Original Article
Food Science and Technology

ISSN 0101-2061 (Print)
ISSN 1678-457X (Online)

DOI: https://doi.org/10.1590/fst.119121

# Cyanogenic compounds removal and characteristics of non- and pregelatinized traditional detoxified wild yam (*Dioscorea hispida*) tuber flour

Teti ESTIASIH[1]*, Dessy Eka KULIAHSARI[2], Erryana MARTATI[1], Kgs AHMADI[3]

**Abstract**

The presence of cyanide compounds restricts the utilization of wild yam (*Dioscorea hispida*) tubers for food or food ingredients. Traditional detoxification, usually used in wild yam chips processing, has not been evaluated for its effectiveness in reducing cyanogenic compounds. Processing into flour will increase wild yam tubers utilization and pregelatinization usually improve flour functional properties. This study aimed to evaluate the effect of 4 traditional detoxification methods and pregelatinization on cyanogenic compounds removal and wild yam tuber flour characteristics. The different methods were in rubbing ash, soaking time, and pregelatinization methods by boiling or steaming. The duration of a particular step was also different. The results showed that traditional detoxification methods reduced total cyanides 97%, cyanogenic glycosides 98-100%, acetone cyanohydrin 89-97%, and HCN 94-95%; also affected degree of cyanogenic compounds removal. Pregelatinization also reinforced the degree of cyanogen removal. Tuber flour physicochemical properties were affected by detoxification methods and pregelatinization. Modification of starch might occur due to the presence of $SiO_2$ and calcium in rubbing ash and affected functional properties of wild yam tuber flour. Starch granule morphology appeared not to be affected by detoxification methods. Traditional detoxification methods could be used to make a safe wild yam tuber flour.

**Keywords:** cyanogenic compounds; detoxification; functional properties; pregelatinization, starch.

**Practical Application:** Traditional detoxification successfully reduced cyanogenic compounds in wild yam tuber to the safe level.

## 1 Introduction

Wild yam is one of the tubers classified as a member of the *Dioscoreaceae* family and well-known as carbohydrates source. Yam tubers also contain some bioactive compounds, and the most recognized are dioscorin, diosgenin, and water-soluble polysaccharides. Dioscorin is a yam tuber storage protein, which is reported to have immunomodulatory activity (Liu et al., 2007), antioxidants (Han et al., 2013), improves metabolic syndrome of obese rats, and reduces systolic blood pressure (Shih et al., 2015). Diosgenin, a natural steroidal sapogenin, occurs abundantly in plants such as yams (Tohda et al., 2017). Dioscin, is a glycoside of diosgenin, in yam reaches 2.7% in yam. Meanwhile, diosgenin is about 0.004% for cultivated yams and 0.12-0.48% for wild yams. Diosgenin has been long used as raw material for steroidal drugs (Son et al., 2007). Diosgenin has many biological activities, such as anti-inflammatory, a potent antioxidant, anti-proliferative, hypoglycemic, and hypolipidemic (Tikhonova et al., 2014; Sethi et al., 2018). The ability to reduce cholesterol is related to cholesterol absorption inhibition and secretion (Son et al., 2007). Yam polysaccharide also reveals some biological activities, such as immunomodulatory, antioxidant, hypoglycemia, and antitumor (Huang et al., 2020).

However, the uses of wild yam tubers are limited due to the presence of toxic compounds in the form of cyanogenic, namely cyanogenic glycosides, acetone cyanohydrin, and cyanide acid (HCN) (Bradbury et al., 2011). Cyanogenic glycoside is a precursor of free cyanide, which might be hydrolyzed to acetone cyanohydrin with the assistance of β-glycosidase enzyme and then changed to free HCN (Panghal et al., 2019). Cyanide can bind oxygen in the blood so that it disrupts the respiratory system and causes symptoms of esophageal inflammation, dizziness, weakness, and seizures (Hendry-Hofer et al., 2019). Wild yam tubers contain HCN of 84.26 ppm (Kumoro et al., 2011) and total cyanide of 379-739 ppm (Saleha et al., 2018). The forms of cyanogenic compounds of wild yams have not been studied. Cyanide ingestion of 50-100 ppm is acute and leads to lethality (Bandna & Chand, 2012). Residual cyanogen causes neurological disorders and paralysis (Siritunga & Sayre, 2004; Kambale et al., 2017). The safe limit for cyanide in food products is 10 ppm (Burns et al., 2012).

Ferraro et al. (2016) reviewed the methods such as grating, grinding, and other tissue disruption techniques, which effectively reduce cyanides by facilitating cyanogenic compounds in vacuole to interact with β glucosidase in the cell wall, converting cyanogenic glucosides into HCN. HCN is easily removed by heating or solubilization. Some efforts to detoxify wild yam tubers have been studied limitedly, including leaching and

---

*Received 05 Dec., 2021*
*Accepted 06 Jan., 2022*

[1] *Department of Food Science and Technology, Universitas Brawijaya, Malang, Indonesia*
[2] *Master Program of Agricultural product Technology, Universitas Brawijaya, Malang, Indonesia*
[3] *Department of Agroindustrial Technology, Tribhuwana Tunggadewi University, Malang, Indonesia*
*Corresponding author: teties@ub.ac.id, teties@yahoo.co.id

steaming (Kumoro et al., 2011), boiling, roasting, and soaking in the flowing water (Ashri et al., 2014).

In Indonesia, traditional detoxification of wild yams are used to process wild yam tuber chips, including ash rubbing, pressing, and soaking, and then steaming/boiling and drying. Different locations of wild yam chip production have slightly different detoxification methods. Processing wild yams into flour as an intermediate ingredient could extend its uses. Traditional detoxification is expected to be used before wild yam flour preparation. The flour characteristics are supposed to be affected by the detoxification method. Therefore, the objective of this study was to evaluate the effect of the detoxification method on cyanogenic compounds removal and the physicochemical and functional properties of wild yam tuber flour.

One of the limitations of native flour is its low swelling and water-binding capacity that restricts its uses. Modification of flour is aimed to resolve such problems, one being the pregelatinization method (Dos Santos et al., 2019). During traditional wild yam detoxification in yam chips processing, the sliced wild yam tubers, after ash rubbing, pressing, and soaking, are steamed and then dried. Pregelatinization causes the starch granule structure to be damaged and thus increases solubility, swelling, viscosity, and water absorption (Sun et al., 2018). Pregelatinized flour is used as a thickener or filler and is also used as the main ingredient in several food products. The purpose of this study was also to determine the effect of pregelatinization on the properties of wild yam tuber flour.

## 2 Materials and methods

### *2.1 Materials*

Wild yam tubers were obtained from the teak forest in Nganjuk Regency, East Java, Indonesia. The ash for rubbing from wood and rice straw was prepared traditionally by burning them until the color of residue was greyish white. Crude salt was also used for rubbing. Chemicals for analysis were pure analytical grades obtained from Merck (Germany).

### *2.2 Traditional detoxification of wild yam tubers*

Detoxification of wild yam tubers followed four methods of traditional wild yam chips processing based on 4 locations: method 1 from Kandangan Sub-District, Kediri Regency, East Java; method 2 from Paiton Sub-District, Probolinggo Regency, East Java, method 3 from Wates Sub-District, Kediri Regency, East Java, and method 4 from Ngaglik Sub-District, Blitar Regency, East Java.

Detoxification of wild yam tubers was as follows: after peeling and washing, wild yam tubers were sliced using a traditional slicer to the thickness of ± 5 mm. The sliced tubers were rubbed by ash with or without salt. The ratios for each method were pure wood ash for method 1, 25:1 wood ash to salt for method 2, 50:1 wood ash to salt for method 3, and 5:1 mixture of wood ash and rice straw ash (2:1) to salt for method 4. The mineral compositions of the ash of each method were analyzed using X-Ray Fluorescence (XRF, PANalytical/Minipal 4). Traditionally, the amount of ash and salt for rubbing was sufficient to cover all of the surfaces of the sliced tubers. The rubbed sliced tubers were let to stand for 2 hours and then were put into gunny sacks. The rubbed sliced tubers were traditionally pressed using a big stone above the gunny sack. This process was conducted for 12 hours.

After pressing, the rubbed sliced tubers were drained for 4 hours. Then the sliced tubers were soaked for 36 hours (method 1), 12 hours (method 2), 48 hours (method 3), or 36 hours (method 4) with the soaking water changed with fresh water every 2 hours. For non-pregelatinized flour, after soaking, the sliced tubers were dried in a cabinet dryer for 4 hours at 60°C. The dried chips were ground and sieved 80 mesh. Pregelatinized wild yam tubers were prepared as usually conducted in traditional wild yam tuber chips processing. In method 1, the soaked sliced yam tubers were boiled for 30 min, boiled for 15 min (method 2), boiled for 15 min (method 3), or steamed for 30 min (method 4). The chips were then dried in a cabinet dryer for 4 hours at 60°C, ground, and sieved for 80 mesh. The differences of each traditional detoxification method are listed in Table 1.

### *2.3 Chemical analysis*

The cyanogenic compounds of fresh tuber and tuber flour of wild yams consisted of cyanogenic glucoside, acetone cyanohydrin, and cyanide acid (HCN) and each was analyzed according to the method of Bradbury et al. (1991). Proximate, starch, and amylose analysis of fresh tuber and tuber flour was according to AOAC (2011) methods. Amylose was measured spectrophotometrically by the iodine method, and starch was quantified by the acid hydrolysis method.

### *2.4 Physical analysis*

A diffractogram of the wild yam flour samples was obtained using an X-ray diffractometer (XRD, PANalytical X'Pert Pro) to

**Table 1.** The differences of 4 traditional detoxification methods of wild yam (*Dioscorea hispida*) tuber chips processing.

| Processing Step | Method 1 | Method 2 | Method 3 | Method 4 |
|---|---|---|---|---|
| Rubbing | Wood ash only | Wood ash: salt 25:1 (w/w) | Wood ash: salt 50:1 (w/w) | Wood ash: rice straw ash 2:1 (w/w) ash: salt 5:1 (w/w) |
| Pressing | 12 hours | 12 hours | 12 hours | 12 hours |
| Soaking | 36 hours | 12 hours | 48 hours | 36 hours |
| Pregelatinization | Boiling 30 min | Boiling 15 min | Boiling 15 min | Steaming 30 min |

Method 1: from Kandangan Sub-district, Kediri Regency; Method 2: from Paiton Sub-district, Probolinggo Regency Regency; Method 3: from Wates Sub-district, Kediri Regency; Method 4: from Ngaglik Sub-district, Blitar Regency.

determine starch diffraction patterns. The analysis conditions were 100 mA for current, Cu target, $2\theta = 4$-60° for the 2° angle, 40 kV for tube pressure, 0.02° for step length, and 6°/min for scanning speed.

### 2.5 Functional properties analysis

Water absorption capacity (WAC), oil absorption capacity (OAC), and swelling power (SP) were determined as described by Manupriya et al. (2020).

### 2.6 Starch granule morphology analysis

Granular morphology wild yam starch samples were observed using scanning electron microscopy (SEM, FEI type Inspect S50). The images were captured at a magnification of 5000.

### 2.7 Data analysis

This study used a completely randomized factorial design. The data were analyzed using a two-way analysis of variance by software Minitab 17. The Least Significant Difference was used to analyze the differences of the treatments.

## 3 Results and discussion

### 3.1 Traditional detoxification of wild yam (Dioscorea hispida) tuber

The predominant mineral in the rubbing ash for all detoxification methods is calcium, and potassium is the second most abundant mineral (Table 2). Other minerals are also found in ash for all methods, such as Ti, Si, Ba, Al, and P, with different concentrations. The ash in method 4 that uses rice straw ash for rubbing contains the highest $SiO_2$. The excessive ash used for rubbing the wild yam tuber slices is aimed to leach out the cell fluid that contains cyanogenic compounds due to different concentrations of minerals between inside and outside tissue. The cell fluid leaching is aggravated by subsequent pressing treatment after ash rubbing.

Cyanogenic glucoside is hydrolyzed by β glucosidase to glucose and acetone cyanohydrin. Furthermore, acetone cyanohydrin decomposes to acetone and hydrogen cyanide under neutral conditions (Bolarinwa et al., 2016), or due to the activity of hydroxy nitrile lyase (HNLase). Slicing before rubbing also disrupts the cell wall and enables the cyanogenic compounds degrading enzymes to contact their substrate. Although, the location of cyanogenic compounds and their degrading enzymes in wild yam tubers are still unknown. However, in many plants, the locations of cyanogenic compounds and their enzymes are different. Bolarinwa et al. (2016) reviewed that in young sorghum leaves. The cyanogenic compound (dhurrin) is located in vacuoles; meanwhile, their hydrolyzing enzyme is located in the cytoplasm. The leaf tissues of cassava are free of cyanide due to the separated locations of the enzyme and the substrates.

Ash rubbing the sliced wild yam tuber increases the pH value into alkaline. In alkaline pH, presumably endogenous wild yam tuber degrading enzymes such as β glucosidase and HNLase were activated. Some studies showed that optimum pH for β glucosidase activity varies depending on the enzyme sources, such as 5.0 for β glucosidase from almond and Thai rosewood and 6.5 from cassava (Svasti et al., 2003). Microbial β glucosidases have optimum activity at pH range of 4-8.5, from the plant at the pH of 5-7, and the animal at pH 5-6.5 (Singh et al., 2016). The β glucosidase activity from wild yam is still limitedly explored, and the optimum pH for its activity has not been studied. Traditionally, the ash rubbing in wild yam tuber detoxification is conducted in the beginning. Presumably, in this step, β glucosidase degrades cyanogenic glycosides. The leaching of cell liquid due to the cell disruption during slicing and subsequently pressing in the presence of ash also reduces the cyanogenic compounds from tuber tissue.

**Table 2.** Mineral composition (%) of rubbing ash for wild yam detoxification.

| Mineral (%) | Oxides | Rubbing Ash | | | |
|---|---|---|---|---|---|
| | | Method 1 | Method 2 | Method 3 | Method 4 |
| Ca | CaO | 82.10 | 78.14 | 61.60 | 58.70 |
| K | $K_2O$ | 8.67 | 6.85 | 16.70 | 8.04 |
| Nd | $Nd_2O_3$ | - | 6.40 | - | 10.00 |
| Si | $SiO_2$ | 4.84 | 4.54 | 8.19 | 18.70 |
| Mo | $MoO_3$ | - | - | 4.17 | - |
| Mn | MnO | - | - | 4.00 | - |
| P | $P_2O_5$ | 1.07 | 1.10 | 1.55 | 1.50 |
| Ba | BaO | 1.70 | 1.00 | 1.00 | 1.00 |
| Al | $Al_2O_3$ | - | - | 1.00 | - |
| Mg | MgO | - | - | 0.81 | - |
| S | $SO_3$ | 0.713 | - | - | - |
| Cr | $Cr_2O_3$ | - | 0.67 | - | 1.00 |
| As | $As_2O_3$ | 0.60 | - | - | - |
| Ti | $TiO_2$ | 0.31 | 0. 31 | 0.44 | 0.10 |
| V | $V_2O_5$ | - | - | 0.20 | - |

Method 1. 2. 3 used wood ash. method 4 used combination of wood ash and rice straw ash.

The soaking time of the pressed sliced wild yam tuber is different for each method. The soaking water is replaced by freshwater every 2 hours thus the microbial growth probability is low. This step is aimed to remove soluble cyanogenic compounds. The primary mechanism of cyanogenic compounds removal in this step is solubilization. Drying after soaking involves heating that evaporates cyanide acid from cyanogenic glucoside and cyanohydrin from the former steps.

### *3.2 Cyanogenic compounds of fresh wild yam (Dioscorea hispida) and tuber flour*

Wild yam tubers contain appreciable amounts of cyanogenic compounds of 83 ppm that exceed the maximum safe consumption limit (Table 3). Hargono et al. (2017) reported total cyanides of 176 ppm in wild yam tubers, meanwhile Kumoro et al., 2011 showed an HCN level of 46.30 ppm. Cyanogenic glycosides are the main cyanogen in wild yam tubers. Other species of yams contained cyanogen of 3-6 ppm (Bhandari & Kawabata, 2004). The difficulty of cyanogen removal from wild yam tubers is high amounts of cyanogenic glycosides. Free HCN is liberated from acetone cyanohydrins spontaneously or with the assistance of HNLase.

Traditional detoxification sharply reduced cyanogenic compounds in wild yam flour. Total cyanides achieved a safe level of consumption below 10 ppm, which ranged from 1.09 to 2.14 ppm. The degree of total cyanides removal for all treatments was 97%. Kumoro et al. (2011) reported that steaming for 2 hours could reduce HCN by 35.28%. Table 3 shows that different traditional detoxification methods revealed a similar degree of total cyanides removal. Bolarinwa et al. (2016) reviewed that cassava dough or pulp fermentation for 4–5 days decreased total cyanide by 52–63%. However, fermentation in cocoyam flour processing decreased cyanogen levels by 98.6%. Successive treatments in the traditional detoxification result in a sharp decrease of all cyanogenic compounds including reduce cyanogenic glycosides successfully. This compound is the primary constraint in wild yam tuber utilization because it is not easily removed by simple and single processing.

The degree of cyanides removal is affected by their compound types, and the highest removal was found in cyanogenic glycosides by about 99%. This compound is not easy to remove because of low volatility during heating. Ash rubbing, pressing, and soaking in traditional detoxification methods are supposed to reduce cyanogenic glycosides. The primary supposed mechanism of cyanogenic glycosides reduction is solubilization and the hydrolysis of cyanogenic glycosides by endogenous β glucosidase. Sufficient contact of this enzyme with its substrate is facilitated by slicing and pressing and ash rubbing also eases the liquid cell containing cyanogen to leach out. Among 4 methods, wild yam tuber flour of method 1 and pregelatinization showed the lowest concentration of cyanogenic glycosides. This method used wood ash only for rubbing, 36 hours of soaking, and 30 min of boiling. Other methods used a combination of ash and salt for rubbing. Perhaps the presence of more minerals, as shown in Table 2 leads to more liquid cells leaching out, thus sharply reducing cyanogenic glycosides. The pressing time of 4 methods was similar, and while pressing, the β glucosidase might react with its substrate to hydrolyze cyanogenic glycosides into acetone cyanohydrin.

The acetone cyanohydrin removal for all methods is about 89-97%. The occurrence of acetone cyanohydrin in fresh wild yam tubers is the lowest among cyanogenic compounds. This intermediate product is naturally found in a low level because of the restricted contact of β glucosidase in the cell wall with its substrate cyanogenic glycosides in vacuoles. It is supposed that acetone cyanohydrin is formed in a large amount during detoxification, possibly in the ash rubbing and pressing steps. Slicing and pressing let the enzyme contact with cyanogenic glycosides substrate. However, heat treatment by boiling/steaming and drying in the next steps might decompose acetone cyanohydrin into acetone and hydrogen cyanide (HCN). Therefore, this cyanogenic compound was found in low amounts. The method 1 and pregelatinization revealed the highest acetone cyanohydrin removal degree in the tuber flour. The lowest removal degree was found in the tuber flour from methods 3 and 4 and non-pregelatinization. Both methods used longer soaking time than methods 1 and 2 but revealed a slightly higher amount of acetone cyanohydrin. Presumably, this difference was related to the rubbing ash used. Longer soaking time resulted in lower acetone cyanohydrin levels due to longer

**Table 3.** Cyanogenic compounds of wild yam (*Dioscorea hispida*) fresh tuber and flour.

| Detoxification | Treatment | Cyanogenic Glycosides | | Acetone Cyanohydrin | | Free HCN | | Total Cyanides | |
|---|---|---|---|---|---|---|---|---|---|
| | | Concentration (ppm) | Removal (%) | Concentration (ppm) | Removal (%) | Concentration (ppm) | Removal (%) | Concentration (ppm) | Removal (%) |
| Method 1 | Non-pregelatinization | 0.45 ± 0.03$^c$ | 99.18 | 0.11 ± 0.01a | 97.19 | 1.33 ± 0.07 | 94.58 | 1.89 ± 0.42 | 97.73 |
| | Pregelatinization | 0.34 ± 0.05$^e$ | 99.38 | 0.10 ± 0.05a | 97.44 | 1.32 ± 0.04 | 94.62 | 1.76 ± 0.28 | 97.88 |
| Method 2 | Non-pregelatinization | 0.49 ± 0.01$^b$ | 99.10 | 0.14 ± 0.09a | 96.42 | 1.34 ± 0.09 | 94.54 | 1.97 ± 0.16 | 97.63 |
| | Pregelatinization | 0.45 ± 0.02$^c$ | 99.18 | 0.13 ± 0.04a | 96.68 | 1.33 ± 0.04 | 94.58 | 1.95 ± 0.16 | 97.66 |
| Method 3 | Non-pregelatinization | 0.56 ± 0.02$^d$ | 98.98 | 0.41 ± 0.05b | 89.51 | 1.37 ± 0.02 | 94.42 | 2.34 ± 0.54 | 97.19 |
| | Pregelatinization | 0.49 ± 0.03$^b$ | 99.10 | 0.13 ± 0.03a | 96.68 | 1.35 ± 0.02 | 94.50 | 1.97 ± 0.09 | 97.63 |
| Method 4 | Non-pregelatinization | 0.56 ± 0.02$^a$ | 98.98 | 0.41 ± 0.05b | 89.51 | 1.37 ± 0.02 | 94.42 | 2.34 ± 0.32 | 97.19 |
| | Pregelatinization | 0.53 ± 0.09$^e$ | 99.03 | 0.36 ± 0.06b | 90.79 | 1.36 ± 0.06 | 94.46 | 2.25 ± 0.42 | 97.30 |
| Fresh yam tuber* | | 54.74 ± 0.17 | | 3.91 ± 0.28 | | 24.55 ± 0.21 | | 83.20 ± 0.11 | |

The value with different notation means significantly different at p > 0.05. * wet basis solubilization. Kumoro et al. (2011) reported that soaking time affected HCN removal in wild yam.

HCN removal degree in the wild yam tuber flour for all methods was 94-95%. Free HCN was also found in the fresh wild yam tuber, although its level was lower than cyanogenic glycosides. Traditional detoxification methods and non- or pregelatinization showed insignificant differences. Nevertheless, the highest HCN removal degree was found in method 1 with pregelatinization. Pregelatinization for all methods showed a lower HCN level than non-pregelatinization. Additional heating by boiling or steaming caused more HCN to evaporate. Boiling is better than steaming to remove HCN, and a longer boiling time also showed better HCN removal.

### *3.3 Physicochemical characteristics of wild yam (Dioscorea hispida) tuber flour*

Fresh yam tubers are a good source of carbohydrates, with low protein and fat content (Table 4). The composition of fresh wild yam tubers is similar to that reported by Padhan et al. (2020). The traditional detoxification methods affected the approximate composition of the flour. A sharp increase of ash was found in the wild yam tuber flour from 0.69% into about 1.05-4.50% (db). The ash from the rubbing step in detoxification might be absorbed into the tuber tissue during rubbing and pressing. This residual ash was found in the final tuber flour product, although most ash is removed during soaking and in some methods also during boiling. The highest ash content was found in the flour from method 4, and the least was found in method 1. Method 4 used a combination of wood and rice straw ash and a combination of ash and salt with ash in the highest portion compared to other methods (except method 1 that only used wood ash). Soaking time did not appear to affect the ash content, indicated by method 4 with the longest soaking time but had the highest ash content. The treatment after detoxification by steaming or boiling did not significantly affect ($p < 0.05$) ash content. Generally, pregelatinized flour had lower ash content except for pregelatinized flour from method 4. Method 1 to 3 used boiling for pregelatinization, meanwhile, method 4 used steaming. During boiling, some absorbed ash from ash rubbing might leach out, thus decreasing the ash content. In steaming, the leaching process was more restricted than boiling.

Detoxification method and pregelatinization significantly affected the protein, fat, and carbohydrate of wild yam tuber flour (Table 4). The changes of proximate composition related to the solubilization of nutritional components due to liquid leaching during ash rubbing, pressing, and soaking. Interestingly, the changes of carbohydrate and starch are most pronounced among components. Pregelatinization generally reduces nutritional components because of component leaching during boiling or steaming after detoxification.

Amylose of tuber flour decreased sharply compared to the fresh wild yam tuber that contains amylose of 1.83%. The degree of amylose leaching was about 63-74%. Some amylose leached out during detoxification and the amylose content in the final wild yam tuber flour was only about 0.47-0.68% (db). Method 1 had the lowest starch content for non- and pregelatinized tuber flour. This method involved 36 hours of soaking, similar to method 3. However, the ash used was only wood ash. The least amylose leaching was observed in method 4. Table 2 shows that rubbing ash of method 1 had the least $SiO_2$, which might severely reduce amylose due to excessive leaching. $SiO_2$ decreased weight loss (Abbasi, 2012) by forming a network structure of starch (Tang et al., 2008). Tattiyakul et al. (2012) reported low solubility and low amylose leaching of wild yam starch (9.92 ± 0.4g/100 g and 15.8 ± 0.6 g/100 g, respectively) measured by heating at 90 °C for 30 min. Traditional detoxification methods involved soaking for 12-48 hours and pregelatinization by boiling or steaming for 15-30 min. Therefore, excessive amylose leaching occurred that lowered the amylose content of tuber flour.

Contrary to amylose content, the lowest starch content was observed at method 4 and the highest at method 1. The predominant starch of wild yam tuber is amylopectin. Amylopectin was greatly responsible for the solubility of starch during detoxification in soaking, pressing, and boiling. The calcium concentration in rubbing ash was different among the methods, and the highest concentration was found in method 1, and the least was method 4. According to Yang et al. (2014), calcium-modified amylopectin behavior prevents hydration, and induces a stable and compact network of amylopectin and whey protein (Liu et al., 2007). Pregelatinized flour showed lower starch content than non-pregelatinized for all methods, because more starch leaching occurred during boiling or steaming.

**Table 4**. Chemical characteristics of wild yam (*Dioscorea hispida*) fresh tuber and flour.

| Detoxification | Treatment | Proximate (% db) | | | | | Starch (% db) | Amylose (% db) |
|---|---|---|---|---|---|---|---|---|
| | | Ash | Moisture | Fat | Protein | Carbohydrate | | |
| Method 1 | Non-pregelatinization | 1.88 ± 0.56b | 11.52 ± 0.26d | 1.69 ± 0.01d | 1.92 ± 0.34d | 85.47 ± 0.12d | 57.82 ± 0.19a | 0.49 ± 0.06b |
| | Pregelatinization | 1.05 ± 0.13a | 9.04 ± 0.15a | 1.08 ± 0.02c | 2.10 ± 0.17bc | 83.14 ± 0.28b | 51.00 ± 0.51b | 0.47 ± 0.02a |
| Method 2 | Non-pregelatinization | 2.16 ± 0.20b | 10.75 ± 0.13c | 1.69 ± 0.04d | 1.22 ± 0.13e | 86.46 ± 0.19e | 36.91 ± 0.11c | 0.53 ± 0.03c |
| | Pregelatinization | 2.04 ± 0.26b | 9.63 ± 0.17b | 1.08 ± 0.14c | 2.08 ± 0.11c | 84.05 ± 0.24c | 29.97 ± 0.25d | 0.47 ± 0.04a |
| Method 3 | Non-pregelatinization | 2.41 ± 0.78c | 10.79 ± 0.19c | 1.05 ± 0.09c | 2.89 ± 0.82b | 82.86 ± 0.43b | 26.24 ± 0.62e | 0.65 ± 0.16f |
| | Pregelatinization | 2.36 ± 0.07bc | 9.26 ± 0.33ab | 1.05 ± 0.12c | 2.10 ± 0.53c | 85.23 ± 0.76 | 25.25 ± 0.71e | 0.55 ± 0.09d |
| Method 4 | Non-pregelatinization | 4.29 ± 0.09d | 11.37 ± 0.11d | 0.87 ± 0.02b | 4.63 ± 0.23a | 78.84 ± 0.32a | 25.98 ± 0.29e | 0.68 ± 0.05g |
| | Pregelatinization | 4.50 ± 0.51d | 9.04 ± 0.15a | 0.24 ± 0.00a | 2.45 ± 0.17b | 84.06 ± 0.24c | 23.36 ± 0.56f | 0.58 ± 0.03e |
| Fresh yam tuber* | | 0.69 ± 0.08 | 72.21 ± 0.54 | 1.95 ± 0.13 | 5.03 ± 0.42 | 20.12 ± 0.35 | 12.76 ± 0.36 | 1.83 ± 0.05 |

The value with different notation means significantly different at $p > 0.05$. * wet basis

The crystallinity of wild yam tuber flour was shown in the diffractogram of Figure 1. All treatments showed a similar diffraction pattern. This diffraction pattern was characterized by a narrow peak between 0-30° and then sloping after 30°. All the flour had strong reflection at about 5°, 17°, 20°, 22°, and 24°, which is a typical β-type crystalline pattern (Zhang et al., 2018). The β-type crystalline structure is typical of high-amylose starches contained in tubers (Dome et al., 2020). The strong reflections were observed at a diffraction angle (2θ) of 17.03° and 17.23° for method 1, non- and pregelatinized tuber flour; 16.99° and 22.07° for method 2, non- and pregelatinized tuber flour; 29.35° and 29.37° for method 3, non and pregelatinized tuber flour; 17.25° and 16.89° for method 4, non and pregelatinized tuber flour. The sloping pattern after 30° indicated that all wild yam tuber flour had amorphous regions.

Pregelatinization shifted the diffraction pattern indicating the changes of semicrystalline structure into amorphous, although some semicrystalline starch structure might remain. All pregelatinized tuber flours had lower diffraction peak intensity than non-pregelatinized for all detoxification methods. According to Wang et al. (2019), pregelatinization reduced starch crystallinity indicated by decreasing diffraction peak intensity due to less structural crystallite. The changes of crystallin into amorphous occurred because of amylopectin and helical structure disruption (Dar et al., 2018). Water absorption during steaming or boiling swelled the starch granule and loosed crystallinity (Bertolini, 2010). Traditional detoxification of method 4 combined with pregelatinization showed the lowest diffraction peak intensity. This treatment involved 30 min steaming, and the crystallinity was almost similar to method 1 with 30 min boiling for gelatinization. Long heating time by boiling or steaming gave sufficient time for the hydrogen bonds among the starch chains to disrupt and loosen the crystallinity.

### *3.4 Functional properties of wild yam (Dioscorea hispida) tuber flour*

Traditional detoxification methods significantly ($p > 0.05$) affected water absorption capacity (Table 5). Among the methods, method 4 showed the lowest water absorption capacity both for non- and pregelatinized tuber flour. The highest water absorption capacity was observed at method 1. The presence of $SiO_2$ in the rubbing ash was responsible for the compactness of the flour (Tang et al., 2008), thus decreasing water uptake of the flour. The calcium concentration in rubbing ash was the highest among other minerals, but the percentage was different for each detoxification method. The lowest calcium was found in the rubbing ash of method 4 with the lowest water absorption capacity. According to Hedayati et al. (2020), $CaCl_2$ enhances water absorption of starch. The inclusion of $CaCl_2$ led to an interconnected network and increased the linkage within the starch matrix. $CaCl_2$ formed electrostatic interactions with water molecules. Pregelatinization increased water absorption capacity for all methods. According to Patindol et al. (2013), pregelatinized starch granules were completely disintegrated and water uptake increased.

The phenomena observed for water absorption capacity were also found in oil absorption capacity. The oil absorption capacity of the flour from method 4 was the lowest and the highest in method 1. The presence of $SiO_2$ modified the starch, and modification intensity depended on the concentration of $SiO_2$ in the rubbing ash. A more compact granular structure, because of the interaction of $SiO_2$ with starch, restricted the oil absorption. Thus, the highest oil absorption capacity was found in the flour with method 1 with the lowest $SiO_2$ concentration. Pregelatinized flour showed higher oil absorption capacity than non-pregelatinized. Patindol et al. (2013) also observed the increase of oil uptake due to pregelatinization. Disintegration due to heating and a more open granular structure made it easier for oil to be absorbed. Denaturation of protein in the flour due to heating also enhanced oil absorption because of exposure to inner hydrophobic protein regions during denaturation.

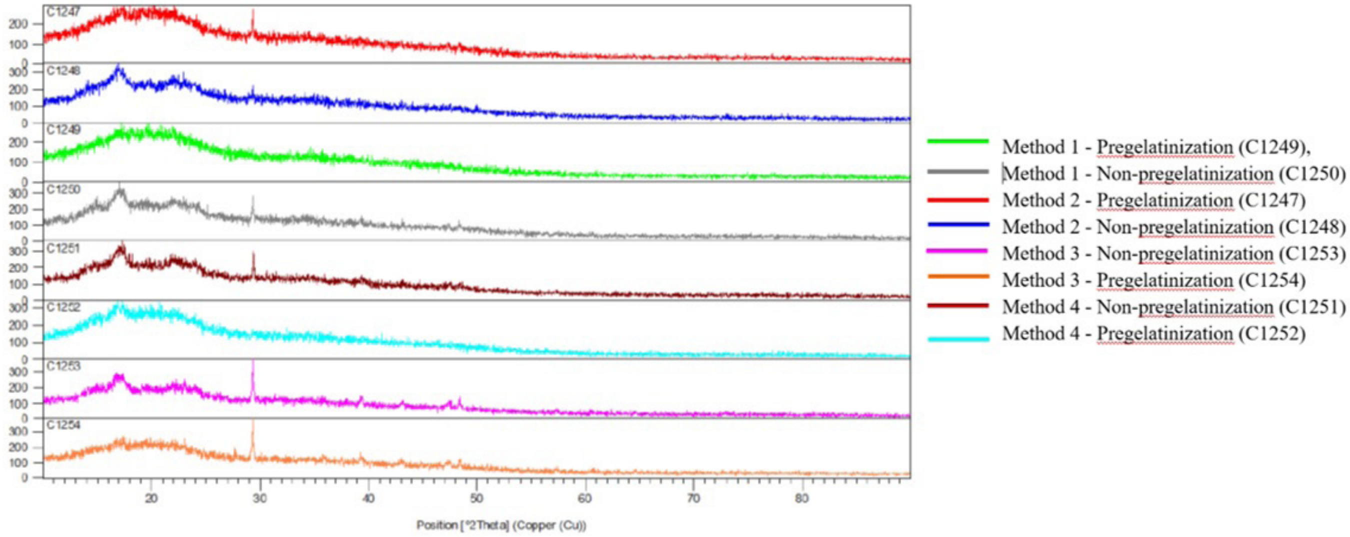

**Figure 1.** X-ray diffractogram of traditional detoxified wild yam (*Dioscorea hispida*) tuber flour.

Swelling power indicated the ability of the starch granule stability during heating in excess water (Waterschoot et al., 2016). The effect of the detoxification method and pregelatinization on swelling power was similar to water and oil absorption capacity. The presence of $SiO_2$ from rubbing ash seemed to affect the swelling power, which related to the modification of the starch granule structure to be more compact. The formed network starch due to the presence of $SiO_2$ restricted the starch granules to absorb water and swell. Pregelatinization increased the swelling power of wild yam tuber flour in all detoxification methods. According to Li & Yeh (2001), amylopectin was responsible for swelling power. The high proportion of long-chain molecules in amylopectin contributes to swelling power. Data in Table 5 shows that starch content of flour decreased in which these starches mainly consisted of amylopectin. Thus, swelling power also decreased in the same manner.

The solubility of tuber flour was affected by the detoxification methods and pregelatinization. Flour detoxified by method 4 revealed the lowest solubility, and the highest was observed in method 1. The solubility is also affected by the composition of minerals in rubbing ash. The higher the $SiO_2$ and the lower Ca content resulted in a lower solubility. As previously discussed, $SiO_2$ induced a network formation that resisted to water (Tang et al., 2008), while Ca increased water absorption due to the formation of more open granular structures (Hedayati et al., 2020). Pregelatinized tuber flour showed better solubility than non-pregelatinized. Zhang et al. (2018) reported that swelling power and water solubility of *Dioscorea opposita* starch gradually increased with increasing heating temperature. The hydration of starch granules reflected the magnitude of interaction mainly between amorphous domains with water, and amylose content affected the extent of this interaction.

### 3.5 Starch granule structure

Figure 2 showed the morphological structure of wild yam tuber starch granules from different detoxification methods

**Table 5.** Functional properties of wild yam (*Dioscorea hispida*) tuber flour.

| Method of Detoxification | Treatment | Water Absorption Capacity (g/g) | Oils Absorption Capacity (g/g) | Swelling Power (%) | Solubility (%) |
|---|---|---|---|---|---|
| Method 1 | Non-pregelatinization | 1.91 ± 0.15b | 1.53 ± 0.02c | 12.28 ± 0.09g | 9.23 ± 0.03d |
| | Pregelatinization | 5.72 ± 0.50e | 1.88 ± 0.24d | 13.93 ± 0.05h | 29.00 ± 0.03g |
| Method 2 | Non-pregelatinization | 1.76 ± 0.10b | 1.36 ± 0.10ab | 11.24 ± 0.08e | 6.97 ± 0.04c |
| | Pregelatinization | 4.59 ± 0.08d | 1.51 ± 0.12c | 12.01 ± 0.09f | 27.50 ± 0.08 |
| Method 3 | Non-pregelatinization | 1.71 ± 0.12b | 1.34 ± 0.35abc | 10.17 ± 0.07c | 6.33 ± 0.02b |
| | Pregelatinization | 4.24 ± 0.37d | 1.44 ± 0.04b | 10.80 ± 0.06d | 13.83 ± 0.03f |
| Method 4 | Non-pregelatinization | 1.51 ± 0.07a | 1.21 ± 0.14a | 9.51 ± 0.06a | 3.43 ± 0.01a |
| | Pregelatinization | 3.74 ± 0.11c | 1.23 ± 0.21a | 9.98 ± 0.04b | 12.60 ± 0.05e |

The value with different notation means significantly different at $p > 0.05$.

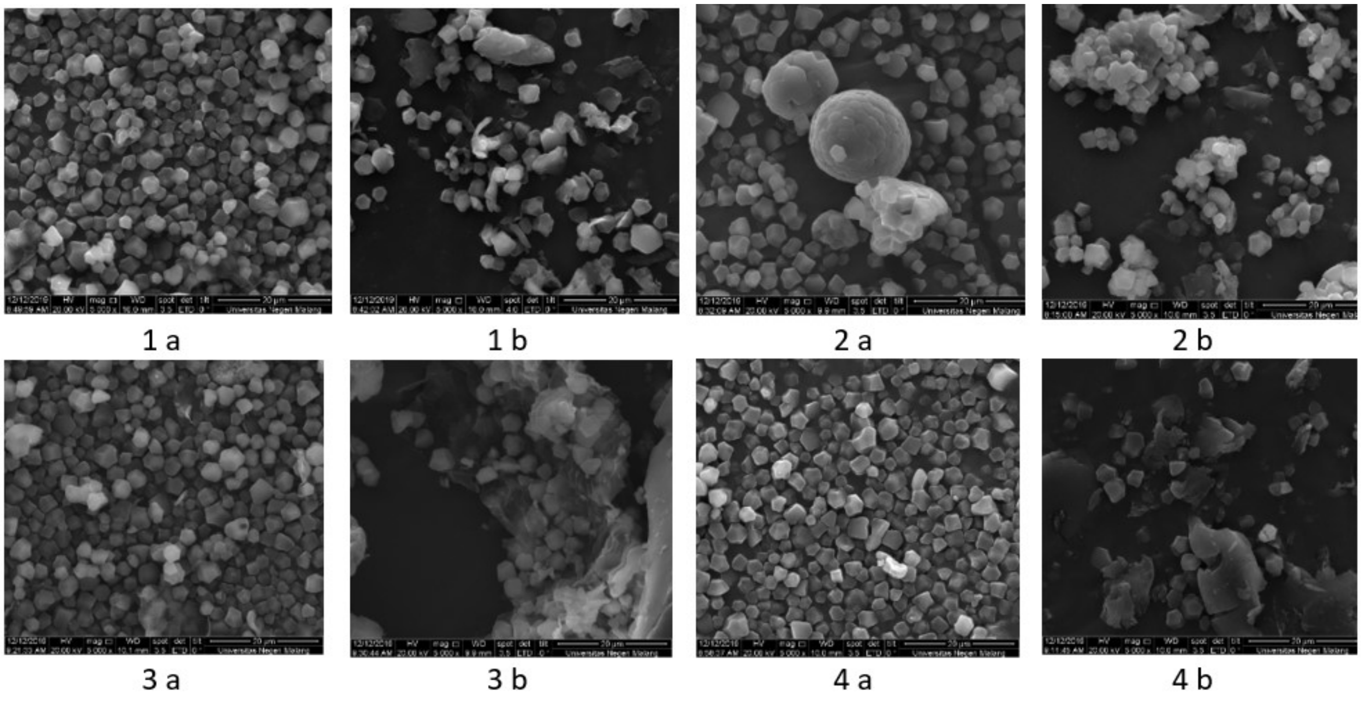

**Figure 2.** Microstructure of wild yam tuber flour from different detoxification methods and pregelatinization (1) Method 1 (a) non pregelatinization, (b) pregelatinization; (2) Method 2 (a) non pregelatinization, (b) pregelatinization; (3) Method 3 (a) non pregelatinization, (b) pregelatinization; (4) Method 4 (a) non pregelatinization, (b) pregelatinization.

and pregelatinization. Starch granules of wild yam tuber had polyhedral starch granules. A similar finding was reported by Tattiyakul et al. (2012) and Ashri et al. (2014) that the native starch granules are polyhedral and have a smooth surface. The starch granules remained intact after the modification by hydrothermal (Tattiyakul et al., 2012). The detoxification methods did not appear to have affected starch granular morphology. Before gelatinization, the detoxification steps were ash rubbing, pressing, and soaking, which did not involve heating. The rubbing by ash and pressing leached out the liquid from the cell, and some starch might also leach out to some extent. However, some water might also be absorbed during soaking, enlarging the size of starch granules. The longest soaking time was in method 3 that revealed a bigger starch granule size than other methods. Method 2 had the shortest soaking time, and some starch granules appeared lumpy. This lumpiness is presumably related to the role of minerals to make a bridge between starch granules. Li et al. (2016) reported the coaggregation of mineral filler particles and starch granules.

Pregelatinization, in all detoxification methods, partially damaged starch granules, both by boiling or steaming. After gelatinization, many starch granules remained intact, although some agglomeration occurred. Aichayawanich et al. (2011) reported that the rubbery phase of starch was strongly agglomerated into large particles. The agglomeration mechanism may be liquid bridge formation. Therefore, method 3 that involved soaking 48 hours and 15 min boiling, had the greater lumpiness. During soaking, water was sufficient to make liquid bridges, and these bridges were still intact because the boiling time was only 15 min. The liquid bridge ruptured during heating due to thermal energy for dissociation. Compared to the method 3, soaking 12 hours followed by 15 min boiling revealed lower lumping. Methods 1 and 4 involved different heating methods and a similar soaking time of 36 hours. The lumpiness was more apparent in method 4 because the energy intensity to rupture agglomeration in steaming (method 4) was lower than in boiling (method 1).

### *3.6 Practical application*

This study reveals that traditional detoxification methods could be used for pre-treatment in wild yam tuber flour preparation. These methods successfully reduce cyanogenic compounds to a safe level for consumption. Traditional detoxification pre-treatment could remove cyanogenic glycosides as the most difficult compound to reduce. Using these methods in wild yam tuber flour preparation would increase and widen the utilization of wild yam tubers. Flour is an intermediate product and an ingredient for many food products. Traditional detoxification method 1 is the best for reducing cyanogenic compounds.

The physicochemical characteristics of wild yam tuber flour, after traditionally detoxifying, reveal some properties such as water and oil absorption, solubility, and swelling power which are important parameters for food processing. This study showed that traditional detoxification method 1 had the highest water and oil absorption, solubility, and swelling power. Gelatinization in traditional detoxification improved wild yam tuber flour functional properties. Pregelatinized wild yam tuber flour is suitable for food products that require good water absorption and solubility, such as instant products, and this tuber flour might also be suitable as a food thickener. Further study is required to investigate the utilization of detoxified wild yam tuber flour in many food products.

## 4 Conclusions

Traditional detoxification of wild yam tubers successfully removed total cyanogenic compounds by about 97%. Different methods revealed similar degrees of total cyanides removal. The degree of removal was the highest for cyanogenic glycoside by about 98-99%. Pregelatinization for all methods showed lower cyanogenic compound levels than non-pregelatinization. The methods affected the approximate composition of the flour due to leaching during ash rubbing, pressing, soaking, and boiling/steaming. Minerals in the rubbing ash modified starch in the flour, thereby influencing flour characteristics. Traditional detoxification methods significantly affected the physical and functional properties of tuber flour. The interaction of starch with $SiO_2$ and calcium modified functional properties. Higher $SiO_2$ and lower calcium concentration in rubbing ash reduced functional properties. Wild yam tubers had polyhedral starch granules that were still intact during detoxification. Some lumpiness was found related to mineral role to make a bridge between starch granules. After gelatinization, many starch granules remained intact, although some agglomeration occurred.

## Conflict of interest

The authors declare no conflict of interest.

## Author contribution

First author has contribution of research planning, data interpretation, and manuscript preparation. Second author contributes to execute the research plan, data analysis, statistical analysis. and data presentation. Third author contributes to the data analysis and interpretation. Fourth author has contribution to manuscript preparation.

## Acknowledgements

The authors are incredibly grateful to Universitas Brawijaya for funding this research through Hibah Guru Besar Year 2019-2020.

## References

Abbasi, Z. (2012). Water resistance, weight loss, and enzymatic degradation of blends starch/polyvinyl alcohol-containing $SiO_2$ nanoparticles. *Journal of the Taiwan Institute of Chemical Engineers*, 43(2), 264-268. http://dx.doi.org/10.1016/j.jtice.2011.10.007.

Aichayawanich, S., Nopharatana, M., Nopharatana, A., & Songkasiri, W. (2011). Agglomeration mechanisms of cassava starch during pneumatic conveying drying. *Carbohydrate Polymers*, 84(1), 292-298. http://dx.doi.org/10.1016/j.carbpol.2010.11.036.

Ashri, A., Yusof, M., Jamil, M., Abdullah, A., Yusoff, S., Arip, M., & Lazim, A. (2014). Physicochemical characterization of starch extracted from Malaysian wild yam (*Dioscorea hispida Dennst.*). *Emirates Journal of Food and Agriculture*, 26(8), 652-658. http://dx.doi.org/10.9755/ejfa.v26i8.17098.

AOAC. (2011). *Official Methods of Analysis of AOAC International* (18th ed.). Gaitherburg: AOAC International.

Bandna, C., & Chand, B. (2012). Effect of Processing on the cyanide content of cassava products in Fiji. *Journal of Microbiology (Seoul, Korea)*, 13(2), 947-958.

Bertolini, A. C. (2010). *Starch: characterization, properties, and applications* (Vol. 4). New York: CRC Press.

Bhandari, M. R., & Kawabata, J. (2004). Assessment of antinutritional factors and bioavailability of calcium and zinc in wild yam (*Dioscorea* spp.) tubers of Nepal. *Food Chemistry*, 85(2), 281-287. http://dx.doi.org/10.1016/j.foodchem.2003.07.006.

Bolarinwa, I. F., Oke, M. F., Olaniyan, S. A., & Ajala, A. S. (2016). A review of cyanogenic glycosides in edible plants. In *Toxicology - New Aspects to This Scientific Conundrum* (Vol. 32, pp. 179-191). London: Intechopen. http://dx.doi.org/10.5772/64886.

Bradbury, J. H., Egan, S. V. & Lynch, M. J. (1991). Analysis of cyanide in cassava using acid hydrolysis of cyanogenic glucosides. *Journal of Science Food and Agriculture*, 55, 211-290. https://doi.org/10.1002/jsfa.2740550213.

Bradbury, J. H., Cliff, J., & Denton, I. C. (2011). Uptake of wetting method in Africa to reduce cyanide poisoning and konzo from cassava. *Food and Chemical Toxicology*, 49(3), 539-542. http://dx.doi.org/10.1016/j.fct.2010.04.049. PMid:20510334.

Burns, A. E., Bradbury, J. H., Cavagnaro, T. R., & Gleadow, R. M. (2012). Total cyanide content of cassava food products in Australia. *Journal of Food Composition and Analysis*, 25(1), 79-82. http://dx.doi.org/10.1016/j.jfca.2011.06.005.

Dar, M. Z., Deepika, K., Jan, K., Swer, T. L., Kumar, P., Verma, R., Verma, K., Prakash, K. S., Jan, S., & Bashir, K. (2018). Modification of structure and physicochemical properties of buckwheat and oat starch by γ-irradiation. *International Journal of Biological Macromolecules*, 108, 1348-1356. http://dx.doi.org/10.1016/j.ijbiomac.2017.11.067. PMid:29133096.

Dome, K., Podgorbunskikh, E., Bychkov, A., & Lomovsky, O. (2020). Changes in the crystallinity degree of starch having different types of crystal structure after mechanical pretreatment. *Polymers*, 12(3), 1-12. http://dx.doi.org/10.3390/polym12030641. PMid:32178224.

Ferraro, V., Piccirillo, C., Tomlins, K., & Pintado, M. E. (2016). Cassava (*Manihot esculenta* Crantz) and yam (*Dioscorea* spp.) crops and their derived foodstuffs: safety, security and nutritional value. *Critical Reviews in Food Science and Nutrition*, 56(16), 2714-2727. http://dx.doi.org/10.1080/10408398.2014.922045. PMid:26165549.

Han, C. H., Liu, J. C., Fang, S. U., & Hou, W. C. (2013). Antioxidant activities of the synthesized thiol-contained peptides derived from computer-aided pepsin hydrolysis of yam tuber storage protein, dioscorin. *Food Chemistry*, 138(2-3), 923-930. http://dx.doi.org/10.1016/j.foodchem.2012.11.101. PMid:23411197.

Hargono, H., Jos, B., & Kumoro, A. C. (2017). Kinetics of the enzymatic hydrolysis of sweet cassava starch, bitter cassava, and gadung (*Dioscorea hispida* dennst) flours at low temperature. *Bulletin of Chemical Reaction Engineering & Catalysis*, 12(2), 256-262. http://dx.doi.org/10.9767/bcrec.12.2.808.256-262.

Hedayati, S., Shahidi, F., Majzoobi, M., Koocheki, A., & Farahnaky, A. (2020). Structural, rheological, pasting and textural properties of granular cold water swelling maize starch: effect of NaCl and $CaCl_2$. *Carbohydrate Polymers*, 242, 116406. http://dx.doi.org/10.1016/j.carbpol.2020.116406. PMid:32564824.

Hendry-Hofer, T. B., Ng, P. C., Witeof, A. E., Mahon, S. B., Brenner, M., Boss, G. R., & Bebarta, V. S. (2019). A review on ingested cyanide: risks, clinical presentation, diagnostics, and treatment challenges. *Journal of Medical Toxicology; Official Journal of the American College of Medical Toxicology*, 15(2), 128-133. http://dx.doi.org/10.1007/s13181-018-0688-y. PMid:30539383.

Huang, R., Xie, J., Yu, Y., & Shen, M. (2020). Recent progress in the research of yam mucilage polysaccharides: Isolation, structure and bioactivities. *International Journal of Biological Macromolecules*, 155, 1262-1269. http://dx.doi.org/10.1016/j.ijbiomac.2019.11.095. PMid:31730974.

Kambale, K. J., Ali, E. R., Sadiki, N. H., Kayembe, K. P., Mvumbi, L. G., Yandju, D. L., Boivin, M. J., Boss, G. R., Stadler, D. D., Lambert, W. E., Lasarev, M. R., Okitundu, L. A., Ngoyi, D. M., Banea, J. P., & Tshala-Katumbay, D. D. (2017). Lower sulfur transferase detoxification rates of cyanide in Konzo—A tropical spastic paralysis linked to cassava cyanogenic poisoning. *Neurotoxicology*, 59, 256-262. http://dx.doi.org/10.1016/j.neuro.2016.05.016. PMid:27246648.

Kumoro, A. C., Retnowati, D. S., & Budiyati, C. S. (2011). Removal of cyanides from gadung (*Dioscorea hispida* Dennst.) tuber chips using leaching and steaming techniques. *Journal of Applied Sciences Research*, 7(12), 2140-2146.

Li, J. Y., & Yeh, A. I. (2001). Relationships between thermal, rheological characteristics and swelling power for various starches. *Journal of Food Engineering*, 50(3), 141-148. http://dx.doi.org/10.1016/S0260-8774(00)00236-3.

Li, T., Fan, J., Chen, W., Shu, J., Qian, X., Wei, H., Wang, Q., & Shen, J. (2016). Coaggregation of mineral filler particles and starch granules as a basis for improving filler-fiber interaction in paper production. *Carbohydrate Polymers*, 149, 20-27. http://dx.doi.org/10.1016/j.carbpol.2016.04.082. PMid:27261726.

Liu, Y. W., Shang, H. J., Wang, C. K., Hsu, F. L., & Hou, W. C. (2007). Immunomodulatory activity of dioscorin, the storage protein of yam (Dioscorea alata cv. Tainong No. 1) tuber. *Food and Chemical Toxicology*, 45, 2312–2318. http://doi.org/10.1016/j.fct.2007.06.009.

Manupriya, B. R., Lathika, Somashekarappa, H. M., Patil, S. L., & Shenoy, K. B. (2020). Study of gamma irradiation effects on the physico-chemical properties of wheat flour (*Triticum aestivum*,L.). *Radiation Physics and Chemistry*, 172, 108693. http://dx.doi.org/10.1016/j.radphyschem.2020.108693.

Padhan, B., Biswas, M., & Panda, D. (2020). Nutritional, anti-nutritional and physico-functional properties of wild edible yam (*Dioscorea* spp.) tubers from Koraput, India. *Food Bioscience*, 34, 100527. http://dx.doi.org/10.1016/j.fbio.2020.100527.

Panghal, A., Munezero, C., Sharma, P., & Chhikara, N. (2019). Cassava toxicity, detoxification and its food applications: a review. *Toxin Reviews*, 40(1), 1-16. http://dx.doi.org/10.1080/15569543.2018.1560334.

Patindol, J., Shih, F., Ingber, B., Champagne, E., and Boue, S. (2013). Porous rice powder from the precipitation of gelatinized flour or starch paste with ethanol. *Starch - Stärke*, 65(3-4), 296-303. https://doi.org/10.1002/star.201200134.

Saleha, S., Saidi, N., Saiful, Murniana, Rasnovi, S., & Iqbalsyah, T. M. (2018). Nutritional composition of *Dioscorea hispida* from different locations around Leuser ecosystem area. *Jurnal Natural*, 18(1), 1-6. http://dx.doi.org/10.24815/jn.v18i1.8504.

Santos, T. P. R., Franco, C. M. L., Mischan, M. M., & Leonel, M. (2019). Improvement in spray-drying technology for preparation of pregelatinized cassava starch. *Food Science and Technology (Campinas)*, 39(4), 939-946. http://dx.doi.org/10.1590/fst.10418.

Sethi, G., Shanmugam, M. K., Warrier, S., Merarchi, M., Arfuso, F., Kumar, A. P., & Bishayee, A. (2018). Pro-apoptotic and anti-cancer properties of diosgenin: a comprehensive and critical review. *Nutrients*, 10(5), 645. http://dx.doi.org/10.3390/nu10050645. PMid:29783752.

Shih, S. L., Lin, Y. S., Lin, S. Y., & Hou, W. C. (2015). Effects of yam dioscorin interventions on improvements of the metabolic syndrome in high-fat diet-induced obese rats. *Botanical Studies*, 56(1), 4. PMid:28510813.

Singh, G., Verma, A. K., and Kumar, V. (2016). Catalytic properties, functional attributes and industrial applications of β-glucosidases. *3 Biotech*, 6(1), 1-14.

Siritunga, D., & Sayre, R. (2004). Engineering cyanogen synthesis and turnover in cassava (*Manihot esculenta*). *Plant Molecular Biology*, 56(4), 661-669. http://dx.doi.org/10.1007/s11103-004-3415-9. PMid:15630626.

Son, I. S., Kim, J. H., Sohn, H. Y., Son, K. H., Kim, J. S., & Kwon, C. S. (2007). Antioxidative and hypolipidemic effects of diosgenin, a steroidal saponin of yam (*Dioscorea* spp.), on high-cholesterol fed rats. *Bioscience, Biotechnology, and Biochemistry*, 71(12), 3063-3071. http://dx.doi.org/10.1271/bbb.70472. PMid:18071250.

Sun, X., Li, W., Hu, Y., Zhou, X., Ji, M., Yu, D., Fujita, K., Tatsumi, E., & Luan, G. (2018). Comparison of pregelatinization methods on physicochemical, functional and structural properties of tartary buckwheat flour and noodle quality. *Journal of Cereal Science*, 80, 63-71. http://dx.doi.org/10.1016/j.jcs.2018.01.016.

Svasti, J., Phongsak, T., & Sarnthima, R. (2003). Transglucosylation of tertiary alcohols using cassava β-glucosidase. *Biochemical and Biophysical Research Communications*, 305(3), 470-475. http://dx.doi.org/10.1016/S0006-291X(03)00793-9. PMid:12763016.

Tang, S., Zou, P., Xiong, H., & Tang, H. (2008). Effect of nano-$SiO_2$ on the performance of starch/polyvinyl alcohol blend films. *Carbohydrate Polymers*, 72(3), 521-526. http://dx.doi.org/10.1016/j.carbpol.2007.09.019.

Tattiyakul, J., Naksriarporn, T., & Pradipasena, P. (2012). X-ray Diffraction pattern and functional properties of *Dioscorea hispida* Dennst starch hydrothermally modified at different temperatures. *Food and Bioprocess Technology*, 5(3), 964-971. http://dx.doi.org/10.1007/s11947-010-0424-3.

Tikhonova, M. A., Yu, C. H., Kolosova, N. G., Gerlinskaya, L. A., Maslennikova, S. O., Yudina, A. V., Amstislavskaya, T. G., & Ho, Y. J. (2014). Comparison of behavioral and biochemical deficits in rats with hereditary defined or d-galactose-induced accelerated senescence: evaluating the protective effects of diosgenin. *Pharmacology, Biochemistry, and Behavior*, 120, 7-16. http://dx.doi.org/10.1016/j.pbb.2014.01.012. PMid:24495488.

Tohda, C., Yang, X., Matsui, M., Inada, Y., Kadomoto, E., Nakada, S., Watari, H., & Shibahara, N. (2017). Diosgenin-rich yam extract enhances cognitive function: a placebo-controlled, randomized, double-blind, crossover study of healthy adults. *Nutrients*, 9(10), 1160. http://dx.doi.org/10.3390/nu9101160. PMid:29064406.

Wang, H., Xiao, N., Wang, X., Zhao, X., & Zhang, H. (2019). Effect of pregelatinized starch on the characteristics, microstructures, and quality attributes of glutinous rice flour and dumplings. *Food Chemistry*, 283, 248-256. http://dx.doi.org/10.1016/j.foodchem.2019.01.047. PMid:30722868.

Waterschoot, J., Gomand, S. V., & Delcour, J. A. (2016). Impact of swelling power and granule size on pasting of blends of potato, waxy rice and maize starches. *Food Hydrocolloids*, 52, 69-77. http://dx.doi.org/10.1016/j.foodhyd.2015.06.012.

Yang, H. S., Che, Y. J., Liao, Z., & Lan, T. (2014). The influence of amylopectin on the hydration products of cement. *Applied Mechanics and Materials*, 638-640, 1485-1489. http://dx.doi.org/10.4028/www.scientific.net/AMM.638-640.1485.

Zhang, B., Guo, K., Lin, L., & Wei, C. (2018). Comparison of structural and functional properties of starches from the rhizome and bulbil of Chinese yam (*Dioscorea opposita* Thunb.). *Molecules (Basel, Switzerland)*, 23(2), 8-11. http://dx.doi.org/10.3390/molecules23020427.