

Preparation and Physicochemical Properties of *Cyperus esculentus* Starch from its Tubers Using Ultrasound-assisted Alkali Method

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Cyperus esculentus tubers are rich in starch, oil, protein, dietary fiber, and other nutrients. Ultrasonic treatment can reduce the combination of starch, protein, and dietary fiber in *C. esculentus* tubers during extraction of *C. esculentus* starch, thereby improving the extraction yield and shortening of the extraction time. In this study, the extraction yield of *C. esculentus* starch was 92.2% using ultrasound-assisted alkali method. The microstructure analysis showed that the granule characteristics of *C. esculentus* starch and other starches were similar. X-ray diffraction analysis showed that *C. esculentus* starch possessed an A-type crystal structure. The onset temperature of gelatinization endotherm and peak temperature of gelatinization of *C. esculentus* starch were only lower than those of sweet potato starch, and higher than other starches, which is 67.9 °C. The content of resistance starch (RS) (11.0%) in *C. esculentus* starch was the highest among the six starches. As an underutilized resource, *C. esculentus* is a new crop with high quality, high yield, and high comprehensive utilization value. Its aerial parts can be used as feed, green manure, and its underground parts can be edible and oily. *C. esculentus* starch can be a valuable source to develop into new functional food.

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Keywords: Starch; *Cyperus esculentus*; Ultrasound-assisted; Physicochemical properties

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INTRODUCTION

Cyperus esculentus L., also known as underground chestnut, underground walnut, tiger nut, etc., is a perennial herb of the Cyperaceae family (Djikeng *et al.* 2022). *C. esculentus* is a crop with stress resistance, high yield, high quality, economic, and ecological benefits. Its aerial parts can be used as feed, green manure, and its underground parts are edible and oily (Adewuyi *et al.* 2015; Li *et al.* 2022). *C. esculentus* has a long history of cultivation. It was brought to Europe by the Arabs in the 8th century BC and is now widely distributed in Africa, Europe, Asia, South America, and North America. At present, the country with the largest planting area of *C. esculentus* is the United States, followed by Canada and Spain. It is also widely planted in Egypt, Ghana, China, Russia, and other countries (Jing *et al.* 2012; López-Cortés *et al.* 2013; Zhang *et al.* 2022). Due to the strong stress resistance and ecological adaptability of *C. esculentus*, it is planted in the northwest of China, Huanghuai, and other areas with severe soil desertification in China (Johnson *et al.* 2007; Shklavtsova *et al.* 2013).

Cyperus esculentus is an important multi-purpose economic crop. Its stems and leaves can be used as green feed or as raw materials for weaving. The tuber system has a fragrant smell and can be used to extract essential oils. *C. esculentus* tubers are rich in fat, starch, protein, dietary fiber, and other ingredients; they are used for manufacturing materials, such as cooking oil, soymilk, leisure food, *etc.* (Adelakun *et al.* 2021; Cui *et al.* 2021). *C. esculentus* starch is an odorless, bright white or off-white powder. Due to the different varieties and sizes of *C. esculentus* tubers, the starch yield varies, ranging from about 14% to 37% (Umerie *et al.* 1997; Liu *et al.* 2020). Similar to other sources of starches, the granules of *C. esculentus* starch are formed by alternating crystalline regions and non-crystalline regions (Manek *et al.* 2012; Yu *et al.* 2022). Compared with potato starch granules, most of the starch granules of *C. esculentus* tubers show spherical and elliptical shapes, and a few showed irregular shapes (Akonor *et al.* 2019; Sabah *et al.* 2019). After using pullulanase (PUL) for debranching reaction of *C. esculentus* starch, the content of slow-digestible starch increased significantly, and the viscoelasticity of starch paste decreased, but the material still showed typical pseudoplasticity, which can be widely used in in soups or condiments (Li *et al.* 2017).

Starch, as one of the main components of *C. esculentus* tubers, has an important influence on the physicochemical properties of products. As an oil crop, it is also valuable to develop into extraction of edible oil (Liu *et al.* 2019). However, due to insufficient understanding of its physicochemical properties and functional properties, the current processing and utilization of *C. esculentus* starch is still limited (Jing *et al.* 2012; Nwosu *et al.* 2022). Ultrasonic treatment can reduce the combination of starch, protein, and dietary fiber, especially in *C. esculentus* tubers, which is rich in dietary fiber. The treatment can improve the extraction of starch from *C. esculentus* tubers, thereby shortening the extraction time (Cui *et al.* 2021; Yusoff *et al.* 2022). In this study, ultrasound-assisted alkali method was used as the method to precipitate the protein in *C. esculentus* tubers, and the starch of *C. esculentus* tubers was obtained. The ultrasonic conditions were optimized to improve the extraction yield of starch. Through scanning electron microscopy, X-ray diffraction, and differential scanning calorimetry (DSC) analyses of the five types of starches, including those from wheat, dent corn, sweet potato, potato, and tapioca, the different characteristics of the *C. esculentus* starch and other starches were analyzed.

EXPERIMENTAL

Materials and Chemicals

Cyperus esculentus tubers were provided by Dingzhou Laowei Agricultural Technology Co., Ltd. (Hebei, China). Five types of starches, including wheat, dent corn, sweet potato, potato, and tapioca, were purchased from Xinxiang Liangrun Whole Grain Food Co., Ltd. (Henan, China). Alpha amylase was purchased from Sigma-Aldrich Trading Co., Ltd. (Shanghai, China). Glucoamylase was provided by Shanghai Yuanye Biotechnology Co., Ltd. (Shanghai, China). All other reagents used were of analytical grade products.

Preparation of *Cyperus esculentus* Starch

After washing *C. esculentus* tubers, they were dried in a convection drying oven at 45 °C for 24 h to keep the moisture content below 20%. A wall breaker (Kenwood BLP 900BK, DeLong Electric Co., Ltd., Shanghai, China) was used for wall breaking treatment.

The particle size at this stage was about 40-mesh, and then petroleum ether was used as a degreasing treatment with solid-liquid ratio 6:1 for 6 h. Then the petroleum ether was filtered out. The primary defatted *C. esculentus* was subjected to secondary defatting. The solid-liquid ratio at this time was 4:1, and the time was 4 h. Then, the petroleum ether was filtered off, and the obtained secondary defatted *C. esculentus* was dried and processed by a wall breaker method again. The particle size was about 60-mesh, and the *C. esculentus* powder was obtained.

About 20 g of *C. esculentus* powder was taken in a 250-mL beaker and mixed with a certain proportion of NaOH solution. After covering with plastic wrap, the mixture was placed in an ultrasonic machine (40KHZ 240W, Keli Ultrasonic Cleaning Equipment Co., Ltd., Shenzhen, China) with a bath, and the required ultrasonic time and temperature adjusted. After sonication, *C. esculentus* solution was taken out and stirred at room temperature for 30 min using a magnetic stirrer, and then using a 50-mL centrifuge cup, the contents were centrifuged with speed of 4000 rpm for 15 min. After the first centrifugation, the sediment was stirred evenly, and 200-mesh polyester gauze was used for filtration. At this time, most of the starch had become filtered. However, the sediment still contained some proteins and lipids, so it needed to be washed three times by water, and the centrifugation conditions remained unchanged. The precipitate was dried in a drying oven at 40 °C for 24 h, and then crushed to 100-mesh size with a wall breaker to obtain *C. esculentus* starch. The equations used for calculating the purity and extraction yield of the starch were as follows:

$$\text{Starch purity (\%)} = \frac{S}{101.50} \times 100\% \quad (1)$$

$$\text{Extraction rate (\%)} = \frac{M_1 \times P_1}{M_2 \times P_2} \times 100\% \quad (2)$$

where S was the spin light value of sample (g), M_1 (g) and P_1 (%) was the quality and purity of *C. esculentus* starch, M_2 (g) and P_2 (%) was the quality and purity of *C. esculentus* powder.

Box-Behnken design (BBD)

The screening data revealed that four factors (ultrasound temperature; ultrasound time; liquid-solid ratio; and pH) significantly influenced the ultrasound-assisted extraction of *C. esculentus* starch. Response surface methodology (RSM) was utilized in this study. Specifically, the Box-Behnken experimental design was adopted. The variables considered for the experiment were ultrasound temperature (30, 40, and 50 °C), ultrasound time (30, 40, and 50 min), liquid-solid ratio (9:1, 12:1, and 15:1), and pH (8, 9, and 10). The Box-Behnken RSM experimental design used for the extraction yield of *C. esculentus* starch is shown in Table 1. From the analysis of the responses obtained from the experimental design, analysis of variance (ANOVA) was carried out to determine the significant variables for each response. A full quadratic model, with the form presented in Eq. 3, was used to develop the empirical models for the prediction of the extraction yield of *C. esculentus* starch,

$$Y = \beta_0 + \sum_{i=1}^n \beta_i \chi_i + \sum_{i=1}^n \beta_{ii} \chi_i^2 + \sum_{i=1}^{n-1} \sum_{j=i+1}^n \beta_{ij} \chi_i \chi_j \quad (3)$$

where Y is the predicted value obtained from the BBD response surface, β_0 , β_i , β_{ii} , and β_{ij} are the constant term, linear coefficient, quadratic term coefficient, and interaction term

coefficient, respectively. The χ_i and χ_j are the coded values (true values) of the experimental independent variables.

Scanning Electronic Microscopy (SEM)

Six kinds of starches from materials such as *C. esculentus*, sweet potato, potato, cassava, wheat, and corn, were selected. For capturing scanning electron micrographs (JSM-6490 SEM, JEOL, Peabody, MA, USA), the starch samples were sprinkled on a double-sided sticky tape embossed over a glass slide and carbon coated. The micrographs were taken at an acceleration voltage of 15 kV (Shrestha *et al.* 2012).

X-Ray Diffraction (XRD)

The above six starch samples selected were compactly packed within the sample port of a diffractometer (D8 Advance A25, Bruker Technology Co., Ltd., Beijing, China) and scanned over 2θ range of 2° to 40° using Cu anode for generating $K\alpha$ radiation with scanning speed of $10^\circ/\text{min}$ (Das *et al.* 2022).

Differential Scanning Calorimetry (DSC)

The degradation characteristics of the six different starches selected were determined *in vitro*, according to the method of Englyst *et al.* (1992) with a slight modification. The starch samples (200 mg; M_1 and M_2) and 15 mL of buffer solution were added to 250-mL conical flask and mixed well using a magnetic stirrer for 5 min. Then, the mixtures were kept in a boiling water bath for 20 min to fully gelatinize the starch, and then kept them in a water bath at 37°C . After equilibration at 37°C for 20 min, 10 mL of porcine pancreatic α -amylase (7500 U/mL) and amyloglucosidase (AMG) (300 U/mL) were added. The mixtures were then incubated in a water bath at 37°C with shaking (180 rpm). After 20 (G20) and 120 (G120) min of incubation, 20 mL of absolute alcohol was added to stop the enzymatic degradation (Zhang and Hamaker 2009). The glucose released in each enzymatic hydrolysate was determined with DNS (3,5-dinitrosalicylic acid) method. The rapidly digestible starch (RDS), slowly digestible starch (SDS), and RS percentage of each sample were calculated from the values of G120, and G20 as follows:

$$RDS / \% = \frac{G_{20} \times 0.9}{M_1} \times 100 \quad (4)$$

$$SDS / \% = \left(\frac{G_{120} \times 0.9}{M_2} - \frac{G_{20} \times 0.9}{M_1} \right) \times 100 \quad (5)$$

$$RS / \% = 1 - RDS / \% - SDS / \% \quad (6)$$

Statistical Analysis

The Design Expert 8.0 Software (Stat-Ease, Inc, Minneapolis, USA) was used for the statistical design of experiments and data analysis. All experiments were performed in triplicate, and the results were expressed as mean value \pm standard deviation (SD). The final results were evaluated using the statistical analysis software SPSS 22.0 (IBM Corp., Armonk, NY, USA). The significant levels were established at $p < 0.05$. Statistical analysis was performed using the software Origin 9.0 (Origin Lab Co., Northampton, MA, USA).

RESULTS AND DISCUSSION

Effect of Single Factor on the Extraction of *Cyperus esculentus* Starch

Effects of single factors on the extraction yield of *C. esculentus* starch are shown in Fig. 1. In the single factor experiment of the starch extraction, four factors were selected: ultrasonic temperature, ultrasonic time, liquid to solid ratio, and pH of alkaline liquor. When one factor was changed, the other three remained unchanged. As shown in Fig. 1A, the extraction yield of *C. esculentus* starch reached the maximum when the temperature was 40 °C, and then the extraction began to decrease. This might be because of the effect of alkaline liquor and protein was more sufficient due to the increase of temperature, but as the temperature continued to increase, the swelling degree of starch increased, resulting in insufficient extraction of starch (Ozturk *et al.* 2021). When the ultrasonic temperature was at 70 °C, the starch gelatinized, and it was unnecessary to measure the starch content at this time. When the ultrasonic temperature was at 40 °C, the extraction yield of *C. esculentus* starch was up to 88.0%. As shown in Fig. 1B, with the prolongation of ultrasonic time, the extraction yield also increased, but when the ultrasonic time reached 40 min, the extraction yield decreased with the increase of time. This might be because other components in the *C. esculentus* affected the extraction of starch due to the long ultrasonic time (Wang *et al.* 2021).

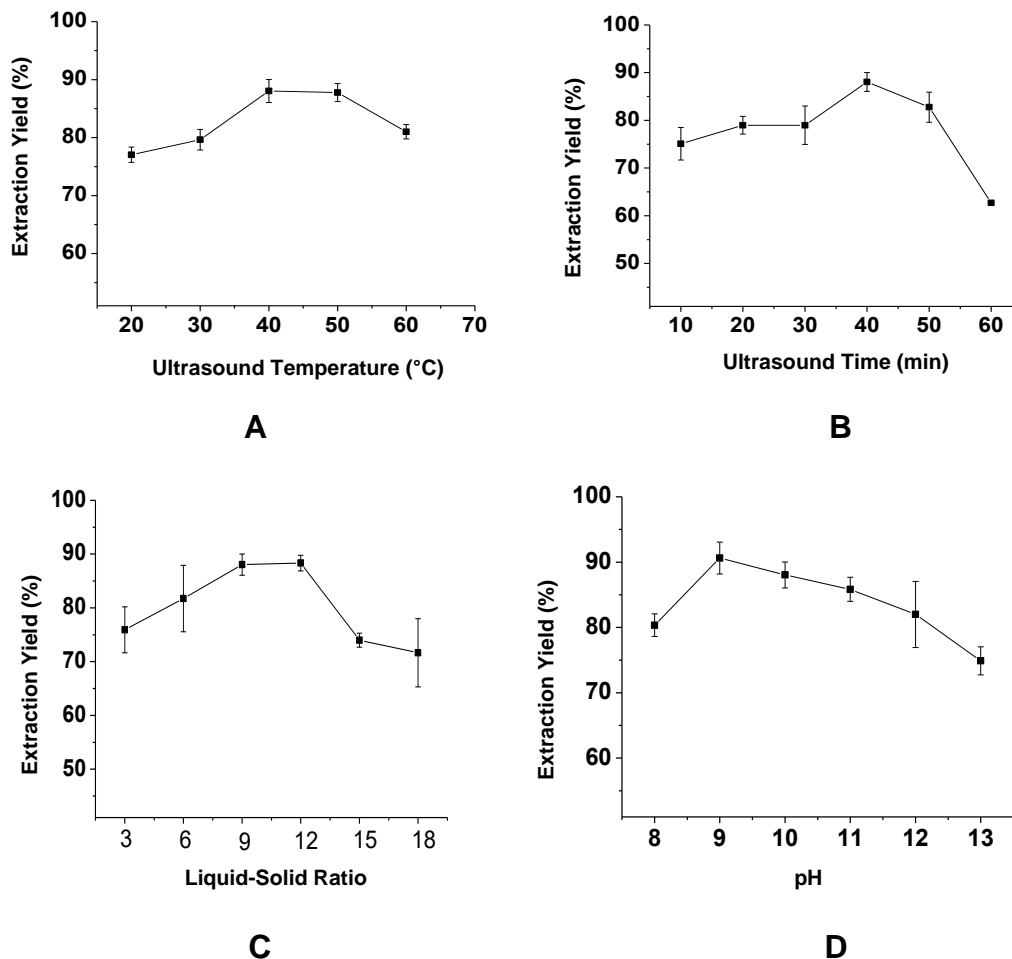


Fig. 1. Effect of single factor on the extraction yield of *Cyperus esculentus* starch: A: Ultrasound temperature; B: Ultrasound time; C: Liquid to solid ratio; and D: pH

As shown in Fig. 1C, with the increase of liquid, the extraction yield of starch showed a trend of first increasing and then decreasing, which might be because the binding force of starch and protein decreased under the action of water. When the liquid to solid ratio was 12:1, the maximum extraction yield of sesame bean starch was 88.3%. As shown in Fig. 1D, When the pH of the alkaline liquor was increased, the starch content of *C. esculentus* first increased and then decreased, which might be because the protein and the *C. esculentus* had a better effect with the increase of the alkaline liquor and pH, but when the protein content reached the optimal pH, the extraction decreased accordingly (Wang *et al.* 2021). When the pH was 9, the extraction yield reached the maximum value.

Extraction Optimization of *Cyperus esculentus* Starch

The effect of the ultrasound variables (ultrasound temperature, ultrasound time, liquid to solid ratio, and pH) on the extraction yield of *C. esculentus* starch was analyzed using response surface methodology (RSM). A four factor and three-level Box-Behnken design (Table 1) was applied to determine the main and interaction effects of the variables on the studied parameters. The F values of the main and interaction effects of the variables are presented in Table 2.

Table 1. Box-Behnken RSM Experimental Design

Exp. No.	Ultrasound Temperature (°C) (A)	Ultrasound Temperature (min) (B)	Liquid-solid Ratio (C)	pH (D)	Extraction yield (%)
1	50	50	12	9	81.85
2	40	40	12	9	83.40
3	30	40	9	9	84.90
4	40	50	12	10	75.48
5	30	40	12	8	88.08
6	40	40	9	8	80.35
7	40	40	12	9	84.82
8	40	30	9	9	86.21
9	40	40	9	10	88.04
10	30	40	15	9	88.03
11	40	30	15	9	84.19
12	30	40	12	10	85.93
13	40	50	15	9	86.53
14	30	30	12	9	86.40
15	50	40	9	9	82.61
16	40	40	12	9	84.72
17	40	50	12	8	87.30
18	40	40	12	9	86.89
19	40	40	12	9	83.09
20	50	40	12	8	76.55
21	40	40	15	8	79.90
22	30	50	12	9	85.71
23	50	40	15	9	79.62
24	40	30	12	10	90.43
25	50	40	12	10	79.47
26	50	30	12	9	87.61
27	40	50	9	9	84.21
28	40	40	15	10	73.99
29	40	30	12	8	72.90

Based on the experimental data (Table 2), a quadratic polynomial model expressed by Eq. 7 was created

$$Y = 84.58 - 2.61X_1 - 0.55X_2 - 1.17X_3 + 0.69X_4 - 1.27X_1X_2 - 1.53X_1X_3 + 1.27X_1X_4 + 1.09X_2X_3 - 7.34X_2X_4 - 3.40X_3X_4 + 0.37X_{12} + 0.63X_{22} - 0.65X_{32} - 3.17X_{42} \quad (7)$$

where Y is the estimated average extraction yield (%) of *C. esculentus* starch, X_1 (°C), X_2 (min), X_3 are the ultrasound temperature, ultrasound time, liquid-solid ratio, respectively, and X_4 is the pH value.

As shown in Table 2, based on the significance analysis, the model $P < 0.01$ indicated that the model was extremely significant, and the lack of fit item $P = 0.118 > 0.05$ indicated that the lack of fit was not significant. The coefficient of determination of the model was $R^2 = 0.8391$, and the signal-to-noise ratio = $8.376 > 4$, which indicated that the model had good fit and reliability and could be used to analyze and predict the extraction yield of *C. esculentus* starch. A smaller coefficient of variation (CV) of Y resulted in greater reliability of the test (Yuksel and Kayacier 2022). The CV value of this experiment was 3.06%, indicating that the reliability was high. As shown in Table 2, the interaction of BD was extremely significant, and the interaction of CD was significant.

Table 2. Box-Behnken RSM Experimental Design

Source of Variance	Degree of Freedom	Mean Square	F-value	P-value	Significance
Model	14	34.04	5.21	0.0019	**
A-Ultrasound Temperature (°C)	1	81.85	12.54	0.0033	**
B-Ultrasound Time (min)	1	3.7	0.57	0.4642	
C-Liquid-Solid Ratio	1	16.47	2.52	0.1345	
D-pH	1	5.69	0.87	0.3665	
AB	1	6.43	0.98	0.3379	
AC	1	9.36	1.43	0.2509	
AD	1	6.43	0.98	0.3379	
BC	1	4.71	0.72	0.4100	
BD	1	215.36	32.99	< 0.0001	**
CD	1	46.24	7.08	0.0186	*
A ²	1	0.91	0.14	0.7147	
B ²	1	2.59	0.4	0.5390	
C ²	1	2.73	0.42	0.5286	
D ²	1	65.11	9.97	0.0070	**
Error	14	6.53			
Lack-of-Fit	10	8.24	3.65	0.1118	
Pure error	4	2.26			
Total	28				

Through the analysis and optimization of Design-Expert 8.0 software, it was concluded that the optimal process parameters for the extraction of *C. esculentus* starch were: Ultrasonic temperature of 30 °C, ultrasonic time of 49.9 min, liquid-solid ratio of 14.99:1, and pH 8. Under these conditions, the extraction yield of *C. esculentus* starch was 93.8%, and the reliability was 0.918. In order to verify the accuracy of the prediction and consider the actual operation, the ultrasonic temperature in the above optimal parameters was adjusted to 30 °C, the ultrasonic time of 50 min, the liquid-solid ratio of 15:1, and pH

8. The experiment was repeated three times under this condition. The extraction yield of ultrasonic-assisted extraction of *C. esculentus* starch was 92.2%. Compared with the theoretical prediction value, the relative error was about 1.66%, which was within the acceptable range, therefore, this solution is feasible.

Microstructure Analysis of Different Starches

Granule characteristics of six different starches various sources, such as *C. esculentus*, sweet potato, potato, cassava, wheat, and dent corn, were observed by SEM (Fig. 2).

Figure 2 shows the scanning electron microscope images of six different starches. The results showed that A (sweet potato starch) was hemispherical, and the particle size is relatively large. Potato starch (B) was shown to be a tuber-shaped particle and its size was similar to sweet potato starch, and cassava starch (C) was identical to sweet potato starch in shape, but with medium particle size. However, wheat starch (D) particle size was different with the large particles that were flat. *Cyperus esculentus* starch (E) was ovoid of size about 2 to 15 μm , and the particle size was smaller among the six starches. The granule characteristics of *C. esculentus* starch were closer to those of tuber starch. The particle size of corn starch (F) was different from that of other starches, and it was irregular tuber-like. In general, the granule characteristics of *C. esculentus* starch and tuber starch were closer, and the particle size was a little smaller.

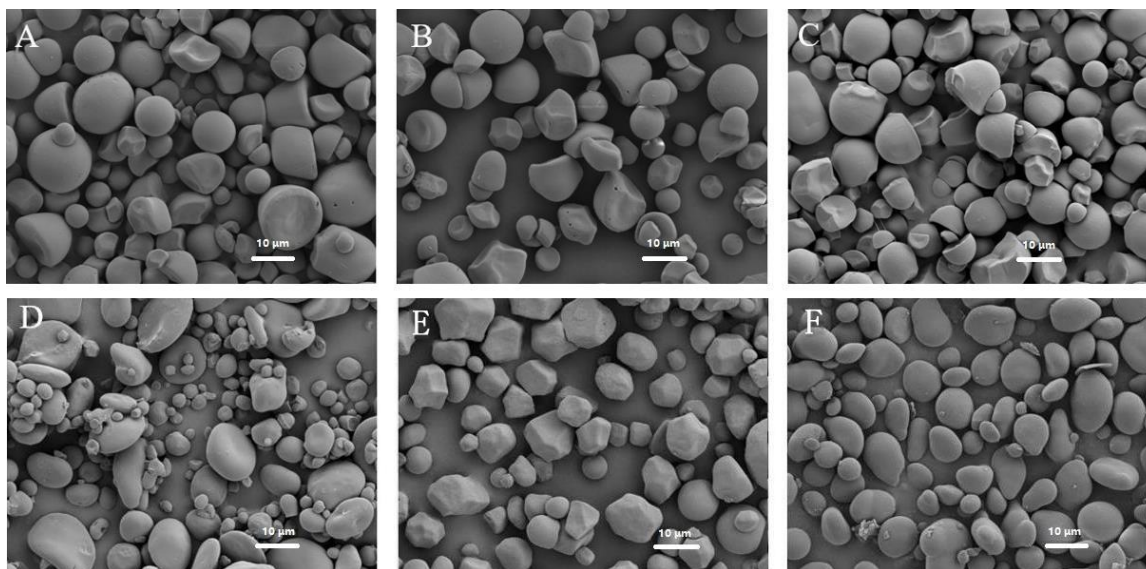


Fig. 2. SEM micrographs of different starches: A: Sweet potato; B: Potato; C: Cassava; D: Wheat; E: Dent corn; and F: *Cyperus esculentus*)

XRD Crystal Analysis of Different Starches

The XRD method was employed to assess and quantify the long-range crystalline order of starch. Various types of XRD patterns were obtained from various starch samples sourced from different botanical sources. For instance, type A, B, and C patterns are found in cereal, tuber, and legume starches, respectively (Rostamabadi *et al.* 2019). The presence of a peak in the regions of 15° and 22° and an intense double peak in the regions of 17° and 18° characterizes type-A starch (Dai *et al.* 2018). The presence of an intense peak in the region of 17° followed by less intense peaks in the regions of 15° , 20° , and 24°

represents type-B starches (Wang and Wang 2001). Therefore, as shown in Fig. 3, the XRD analysis of the five starches, including *Cyperus esculentus*, sweet potato, cassava, wheat, and corn had strong diffraction peaks at 15°, 17°, 18° and 22°, so they were of A-type starch. However, potato starch was B-type starch.

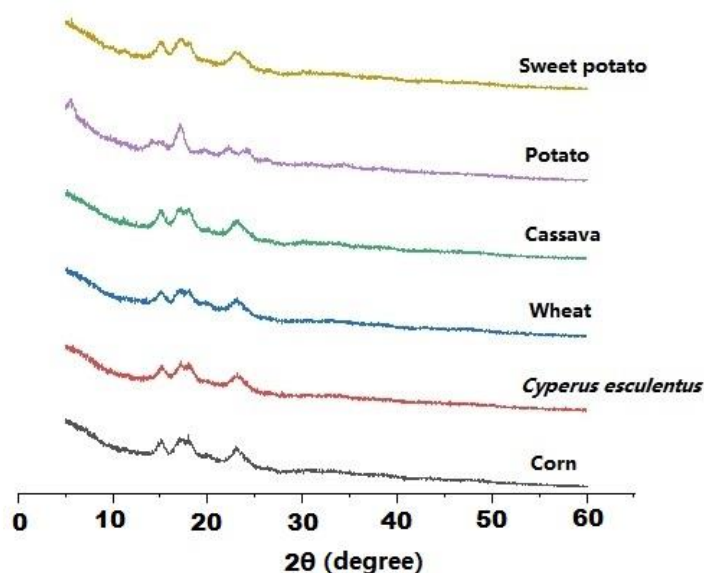


Fig. 3. XRD crystal structure of different starches

Thermal Properties of Different Starches

DSC parameters (onset temperature of gelatinization endotherm, peak temperature, conclusion temperature, transition temperature, and enthalpy change) of different starches are shown in Table 3. Starch granules are made up of a semi-crystalline structure. During gelatinization, this ordered structure was disrupted and melted, which results in uptake of water by amorphous and crystalline regions of starch and thus results in swelling of starch (Autio and Eliasson 2009). As shown in Table 3, the onset temperature of gelatinization endotherm and peak temperature of gelatinization of *C. esculentus* starch were only lower than those of sweet potato starch, and higher than other starches, which was 67.9 °C. Additionally, the internal arrangement of starch within various starch samples, the shape and size of the different starches, the distribution of amylopectin chains, lipid amylose complexes, and the amount of amylose may affect the thermal properties of the starch granules (Miao *et al.* 2010).

Table 3. Thermal Properties of Different Starches

Sample	T_o (°C)	T_p (°C)	T_e (°C)	ΔH (J/g)
<i>Cyperus esculentus</i>	67.86 ± 0.24 ^{ab}	72.13 ± 0.49 ^b	79.61 ± 0.02 ^b	-12.94 ± 1.17 ^b
Wheat	56.71 ± 0.49 ^d	61.28 ± 0.72 ^e	68.76 ± 0.78 ^d	-9.03 ± 0.05 ^a
Cassava	61.92 ± 0.47 ^c	68.10 ± 0.25 ^{cd}	79.79 ± 0.98 ^b	-12.68 ± 2.56 ^{ab}
Dent corn	64.84 ± 3.79 ^{bc}	69.57 ± 3.02 ^{bc}	80.59 ± 0.88 ^b	-14.40 ± 0.52 ^b
Sweet Potato	69.62 ± 0.88 ^a	75.36 ± 0.28 ^a	85.21 ± 0.95 ^a	-14.19 ± 0.81 ^b
Potato	60.88 ± 0.09 ^c	65.05 ± 0.22 ^d	73.18 ± 0.04 ^c	-14.12 ± 2.31 ^b

Means with different letters within the same row are significantly different ($p < 0.05$)

Degradation Characteristics of Different Starches

The degradation characteristics of different starches under the action of α -amylase and glucoamylase *in vitro* were analyzed. The starch *in vitro* hydrolysis can be divided into three types: RDS, SDS, and RS (Sajilata *et al.* 2006). It can be seen from Table 4 that among all groups, the content of RS (11.01%) in *C. esculentus* starch was the highest, while the content of RS (2.39%) in wheat starch was the lowest. The RS contents are influenced by the amorphous and ordered structures of starch granules (Bian *et al.* 2020). However, the content of RS in kiwi starch significantly increased and the content of RDS and SDS significantly reduced after different high-power ultrasound treatments (Wang *et al.* 2022). Meanwhile, the contents of RS in sweet potato and cassava starch were much higher than that of other starches (except *C. esculentus* starch). Therefore, *C. esculentus* and sweet potato starches can be used to prepare some specialty foods to satisfy people who need slow digestion.

Table 4. Degradation Characteristics of Different Starches

Sample	<i>Cyperus esculentus</i>	Wheat	Cassava	Dent corn	Sweet potato	Potato
RDS (%)	75.13 ± 0.37 ^b	81.42 ± 1.17 ^a	72.30 ± 1.19 ^c	83.60 ± 0.59 ^a	67.99 ± 0.27 ^d	76.28 ± 2.04 ^b
SDS (%)	13.87 ± 0.17 ^b	16.20 ± 0.28 ^{ab}	18.05 ± 0.72 ^{ab}	8.79 ± 0.29 ^c	22.06 ± 0.17 ^a	19.22 ± 0.49 ^{ab}
RS (%)	11.01 ± 0.20 ^a	2.39 ± 0.16 ^d	9.65 ± 0.99 ^a	7.62 ± 0.23 ^b	9.95 ± 0.17 ^a	4.51 ± 0.16 ^c

The rapidly digestible starch (RDS), slowly digestible starch (SDS), and resistant starch (RS) percentage each sample were calculated with different letters within the same row are significantly different ($p < 0.05$).

CONCLUSIONS

1. As an under-utilized resource, the starch from *Cyperus esculentus* can be regarded as a valuable source to develop into new functional food. Ultrasonic treatment can break the combination of starch, protein, and dietary fiber, especially in *C. esculentus*, which is rich in dietary fiber, and can better extract starch from *C. esculentus* tubers, thereby improving the extraction yield and shortening the extraction time.
2. The extraction yield of *C. esculentus* starch was 92.2% using ultrasound-assisted alkali method. The microstructure results showed that the granule characteristics of *C. esculentus* starch and tuber starch were closer. X-ray diffraction analysis showed that *C. esculentus* starch had an A-type crystal structure.
3. The onset temperature of gelatinization endotherm and peak temperature of gelatinization of *C. esculentus* starch were only lower than those of sweet potato starch, and higher than other starches, which is 67.9 °C. The content of RS (11.01%) in *C. esculentus* starch was the highest among the six starches.

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Conflict of Interest

The authors declare that there is no conflict of interest

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