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Physicochemical properties and starch digestibility of *Scirpus grossus* flour and starch

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ABSTRACT

Flour and starch isolated from the tubers of *Scirpus grossus* were investigated for their physicochemical properties and starch digestibility. The flour was extracted using two different processes namely peeled and unpeeled processes. Proximate analysis revealed that the flours from both processes contain considerably high total starch, more than 80%, which indicate their potential use as starchy foods. The amylose content of the flours and starches ranged from 29 to 32%. Starch granules of *S. grossus* were oval in shape with smooth surface and small diameters ranging from 6 to 15 μm. All samples exhibited high swelling pasting behaviors with pasting temperatures ranging from 78 to 79 °C, indicating the strong bonding forces within the granule interiors. Differential scanning calorimetry (DSC) results suggested that the samples gelatinized at temperatures ranging from 71 to 81 °C. In vitro starch digestion assay found that all samples provided the estimated glycaemic index (GI) values of approximately 55 or less.

1. Introduction

Wetlands are vital ecosystems which perform some important functions in relation to climate changes such as their ability to sink carbon, store and regulate water. The plants of wetland ecosystems played fascinating role in the life of human beings in earlier days as food, fodder, medicine, etc. But with the advancement of life pattern, the uses of wetland plants are foregone and they are treated as noxious weeds (Swapna, Prakashkumar, Anoop, Manju, & Rajith, 2011). Currently, with rising concerns on climate changes and food security, wetland plants have gained interest with particularly as food sources. The potentials of these plants for use as foods rely on their tuber and root starches. Recent researches have investigated structure and physicochemical properties of several underutilized tropical tuber and root starches (Hoover, 2001; Jayakody, Hoover, Liu, & Weber, 2005, Jayakody, Hoover, Liu, & Donner, 2007).

Scirpus grossus, is a wetland weed of the family Cyperaceae which are perennial grass-like plants and can grow to 3 m tall in shallow water or in moist soils. The most important reserve substance in the rhizome of Cyperaceae is starch, which accounts for 15% of fresh weight in winter. During the formation of new shoots in spring almost all the starch is mobilized (Steinmann & Brändle, 1984). Local people who make use of these rhizomes harvest them during winter. Like other tuber and root starches, many of the developing world's poorest and most food insecure households look to these crops as a contributing, if not the principle, source of food, nutrition and cash income. Among other things, farm households see the value of roots and tubers in their ability to produce edible energy and in their capability to generate yields under conditions where other crops may fail.

Among many species of the family Cyperaceae, *Cyperus rotundus* has received much attention. The plant is one of the most invasive weeds known, having spread out to a worldwide distribution in tropical and temperate regions. *C. rotundus* has been called "the world's worst weed" as it is known as a weed in over 90 countries and infests over 50 crops worldwide. On the other hand, it is a traditional herbal medicine used widely as analgesic, sedative, antispasmodic, antimalarial, stomach disorders and to relieve diarrhea (Zhu, Luk, Fung, & Luk, 1997). The tuber part of *C. rotundus* is one of the oldest known medicinal plants used for the treatment of dysmenorrhea and menstrual irregularities (Bhattarai, 1993). Infusion of this herb has been used in pain, fever, diarrhea, dysentery, an emmenagogue and other intestinal problems (Uddin, Mondal, Shilpi, & Rahnan, 2006). Umerie and Ezeuzo (2000) have reported that the *C. rotundus* starch is used in the food and confectionary industries. Phytochemical studies have shown that the major chemical components of this herb are essential oils, flavonoids, terpenoids, mono- and sesquiterpenes (Ohira

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et al., 1998; Kilani et al., 2005). Several investigators have reported its potential in antibacterial, antioxidant, cytotoxic and apoptotic activities (Ardestani & Yazdanparast, 2007; Kilani et al., 2008).

In the Cyperaceae family, *S. grossus* which is found extensively in South East Asia has not yet been investigated for its potential application. Though, local people extract its tuberous flour and use as foods. The yields are considerably high due to the large size of the tubers when compared to other species in the family (Fig. 1). This study investigated the physicochemical properties and starch digestibility of *S. grossus* flour and starch isolated from the tubers in order to find the potential as functional food source.

2. Materials and methods

2.1. Materials

S. grossus tubers were purchased from local markets in Phitsanulok Province, Thailand during winter of 2011.

2.2. Flour preparation

The tubers were brushed in tap water to remove adhering dirt. Flour was prepared by two different methods (peeled and unpeeled). These two processes represent the methods used by local people and industry. The peeled and unpeeled tubers (wet forms) were ground using a mortar. Distilled water was added at the ratio of 1:3 (sample:water) and the samples were ground using a blender until fine particles were obtained. The ground samples were sieved through a 100-mesh screen and rewashed with water for three times. The extracted flour was dried at 50 °C until the moisture content reached 10–13%. Notably that drying at 50 °C in this study cannot anneal starches in the samples as the water content is not sufficient, only excess water (more than 60%, w/w) can induce annealing process (Tester & Debon, 2000). The samples were sieved through a 100-mesh screen.

2.3. Starch extraction

Starch was isolated from the flour (unpeeled samples) using the alkaline extraction method (Lee, Htoon, & Paterson, 2007). The flour was dispersed in water (1:10, w/w) and pH was adjusted to 9 by adding 0.1 M NaOH, and then stored at 30 °C for 2 h. The slurry was filtered through a 100-mesh sieve. The filtrate was centrifuged at 3000 × *g* for 30 min. After centrifugation, the supernatant was discarded and the yellow layer (fat) was manually scraped off. The sediment or starch portion was washed with 0.01% sodium metabisulfite. Subsequently, it was washed three times with water and centrifuged at 3000 × *g* for 15 min. The starch portion was filtered again through a 100-mesh sieve and dried in a hot-air oven at 50 °C for 16 h. The dried starch samples were ground using a hammer mill fitted with a 0.5-mm sieve and sifted through 100 mesh sieve.

2.4. Physicochemical properties

2.4.1. Proximate analysis, total starch and amylose content

Proximate analysis was determined using standard AOAC methods (AOAC, 2000). Total starch was determined enzymatically using the total starch assay kit (Megazyme International, Ireland) following the standard AOAC Method 996.11. About 100 mg of sample was wetted with ethanol, mixed in KOH and sodium acetate buffer (pH 3.8). The samples were digested with thermostable α-amylase and amyloglucosidase and incubated at 50 °C for 30 min. The glucose released was determined using an enzymatic glucose reagent (GOPOD method), and the absorbance of the coloration was measured spectrophotometrically at 510 nm. For amylose, it was determined by colorimetric measurement of the blue amylose–iodine complex (Juliano, 1971). The samples were analyzed in triplicate.

2.4.2. Scanning electron microscope (SEM)

Dried samples were dispersed on double-stick adhesive tapes mounted on SEM aluminum stubs, coated with a thin layer of gold in a vacuum evaporator (EMITEX K 550X), and examined with the SEM (Phillips XL30) at 1000–1500 magnifications.

2.4.3. Swelling power and solubility

The solubility and swelling power were obtained using the method from Schoch (1964) with slight modifications. Samples (0.5 g) were dispersed in 15 mL distilled water. The suspensions were heated to 55, 65, 75, 85 °C in a water-bath with periodic mixing over a 30 min period. The cooked paste samples were centrifuged at 2200 rpm for 15 min. The supernatants were taken and placed in pre-weighed aluminum can before drying at 105 °C to gain constant weight. The dried supernatants were weighed as soon as the samples reached room temperature. After the supernatants were removed the swollen sediment samples were weighed. The solubility and swelling power were then calculated using Eqs. (1) and (2):

$$\text{Solubility}\,(\%) = \frac{\text{Weight of soluble matter in supernatant}\,(\text{g})}{\text{Weight of sample}\,(\text{g dry basis})} \times 100 \quad (1)$$

$$\text{Swelling power}\,(\%) = \frac{\text{Weight of swollen matter}\,(\text{g})}{\text{Weight of sample}\,(\text{g dry basis}) \times (100 - \text{solubility})} \times 100 \quad (2)$$

2.4.4. Pasting properties by Rapid Visco-Analyser (RVA)

Pasting properties were investigated using the Rapid Visco-Analyser (RVA-4D, Newport Scientific Pvt. Ltd., Australia) following the approved method 61.02 (AACC, 2009). A 13-min RVA profile was used with 3.0 g ground samples (adjusted to 14% moisture content) in 25 mL distilled water. The RVA Thermocline™ software (ver. 2.6) was used to obtain the RVA profiles and pasting characteristics. Each sample was analyzed in triplicate.

2.4.5. Differential scanning calorimetry (DSC)

Distilled water was added into the dried samples at the ratio of 3:1 (w/w). The DSC (Mettler Toledo DSC 1) equipped with a refrigerated cooler was used. The hydrated samples (20 ± 5 mg) were weighed into the aluminum DSC pans and hermetically sealed. An empty pan was used as the reference, and DSC analysis was done by scanning from 30 to 120 °C, ramping at 10 °C/min. Nitrogen was used as a purged gas. The resulting thermograms were analyzed using Mettler Toledo Star^e software (ver. 9.20) for the onset temperature (T_o), peak temperature (T_p), conclusion temperature (T_c) and transition enthalpy (ΔH). Each sample was analyzed in triplicate.

2.5. In vitro starch digestibility and modeling of starch digestogram

Time-course starch digestion in the samples was determined using a rapid in vitro digestibility assay based on glucometry (Mahasukhonthachat, Sopade, & Gidley, 2010; Sopade & Gidley, 2009). About 0.5 g of ground sample was treated with artificial saliva containing porcine α-amylase (Sigma A-3176 Type VI-B) before pepsin (Sigma P-6887; pH 2.0) was added and incubated at 37 °C for 30 min in a reciprocating water bath (85 rpm). The digesta was neutralized with NaOH before adjusting the pH to 6 (sodium