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**BIOCHEMISTRY**

# Effect of Debittering on the Physical and Chemical Properties of Palmyrah Young Shoots Flour

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**ABSTRACT**

Palmyrah young shoot is rich in starch and fibre, which is helpful in controlling various diseases especially diabetic. Regular consumption of this flour increase the body strength, reduce hunger and incorporation of it in other foods would positively reduce the malnutrition. However, presence of bitter compounds (flabelliferin or steroidal saponin) limited its consumption rate. Very few studies are carried out in this area and hence the present study was aimed to remove the bitter compounds by aqueous extraction and to evaluate the effect of debittering on changes of palmyrah young shoot flour. The starch and fiber content were dramatically increased and phenolic and saponin contents significantly decreased in debittered flour (DBF) than raw flour (RF). In addition, the color value 'L' value increased whereas 'a' and 'b' value decreased in DBF. The debittered flour had good thickening and gelling properties, which can sustain the use of debittered palmyarh flour in food industries.

**Highlights**

- Palm young shoot is a fibre rich food but still an underutilized crop. The reason behind the under utility is the bitterness in the palmyrah young shoot which limits it consumption. Addressing this problem, the present study was conducted to debitter the flour of palm young shoot and increase its utility among the people as a fibre rich food.
- The debittered palmyrah young shoot flour can be industrially utilized and commercialized for its fibre content and can be easily blended with other foods for processing fibre rich foods.
- The yield of fibre obtained from debittered flour was found to be higher when compared with raw flour. The manufacturing of debittered flour will create great scope in food and agriculture industries. The comparative study conducted on the effect of debittering on the physical and chemical properties on the palmyarh young shoot flour will prove the importance of analytical chemistry to researchers.

**Keywords:** Palmyrah young shoot; debittered flour; physical and chemical properties

Owing to the presence of bitterness and inadequate supplies of starch and fiber, research focused to the food properties and the possible use of starch and fiber from economic, underutilized fiber rich food plant based foods has become of expected attention (Enujiugha and Ayodele-Oni 2003). The starch and fiber rich flour from unknown sources must attain proper interaction characteristics with other components of food (e.g., water, lipid) in order to improve their addition in new food product formulations (Kinsella 1982). Palmyrah (*Borassus flabellifer* L) is a miracle tree in the family of Arecaceae is widespread in the Asian region including the northern and eastern parts of Sri

Lanka, Southern parts of India and in the most of other tropical countries (Thabrew and Jansz 2004). Asian variety of palm have genus of six species of fan palms in which B. aethiopum Mart, found in Africa, *Borassus flabellifer* L. in coastal areas of southeast Asia including India, B. sundaicus Becc restricted to Indonesia (Davis and Johnson 1987). India is one of the major producers of palms nearly 122 million palms per year (Saravanya and Kavitha 2017) and users of palmyrah young shoot in which the Tamil Nadu state having an area harvested, yield and production of approximately 10867.00 ha in the season 2015-2016. In India, palm is largely grown in Andhra Pradesh, Tamil Nadu, Bihar and Orissa. It grow very slow and takes from 15-30 years to bear, growing up to 30m high, the trunk may have a 1.5m circumference at the base and may have 25-40 leaves. Borassus palms are three most important economic species that provide significant value to local populations. It has nearly 800 uses including food and beverage, fiber, ayurvedic and medicinal (Arulraj and Augustine 2008). Neera, toddy, palm sugar, palm jaggery, tender palm fruit or nungu, ripe palm fruit and palm young shoot or young shoot are various edible products of palmyrah palm (Sandhya 2010).

The young shoot of palm is an unknown food, which is consumed predominantly in the north and northeast provinces of Sri Lanka and some parts of Southeast Asia (Mason and Henry 1994). It is the 6-8 months germinated seed of the palmyrah palm that grows downwards into the earth, which is known as palm, shoots and has 98% fiber and 95% starch content (Saravanya and Kavitha 2017). The young shoot is spindle shaped with maximum swelling at the bottom and tapers towards the tipper end (Balasubramanium *et al.* 1999; Mohandas 1983) and it contains 44% organic matter, 50-55% moisture and 2% minerals (Jeyaratnam 1986; Jansz *et al.* 1992). It is a seasonal product, available from November to January. The production of palmyrah young shoot and its products were produced around 600T per year (Mohandas 1983). The shoot generally grows 12-15cm in height before harvesting. It can be eaten boiled, sun dried and powdered, or used in the preparation of various foods. It is considered as good source of carbohydrates such as fiber and starch and some micronutrients such as calcium, magnesium and ferrous ions (Mason and Henry 1994). Starch is the major carbohydrate present in palmyrah shoot flour. Sucrose, glucose and fructose are major sugars present (Jeyaratnam 1986). It has considerable fiber feeding low glycaemic index (Jansz *et al.* 2002). The palmyrah young shoot based products are Tuber starch, Palmosha, Biscuits, Porridge and steamed food or "pittu" respectively (Balasubramanium *et al.* 1999). Owing to the presence of variety of steroidal saponins (flabelliferin) and an antimicrobial flabelliferin FB (Wickramasekara and Jansz 2003) which makes the young shoot flour to bitter taste and presence of toxic effects leads to its unacceptability. Spirosterols is the most dominant aglycone in shoot flour and in palmyrah inflorescence (Theivendirarajah 1994; Yoshikawa *et al.* 2007). Debittering of unboiled palmyrah tuber flour was done by soaking tuber flour in distilled water for three hours at ambient conditions thrice while replacing water per hour. Bitterness removal from un-boiled tuber flour was seen by the color change of water to dark brown, light brown and light yellow during the first, second and third treatments respectively (Sharaniya *et al.* 2015). Therefore, the purpose of present work was to optimize the precise conditions for bitterness removal from palmyrah young shoot flour by aqueous extraction and to determine the physico-chemical and functional properties of raw shoot flour and debittered or processed flour.

## MATERIALS AND METHODS

### Plant Materials

Palmyrah young shoot (*Borassus flabellifer* L.) were obtained from grower located in Valantharavai and Therkuvaniveethi, Ramanathapuram district, Tamil Nadu, India.

### Flour Preparation

The palmyrah young shoots were cleaned, removed outer layer and centre stick like portion, cut into small pieces, dried at 60°C for 12 hours in cabinet dryer, milled by using pin mill and hammer mill and sieved by sieve analyzer. Debittered flour was prepared by aqueous extraction and the extraction conditions were used for debittering 18.05 g in 90.27ml water at 53°C for 1 hour. After extraction at this conditions the mixture were stirred continuously for 1 hour at 45°C. Then it was centrifuged at 1000 rpm at 4°C for 10 mins, removed the supernatant and this process were repeated 3 times. The debittered residues were dried in hot air oven at 60°C for 6 hours and powdered using pulverizer. The raw flour and debittered or processed flour were stored in airtight pouches for analysis (Al-Farsi and Lee 2008).

### Proximate Analysis

Flour was tested for their proximate composition such as moisture, protein, ash, fat and crude fiber according to AOAC Method. Total carbohydrate and starch was determined by Anthrone method (Sadasivam 2005).

### Physical Analysis

**Bulk density:** The flour was filled into 10 mL graduated cylinder and their weight noted. The cylinder was tapped continuously until there was no further change in volume. Bulk density expressed as weight of flour per unit volume (g/mL) (Narayana and Rao 1984).

**pH:** The pH of palmyrah young shoot flour was determined according to the method of Falade & Okafor (Falade and Okafor 2015). The method described as 10 g of flour was suspended in 100mL (10% w/v) distilled water in cleaned 200mL beaker and it was allowed to settle at 30±2 °C for 15 min. Then the pH of the sample was determined by using pH meter (Model-ELICO LI-120). The measure of pH value gives a nature of either acidity or alkalinity of the flour (Vengaiah *et al.* 2013).

**Water activity ($a_w$):** It was determined using the Model 4TE Aqua Lab Dew Point water activity meter at 25 °C. The water activity meter was standing for 2 minute before analysis of flour sample. After that, the sample was filled 3/4$^{th}$ in sample cup. Then the sample cup was placed in sample holder and then turned the switch on mode. Finally the value obtained within 10-15 minutes in display of $a_w$ meter was recorded (Selani *et al.* 2016).

### Color analysis

Color value of flour was measured by the colorimeter, with illuminant C, based in the CIELAB color space. All the analyses were measured in triplicate. The following color coordinates were determined: lightness (L*), redness (a* ± red-green) and yellowness (b* ± yellow-blue) (Lucas-González *et al.* 2017).

### Anti-nutritional factor Analysis

**Total phenol content:** It was determined using the Folin-Ciocalteu reagent method (Bray and Thorpe 1954). One gramme of palmyrah young shoot flour were taken in 50mL centrifuge tube containing 10mL 80 % ethanol and homogenize well using vortex mixer for 5 min. Then centrifuge the sample at 5000rpm for 20 min. After that, collect the supernatant. In that freshly prepared supernatant 0.5 or 1mL of sample was taken in cleaned and dried test tubes. Add 8mL of distilled water in each test tube. Simultaneously the standard Gallic acid was taken at different concentration (0.2, 0.4, 0.6, 0.8 and 1mL) in test tube. Then all the concentration were made up to 8.5mL using distilled water. 0.5mL of Folin's Ciocalteau reagent added to all the test tubes. All the tubes were kept for incubation at 40°C for 10 minutes. Then, 1mL of Sodium Carbonate solution was added and test tubes were kept in the dark for incubation for 1 hour. After incubation purple color developed. Then the solution was read at 660 nm in a UV-spectrophotometer.

**Total saponin content:** The saponin content of the samples was determined by double extraction gravimetric method (Harborne 1973). Five gram of flour was taken in a conical flask and 100 cm$^3$ of 20% aqueous ethanol were added. The samples were heated over a hot water bath for 1hr 30 min with continuous stirring at about 55 °C. The mixture was filtered and the residue re-extracted with another 100 cm$^3$ of 20% ethanol. The combined extracts were reduced to 40 ml over water bath at about 90 °C. Then it was transfered into 250 mL separating funnel and add 20 mL of diethyl ether and shaken vigorously. The aqueous layer was collected while the ether layer was discarded and this purification process was repeated. Then 60 mL of n-butanol was added and it was washed twice with 10 ml of 5% aqueous sodium chloride. The collected solution was heated in a water bath for partial evaporation. After evaporation, the samples were dried in the oven at 60 °C to constant weight and the saponin content was calculated. The analysis were conducted in triplicate.

$$\text{Saponin (\%)} = (W_2 - W_1) / \text{weight of the sample} * 100$$

Where,

W1 = Weight of empty dish

W2 = Weight of empty dish + sample

### Statistical analysis

All the experiments were done in triplicate and the values were presented as mean ± SD. The Microsoft excel software (version 7.0) was used for statistical analysis. The statistical significance difference between values was analyzed by Paired Student's *t* test. Results were considered to be significant at *p*>0.05.

## RESULTS AND DISCUSSION

### Proximate composition

The nutrient composition of raw and debittered flour such as carbohydrate, protein, crude fibre, ash, fat, starch and energy values were 78.28%, 7%, 4.1%, 1.25%, 0.15%, 43.65%, 342.47kcal and 80.51%, 8.5%, 7.41%, 1.29%, 0.05%, 78.55% and 356.49kcal respectively. The result shown starch is major carbohydrate also three is no significant differences observed between the carbohydrate, starch, crude fibre, protein and ash (p<0.05). The starch and crude fiber content of raw and DBF was compared to African variety (Narayana and Rao 1984) and Asian, Andhra Pradesh variety (Vengaiah *et al.* 2013). The raw flour had less fiber than both variety, which may due to the variation of cultivation and soil and environmental conditions. The starch and fiber content were increased in DBF than RF, which may due to aqueous extraction method that facilitated the breakdown of complex substances into simpler substances leading to increased recovery of polar compounds.

**Table 1:** Proximate composition of palmyarh young shoots flour

| Proximate composition | Sample (%) | |
|---|---|---|
| | Raw flour | Debittered flour |
| Moisture | 9.22 ± 0.017 | 8.97 ± 0.020 |
| Carbohydrate | 78.28 ± 0.090 | 80.51 ± 0.046 |
| Fat | 0.15 ± 0.010 | 0.05 ± 0.026 |
| Crude fiber | 4.1 ± 0.100 | 7.41 ± 0.174 |
| Protein | 7 ± 0.100 | 6.4 ± 0.300 |
| Ash | 1.25 ± 0.050 | 1.29 ± 0.114 |
| Starch | 43.65 ± 0.017 | 78.55 ± 0.695 |
| Energy | 342.47 ± 11.738 | 356.49 ± 8.821 |

Table 1 shows that debitterd young shoot flour is rich in carbohydrates especially starch and fiber hence they can potentially be fortified with other foods for fiber enrichment and can able to used as thickeners in food industries. There is some significant difference observed in the moisture content of RF (9.22%) and DBF (8.97%) (p>0.05).

### Physical Analysis

**Bulk density:** It is a measure of weightiness of a flour sample and expressed as weight by volume (g/mL). Bulk density of RF and DBF was 0.73 and 0.86 g/mL (Table 2) respectively, which depends upon the particle size of the samples.

**Table 2:** Physical analysis of palmyarh young shoots flour

| Proximate composition | Sample (%) | |
|---|---|---|
| | Raw flour | Debittered flour |
| Bulk density (g/mL) | 0.73 ±0.026 | 0.857 ± 0.042 |
| pH | 6.27 ± 0.026 | 5.93 ± 0.885 |
| Water activity ($a_w$) | 0.45 ± 0.006 | 0.08 ± 0.010 |

Statistically no differences found between the two samples (p>0.05). It is important for determining packaging requirements, material handling and application in wet processing in the food industry. The bulk density of the shoot flour was relatively higher than wheat flour (0.7 g/mL) so that can be used as thickeners in food products (Ali1 *et al.* 2010).

**pH:** The pH is the important parameter for estimating the quality of flour. The pH value of the raw and dibittered flour were 6.29 and 5.93 respectively and it was obtained within the neutral pH range, that is the palmyrah young shoot flour is considered as low acid food (>4.6). The debitterd flour has low pH than RF this may due to the presence of alkaline substances released away during dibittering process. Hence, consumption of shoot flour may help limit muscle loss, strengthen memory and alertness, and increase the living years. The pH of the DBF was compared with previous report (pH-5.8) but not much deviation was observed in the both samples (Vengaiah *et al.* 2013). Statistically significant differences observed between the pH of RF and DBF (p>0.05).

### Water activity

Water activity defines the water in the food is not available for the growth of microorganisms, measured by water activity meter, which contain the scale range from zero to one. Water activity of raw shoot flour is 0.45 and DBF is 0.08 (Table 2) since no significant differences observed in both samples ($p<0.05$). The results showed that food which has less than 0.85 were considered as non-hazardous since palmyrah shoot flour may not support the growth of harmful bacteria and may have better shelf life because the $a_w$ directly correlated with shelf life of food.

### Color analysis

The color values of flour were shown in (Table 3), $L^*$ value of DBF increased than RF whereas $a^*$ $b^*$ value decreased. $L^*$ of DBF was almost similar to the freeze dried mango coproducts (81.09) (Selani *et al.* 2014). This result was correlated with (Lucas-González *et al.* 2017) report, which proved that decreased lightness with increasing particle size. $L^*$ value of DBF was increased due to the surface area that is greater the surface area greater the reflection of light. The surface area and reflection of light directly correlated with each other (Ahmed *et al.* 2015). The 'a', 'b' value of debitteted flour (1.99, 0.28) decreased which might be due to the decreasing of total phenolic compounds and total saponin content during debittering process since these compounds are leached out into water from flour. Greenness ($a^*$) and yellowness ($b^*$) were decreased with decreased particle size and with greater surface area. The significant differences in color values related with particle size and processing method could be due to the loss of pigment during milling and debittering process ($p>0.05$). Any change in the color parameters may be considered unsuitable and influence consumer acceptance (Selani *et al.* 2014).

**Table 3:** Color value of palmyarh young shoot flour

| Color value | Sample | |
|---|---|---|
| | Raw flour | Debittered flour |
| L | 80.08 ± 1.386 | 71.85 ± 0.867 |
| a | 1.99 ± 0.118 | 0.28 ± 0.057 |
| b | 17.78 ± 0.389 | 7.92 ± 0.142 |

### Anti-nutritional properties

**Total phenolic content:** TPC of DBF decreased significantly than RF. In addition, there was a significant difference between the RF (320 mg/100g) and DBF (290 mg/100g) ($p>0.05$). These results were compared with (Ahmed *et al.* 2016) results showed the *B. Flabellifer* L. has more phenolic compounds than *B. aethiopum* Mart (275.75 mg/100g) (Table 4).

**Total saponin content:** Total saponin content of DBF (0.19 mg/100g) was reduced favorably than RF (2.04 mg/100g) ($p>0.05$). The saponin content in RF was similar to *B. aethiopum* Mart (2.18 mg/100g). The total phenolic and saponin contents were reduced due to leaching of these compounds from flour during debittering process. Total phenolic and saponin content is shown in (Table 4).

**Table 4:** Antinutrional factors of palmyarh young shoot flour

| Antinutrional factors | Amount (mg/100g) | |
|---|---|---|
| | Raw flour | Debittered flour |
| Saponin | 2.04 ± 0.113 | 0.19 ± 0.010 |
| Total polyphenol (TPP) | 320 ± 3.464 | 220 ± 3.606 |

## CONCLUSION

The palmyrah young shoot is much underutilized, non-popular and cheapest product widespread in Asian and African countries. It is rich in carbohydrates especially fiber and starch however presence of bitterness makes the limit for consumption. The present study is focused on the bitterness removal and effect of changes on physical and chemical properties of young shoot flour. Debittering by aqueous extraction was increased the starch, fiber and protein content as well as the anti-nutritional factors such as total polyphenols and saponin content was reduced in debittered flour than raw flour. Bulk density of DBF (0.86 g/mL) was increased whereas pH, water activity and color value was decreased than raw flour. Hence this study was clearly explained bitterness was reduced in palmyrah young shoot flour due to changes of physical and chemical properties by aqueous extraction.

## ABBREVIATIONS USED

RF, raw flour; DBF, Debittered flour

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