed the lowest PV and FV, and GQ had relatively higher FV. MS was found to have lower PV than the CBS, while having similar TV, BD, FV, and SB to some of the CBS, and PS had the highest PV and FV. PT shows that starch viscosity starts to increase during heating [38]. Compared with MS and PS, CBS has the highest PT value, which may be related to the molecular weight distribution. Zhou et al. [39] found that high amylose contents contributed to higher pasting temperatures, these data indicated that CBS had the highgest amylose content among the tested samples. Amylopectin branch chain distribution and molecular weight are the main factors affecting starch pasting performance [40]. Patindol et al. [41] found that PV may be determined primarily by the size of amylopectin molecules. Higher PV in PS might be attributed to its higher M<sub>W</sub> and ACL (Table 1).

#### 3.8. Thermal properties

The starch gelatinization transition temperatures (onset,  $T_o$ ; peak,  $T_p$ ; and conclusion;  $T_c$ ), along with gelatinization enthalpy ( $\Delta H$ ) of starches were summarized in Table 3. Compared with MS and PS, it was found that CBS exhibited the highest  $T_o$  and  $T_c$  but the lowest  $\Delta H$ .

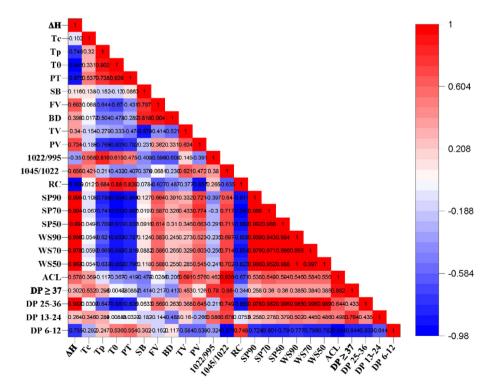


Fig. 4. Pearson's correlation analysis of structure-property relationships of common buckwheat, maize, and potato starches.

In contrast, the highest  $\Delta H$  was shown in PS. The results of this study were consistent with those of Gao et al. [10]. For CBS, the T<sub>0</sub>, T<sub>p</sub>, T<sub>c</sub>, and  $\Delta H$  varied significantly among different cultivars. For example, the To of five cultivars ranged from 62.59 °C to 66.52 °C (XN < GQ < ME < TQ < CT), and the  $\Delta H$  were found in a range of 6.83-8.30 J/g (TQ < ME < GQ < XN < CT), the highest thermal performance of CT may be related to the higher average chain length of amylopectin. Though the five common buckwheat cultivars had typical A starch, different amylose content, Mw, and ACL may lead to different thermal properties. Gelatinization temperature can be used to evaluate whether the crystal is perfect, the higher the gelatinization temperature is, the more perfect the crystal structure is [42,43], and the molecular structure of amylopectin is related to the crystal structure of starch granules [44]. Park et al. [45] reported that gelatinization temperatures were positively correlated with long chains of amylopectin. Noda et al. [46] found that short chains (DP 6–12) of amylopectin were negatively correlated with gelatinization temperatures, and the short chains of amylopectin might induce a decrease in the stability of the double helices. Shin et al. [18] also determined that amylopectin with longer average chain lengths could accelerate the retrogradation. Gidley and Bulpin [44] reported that high proportion of short amylopectin chains contributed to lower gelatinization temperatures, while our results showed that CBS with the highest amount of short chains compared with MS and PS exhibited much higher  $T_0$ ,  $T_p$  and  $T_c$ . The difference could be influnced by the molecular architecture of the crystalline region [28].

#### 3.9. Correlation analysis of starches

Pearson's analysis revealed the correlation among the molecular structures and physicochemical properties of CBS, MS, and PS (Fig. 4). In this study, the data reflected that WS and SP (at 50 °C, 70 °C, and 90 °C) were significantly negatively correlated with the short chains of amylopectin and positively correlated with ACL. Shi et al. [47] found that the long branched chains in amylopectin could increase the stability of the double helix and induce higher gelatinization temperature. The gelatinization temperature was positively correlated with amylopectin long branch chains (DP  $\geq$ 37) and negatively correlated with ACL, while the gelatinization enthalpy was positively correlated with average chain length and significantly negatively correlated with amylopectin proportion of DP 6–12. The results were consistent with the various maize starches reported by Lin et al. [33].

# 4. Conclusion

Five common buckwheat varieties collected from three regions were investigated compared with maize and potato starches in this study. Starches from different cultivars had different molecular weight distribution, amylopectin chain length distribution, water solubility, swelling power, pasting properties and thermal characteristics, but all had one subgroup and exhibited typical A crystalline structure, while potato starch were divided two subgroups and showed typical B polymorph pattern. The differences of physicochemical properties of starches had significant relationship with the molecular structure. The results of this study indicated that common buckwheat starch can be a new source of starch with potential food and nonfood applications.

#### **CRediT** authorship contribution statement

**Licheng Gao:**Conceptualization, Software, Writing - original draft, Writing - review & editing.**Honglu Wang:**Investigation, Data curation. **Chenxi Wan:**Methodology, Software.**Jiajun Leng:**Investigation, Data curation.**Pu Yang:**Writing - review & editing.**Xiaoli Gao:**Writing - review & editing.**Jinfeng Gao:**Resources, Conceptualization, Funding acquisition, Supervision.

# **Declaration of competing interest**

The authors declare that there is no conflict of interests regarding the publication of this paper.

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#### References

- [1] H. Yong, X. Wang, J. Sun, Y. Fang, J. Liu, C. Jin, Comparison of the structural characterization and physicochemical properties of starches from seven purple sweet potato varieties cultivated in China, Int. J. Biol. Macromol. 120 ( (2018) 1632–1638
- [2] F. Zhu, Relationships between amylopectin internal molecular structure and physicochemical properties of starch, Trends Food Sci. Technol. 78 (2018) 234–242.
- [3] N. Vanier, S. Halal, A. Dias, E. Zavareze, Molecular structure, functionality and applications of oxidized starches: a review, Food Chem. 221 (2016) 1546–1559.
- [4] F. Zhu, C. Hao, Physicochemical properties of Maori potato starch affected by molecular structure, Food Hydrocoll. 90 (2019) 248–253.
- [5] X. Fang, Y. Li, J. Nie, C. Wang, K. Huang, Y. Zhang, Y. Zhang, H. She, X. Liu, R. Ruan, X. Yuan, Z. Yi, Effects of nitrogen fertilizer and planting density on the leaf photosynthetic characteristics, agronomic traits and grain yield in common buckwheat (Fagopyrum esculentum M.), Field Crop Res. 219 (2018) 160–168.
- [6] D. Yu, J. Chen, J. Ma, H. Sun, Y. Yuan, Q. Ju, Y. Teng, M. Yang, W. Li, K. Fujita, E. Tatsumi, G. Luan, Effects of different milling methods on physicochemical properties of common buckwheat flour, LWT-Food Sci. Technol. 92 (2018) 220–226.
- [7] Y. Xiao, H. Liu, T. Wei, J. Shen, M. Wang, Differences in physicochemical properties and in vitro digestibility between tartary buckwheat flour and starch modified by heat-moisture treatment, LWT-Food Sci. Technol. 86 (2017) 285–292.
- [8] S. Lee, J. Lee, H. Chung, Impact of diverse cultivars on molecular and crystalline structures of rice starch for food processing, Carbohydr. Polym. 169 (2017) 33–40.
- [9] J. Huang, Z. Shang, J. Man, Q. Liu, C. Zhu, C. Wei, Comparison of molecular structures and functional properties of high-amylose starches from rice transgenic line and commercial maize, Food Hydrocoll. 46 (2015) 172–179.
- [10] J. Gao, I. Kreft, G. Chao, Y. Wang, X. Liu, L. Wang, P. Wang, X. Gao, B. Feng, Tartary buckwheat (Fagopyrum tataricum Gaertn.) starch, a side product in functional food production, as a potential source of retrograded starch, Food Chem. 190 (2016) 552–558.
- [11] F. Zhu, R. Cui, Comparison of molecular structure of oca (Oxalis tuberosa), potato, and maize starches, Food Chem. 296 (2019) 116–122.
- [12] Q. Yang, P. Zhang, Y. Qu, X. Gao, J. Liang, P. Yang, B. Feng, Comparison of physico-chemical properties and cooking edibility of waxy and non-waxy proso millet (Panicum miliaceum L.), Food Chem. 257 (2018) 271–278.
- [13] K. Guo, T. Liu, A. Xu, L. Zhang, X. Bian, C. Wei, Structural and functional properties of starches from root tubers of white, yellow, and purple sweet potatoes, Food Hydrocoll. 89 (2019) 829–836.
- [14] X. Zhang, J. Feng, H. Wang, J. Zhu, Y. Zhong, L. Liu, S. Xu, R. Zhang, X. Zhang, J. Xue, D. Guo, Bivariate flow cytometric analysis and sorting of different types of maize starch grains, Cytometry A 93 (2018) 213–221.
- [15] H. Liu, L. Wang, R. Cao, H. Fan, M. Wang, In vitro digestibility and changes in physicochemical and structural properties of common buckwheat starch affected by high hydrostatic pressure, Carbohydr. Polym. 144 (2016) 1–8.
- [16] W. Zhang, Q. Yang, M. Xia, W. Bai, P. Wang, X. Gao, J. Li, B. Feng, J. Gao, Effects of nitrogen level on the physicochemical properties of Tartary buckwheat (Fagopyrum tataricum (L.) Gaertn.) starch, Int. J. Biol. Macromol. 129 (2019) 799–808.
- [17] F. Zeng, Q. Gao, Z. Han, X. Zeng, S. Yu, Structural properties and digestibility of pulsed electric field treated waxy rice starch, Food Chem. 194 (2016) 1313–1319.
- [18] H. Shin, S. Choi, C. Park, T. Moon, Preparation of starches with low glycaemic response using amylosucrase and their physicochemical properties, Carbohydr. Polym. 82 (2) (2010) 489–497.
- [19] S. You, S. Oh, H. Kim, H. Chung, Influence of molecular structure on physicochemical properties and digestibility of normal rice starches, Int.J.Biol. Macromol. 77 (2015) 375–382.
- [20] Q. Yang, W. Zhang, Y. Luo, J. Li, J. Gao, P. Yang, X. Gao, B. Feng, Comparison of structural and physicochemical properties of starches from five coarse grains, Food Chem. 288 (2019) 283–290.
- [21] M. Millard, Light scattering molecular weights and intrinsic viscosities of processed waxy maizes starches in 90% dimethyl Sulfoxide and H<sub>2</sub>O, Cereal Chem. 74 (5) (1997) 687–691
- [22] S. Shin, C. Lee, M. Kim, S. Choi, H. Choi, Y. Kim, T. Moon, Structural characteristics of low-glycemic response rice starch produced by citric acid treatment, Carbohydr. Polym. 78 (3) (2009) 588–595.
- [23] G. Lowry, Polydispersity index having particular applicability to narrow molecular weight distribution polymers, J. Polym. Sci., Part B: Polym. Lett. 1 (9) (1963) 489–490.

- [24] I. Hanashiro, J. Abe, S. Hizukuri, A periodic distribution of the chain length of amylopectin as revealed by high-performance anion-exchange chromatography, Carbohydr. Res. 283 (1996) 151–159.
- [25] J. Lin, Y. Chang, Effects of type and concentration of polyols on the molecular structure of corn starch kneaded with pullulanase in a Farinograph, Food Hydrocoll. 20 (2–3) (2006) 340–347.
- [26] Y. Kim, K. Woo, H. Chung, Starch characteristics of cowpea and mungbean cultivars grown in Korea. Food Chem. 263 (2018) 104–111.
- [27] W. Norman, L. Tao, Variation in crystalline type with amylose content in maize starch granules: an X-ray powder diffraction study, Carbohydr. Polym. 36 (4) (1988) 277–284.
- [28] R. Raghunathan, R. Hoover, R. Waduge, Q. Liu, T. Warkentin, Impact of molecular structure on the physicochemical properties of starches isolated from different field pea (Pisum sativum L.) cultivars grown in Saskatchewan, Canada, Food Chem. 221 (2017) 1514–1521.
- [29] F. Zhu, C. Hao, Molecular structure of Maori potato starch, Food Hydrocoll. 80 (2018) 206–211.
- [30] H. Zhou, J. Wang, H. Zhao, X. Fang, Y. Sun, Characterization of starches isolated from different Chinese Baizhi (Angelica dahurica) cultivars, Starch - Stärke 62 (3-4) (2010) 198-204.
- [31] K. Guo, L. Lin, X. Fan, L. Zhang, C. Wei, Comparison of structural and functional properties of starches from five fruit kernels, Food Chem. 257 (2018) 75–82.
- [32] L. Zhang, L. Zhao, X. Bian, K. Guo, L. Zhou, C. Wei, Characterization and comparative study of starches from seven purple sweet potatoes, Food Hydrocoll. 80 (2018) 168–176.
- [33] L. Lin, C. Cai, R. Gilbert, E. Li, J. Wang, C. Wei, Relationships between amylopectin molecular structures and functional properties of different-sized fractions of normal and high-amylose maize starches, Food Hydrocoll. 52 (2016) 359–368.
- [34] C. Lii, M. Tsai, K. Tseng, Effect of amylose content on the rheological property of rice starch, Cereal Chem. 73 (4) (1996) 415–420.
- [35] H. Salman, J. Blazek, A. Lopez-Rubio, E. Gilbert, T. Hanley, L. Copeland, Structure-function relationships in A and B granules from wheat starches of similar amylose content, Carbohydr. Polym. 75 (3) (2009) 420–427.

- [36] Q. Liu, E. Donner, Y. Yin, R. Huang, M. Fan, The physicochemical properties and in vitro digestibility of selected cereals, tubers and legumes grown in China, Food Chem. 99 (3) (2006) 470–477.
- [37] P. Ambigaipalan, R. Hoover, E. Donner, Q. Liu, S. Jaiswal, R. Chibbar, K. Nantanga, K. Seetharaman, Structure of faba bean, black bean and pinto bean starches at different levels of granule organization and their physicochemical properties, Food Res. Int. 44 (9) (2011) 2962–2974.
- [38] A. Kaur, N. Singh, R. Ezekiel, N. Sodhi, Properties of starches separated from potatoes stored under different conditions, Food Chem. 114 (4) (2009) 1396–1404.
- [39] Q. Zhou, W. Shi, X. Meng, Y. Liu, Studies on the morphological, crystalline, thermal properties of an under utilized starch from yam, Dioscoreae zingiberensis, C. H. Wright, Starch - Stärke 65 (1–2) (2013) 123–133.
- [40] J. Jane, Y. Chen, L. Lee, A. Mcpherson, T. Kasemsuwan, Effects of amylopectin branch chain length and amylose content on the gelatinization and pasting properties of starch 1, Cereal Chem. 76 (5) (1999) 629–637.
- [41] J. Patindol, B. Gonzalez, Y. Wang, A. McClung, Starch fine structure and physico-chemical properties of specialty rice for canning, J. Cereal Sci. 45 (2) (2007) 209–218
- [42] H. Chung, Q. Liu, L. Lee, D. Wei, Relationship between the structure, physicochemical properties and in vitro digestibility of rice starches with different amylose contents, Food Hydrocoll. 25 (5) (2011) 968–975.
- [43] K. Vallons, E. Arendt, Effects of high pressure and temperature on buckwheat starch characteristics, Eur. Food Res. Technol. 230 (2) (2009) 343–351.
- [44] M. Gidley, P. Bulpin, Crystallization of malto-oligosaccharides as models of the crystalline forms of starch: minimum chain-length requirement for the formation of double helices, Carbohydr. Res. 161 (2) (1987) 291–300.
- [45] I. Park, A. Ibanez, F. Zhong, Gelatinization and pasting properties of waxy and nonwaxy rice starches, Starch - Stärke 59 (8) (2007) 388–396.
- [46] T. Noda, Y. Nishiba, T. Satto, I. Soda, Properties of starches from several low-amylose rice cultivars, Cereal Chem. 80 (2003) 193–197.
- [47] Y. Shi, T. Capitani, P. Trzasko, R. Jeffcoat, Molecular structure of a low-amylopectin starch and other high-amylose maize starches, J. Cereal Sci. 27 (3) (1998) 299.