

Article

# Extraction and Characterization of Crude Arabinoxylan from Breadfruit (*Artocarpus altilis*)

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## Abstract

Arabinoxylan is a dietary fiber belonging to the non-starch polysaccharide family and is known for its health-promoting properties, such as antioxidant and immunomodulatory effects. However, the extraction efficiency and structural integrity of arabinoxylan are strongly influenced by processing conditions, particularly temperature. Breadfruit (*Artocarpus altilis*) is a potential local source of arabinoxylan, containing up to 2.49% fiber, yet its extraction characteristics have not been widely investigated. Therefore, this study aimed to determine the effect of different extraction temperatures on the characteristics of crude arabinoxylan extracted from breadfruit using water as the solvent. A completely randomized design with one factor, extraction temperature (40°C, 45°C, and 50°C), was used with a 24-hour extraction period. The crude arabinoxylan obtained was characterized for yield, moisture content, ash content, starch content, and monosaccharide profile using HPLC. The results showed that extraction temperature significantly affected the physicochemical characteristics of crude arabinoxylan. The highest yield (4.1%) was obtained at 50°C, while the lowest starch and ash content occurred at 45°C. HPLC analysis revealed that arabinose was present in all treatments, whereas xylose was not detected; the highest arabinose concentration (1.04%) was found at 40°C. These findings indicate that lower extraction temperatures better preserve arabinose content, while higher temperatures enhance yield, suggesting that extraction temperature plays a crucial role in optimizing both purity and quantity of arabinoxylan from breadfruit.

**Keywords:** arabinoxylan, breadfruit, water extraction

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## 1. Introduction

Dietary fiber is a plant component that is resistant to digestion. Consuming dietary fiber is

increasingly popular due to its beneficial effects on the body. Arabinoxylan is one component of dietary fiber and is a non-starch polysaccharide composed of arabinose and xylose, and it is often

referred to as pentosan. Sources of arabinoxylan are generally found in cereals and other plant-based materials, such as wheat, bamboo shoots, banana peels, and corn bioethanol waste [15, 20]. The role of arabinoxylan in the food sector has been widely reported, for example in microencapsulation systems, bakery products, and edible films [6,30]. In the field of health, arabinoxylan has the potential to prevent several cardiovascular diseases, exhibits antioxidant activity, and stimulates immune responses; moreover, the structural and functional properties of water-unextractable arabinoxylan have been elucidated in recent studies [16,1,12,14]. The molecular structure of arabinoxylan is shown in Fig. 1.

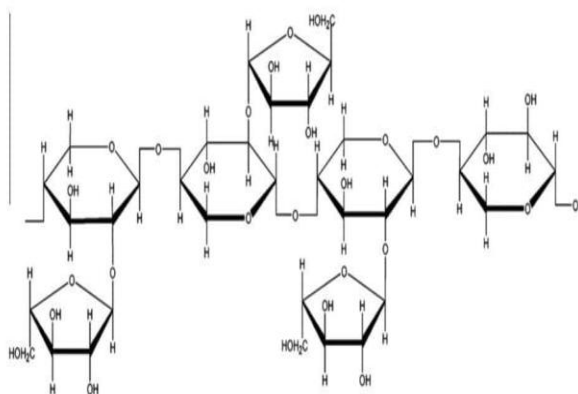


Fig 1. Molecular structure of arabinoxylan  
Source: (Sinha *et al.*, 2011)

Breadfruit (*Artocarpus altilis*) is a climacteric fruit with an oval or round shape that is widely distributed across Indonesia. It is often processed into secondary food products such as chips or flour. Breadfruit contains 1.21% ash, 61.8% water, 35.5% carbohydrates, 0.1% protein, 0.21% calcium, and 2–2.49% fiber [29,3]. Although a recent study [19] reported that arabinoxylan was not directly identified as a compositional component of breadfruit, its presence in other fiber-rich tropical plants suggests that potential non-starch polysaccharides such as arabinoxylan could be present in breadfruit fiber but remain uncharacterized. Therefore, the exploration of arabinoxylan in breadfruit represents a novel

approach to understanding its polysaccharide composition and functional potential.

Extraction is a key process for isolating bioactive compounds, including polysaccharides. The arabinoxylan extraction process can be categorized into water-extractable and non-water-extractable types. The choice of solvent must consider selectivity, solubility, cost, and safety [31]. Among available solvents, water is the simplest, safest, and most environmentally friendly, making it suitable for food-grade extraction.

The goal of this study was to determine the effect of different extraction temperatures on the characteristics of crude arabinoxylan extracted from breadfruit using water as the solvent. To achieve this objective, the research employed a completely randomized design (CRD) with one factor—extraction temperature (40°C, 45°C, and 50°C)—and characterized the crude arabinoxylan obtained based on yield, moisture content, ash content, starch content, and monosaccharide profile using high-performance liquid chromatography (HPLC).

## 2. Material and Method

### 2.1 Extraction

The procedure for extracting arabinoxylan from breadfruit flour followed the method of [6] with modifications. A total of 50 g of breadfruit flour was first dried at 55°C for 24 hours in a cabinet dryer to remove residual moisture and ensure uniform sample conditions prior to extraction. The dried flour was then subjected to defatting using hexane for 2 hours at a sample-to-solvent ratio of 1:3 to remove lipids.

After defatting, water extraction was performed by mixing the flour and distilled water at a ratio of 1:20 (w/v). The mixture was incubated for 24 hours at controlled extraction temperatures of 40°C, 45°C, or 50°C using a shaking water bath. These extraction temperatures were used to evaluate their effect on the solubility and yield of crude arabinoxylan.

Subsequently, the extract was treated with  $\alpha$ -amylase enzyme for 60 minutes at 85°C to remove starch, followed by enzyme inactivation and soluble protein coagulation by heating at 95°C for 30 minutes. The mixture was filtered through filter cloth and filter paper under vacuum. A fourfold volume of ethanol/acetone solution (1:1, v/v) was then added to the clear filtrate to precipitate arabinoxylan. The precipitate was centrifuged at  $1250 \times g$  for 10 minutes and washed repeatedly with acetone to remove water. Finally, the precipitate was dried and weighed to determine the yield.

## 2.2 Characterization of crude Arabinoxylan

At this stage, crude arabinoxylan is characterized, the tests carried out include: determination of yield, determination of water content (AOAC 2005), determination of ash content (AOAC 2005), determination of total starch content using the Luff Schoorl method (AOAC 1995), and identification of the monosaccharide content of crude arabinoxylan (HPLC). HPLC was used to detect the certain compound [13]. The following are HPLC conditions:

Column	: Metacharb H plus
Eluent / mobile phase	: H <sub>2</sub> O
Flow	: 0,5 ml/min
Temperature	: 70°C
Detector	: RID
LOD (limit of detection)	: Arabinose (0,2 ppm) Xylose (0,39 ppm)

Sample identification is carried out in two main steps: sample preparation and standard preparation. Sample preparation begins with careful weighing of the sample, followed by the addition of 2 ml of distilled water. Extraction is performed using a vortex (2 minutes), followed by sonication with water solvent (15 minutes), and centrifugation (5 minutes). The liquid phase is collected, and the extraction is repeated with 2 ml of distilled water. and added to a volume of 5 ml,

then filtered with 0.45  $\mu$ m millex and injected into a 20  $\mu$ L HPLC. Then the second step is standard preparation. This is done by making a mixture of (D+) xylose and arabinose 6.25; 12.5; 25; 50; 100; 250; 500; 1000 ppm and injected into a 20  $\mu$ L HPLC.

## 3. Results and Discussion

### 3.1 Yield

Extraction yield represents the efficiency of solvent and temperature in isolating arabinoxylan from breadfruit flour. As shown in Fig. 2, yield increased with extraction temperature, ranging from 3.2% at 40°C, 3.6% at 45°C, to 4.1% at 50°C. This trend indicates that higher temperatures enhance the diffusion of solvent into the plant matrix, thereby improving solubilization of polysaccharides.

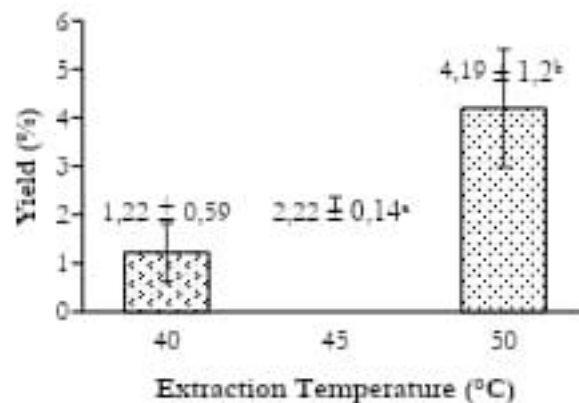


Fig 2. Crude Isolation Yield / Yield Graph Arabinoxylan

Note: The same letter notation indicates no significant difference ( $\alpha = 0.05$ ).

Compared to previous studies on cereals such as wheat and rye [6], the yield obtained from breadfruit was relatively higher, suggesting that breadfruit fiber could be a promising alternative source of arabinoxylan. However, excessive temperature may also promote partial degradation or denaturation of other compounds, which can influence purity. Therefore, while 50°C provides the highest yield, further compositional analysis is

required to confirm its selectivity toward arabinoxylan.

### 3.2 Water Content

Water content is a parameter used to determine the water residue after the drying process. Determining water content is related to the purity of the extraction results [28]. Some of the purposes of determining water content include determining the amount of water content as a percentage of the sample in the dry material, determining the sample's durability during storage, and establishing a lower limit for water content in the extract. Increasing the water content makes it easier for mold and mildew to grow, resulting in decreased biological activity during storage [21]. The water content value for crude arabinoxylan can be seen in the fig. 3.

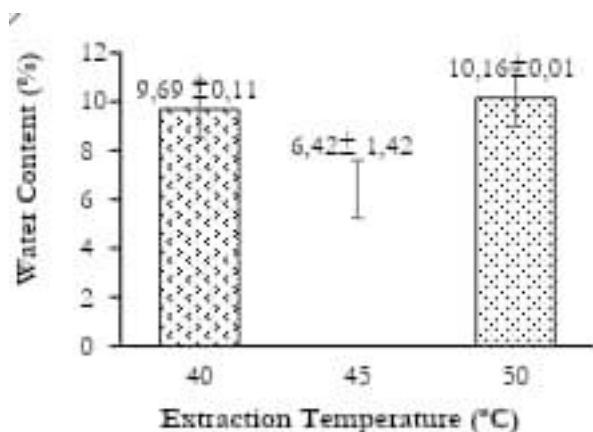


Fig 3. Crude Water Content Results Graph Arabinoxylan

Note: The same letter notation indicates no significant difference ( $\alpha = 0.05$ ).

Based on Fig. 3, it can be observed that the water content of crude arabinoxylan at extraction temperatures of 40 °C and 50 °C did not differ significantly, whereas a significant difference was found at 45 °C. The average water content ranged from 6.42% to 10.16%. The highest value was obtained at 50 °C, while the lowest was observed at 45 °C. The water content in this study was lower than that reported by Bell, 2015 [4], who extracted arabinoxylan from wheat bioethanol production

waste using water as a solvent, yielding a value of 12.1%. However, it was higher than the results of Cui et al. (1999), who extracted arabinoxylan using ethanol as a solvent and obtained a water content of 5.6% [11]. These findings indicate that the type of solvent influences the resulting water content value. This is consistent with the statement of Courtin and Delcour (1998), who noted that arabinoxylan has a high capacity to absorb water. Consequently, arabinoxylan extracted with water as the solvent tends to exhibit a higher water content [10].

### 3.3 Ash Content

Ash content determination is used to determine the inorganic or mineral content of a food ingredient. The ash content of crude arabinoxylan can be seen in fig. 4.

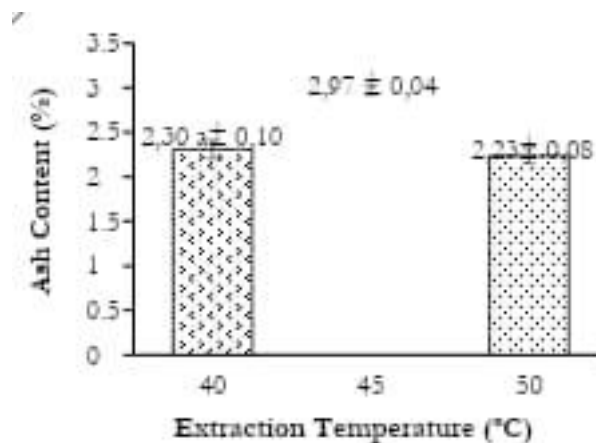


Fig 4. Crude Ash Content Results Graph Arabinoxylan

Note: The same letter notation indicates no significant difference ( $\alpha = 0.05$ ).

Based on fig. 4, the extraction temperature treatments of 40°C and 50°C did not have a significant effect, while the extraction temperature treatment of 45°C had a significant effect on the resulting ash content. The highest ash content value was found in the 45°C treatment with a value of 2.97%, while the lowest value was found in the 50°C treatment with a value of 2.23%. Obtaining ash content in crude arabinoxylan extraction from breadfruit producing an average

of between 2.23% - 2.97%. These results are in accordance with research by Zheng *et al.*, (2011) which extracted arabinoxylan from barley with an ash content of 2.06% [32]. However, it is lower than the research of Maes & Delcour (2002) who extracted arabinoxylan from wheat bran with an ash content of 5-9% [18]. The low ash content obtained is expected. In accordance with the statement of Salim *et al.*, (2016) the determination of ash content should obtain a low value because it is related to the factor of metal contamination which at high temperatures does not disappear [25]. Factors that influence the determination of ash content are the ashing method, type of material, time, and temperature used during heating (Sudarmadji 1989).

### 3.4 Total Crude Arabinoxylan Starch

Starch is a homopolymer carbohydrate of glucose with  $\alpha$ -glycosidic bonds [17]. Determination of total starch in crude the presence of arabinoxylan is related to the residue produced during the extraction process. Some residue may remain in the sample at the end. A graph of total starch yield is presented in fig. 5.

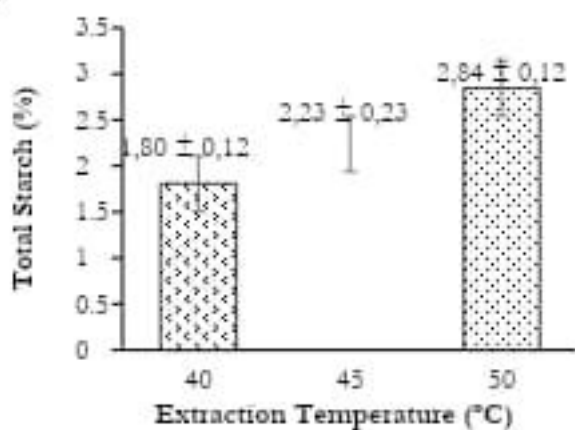


Fig 5. Graph of Total Crude Starch Yield Arabinoxylan

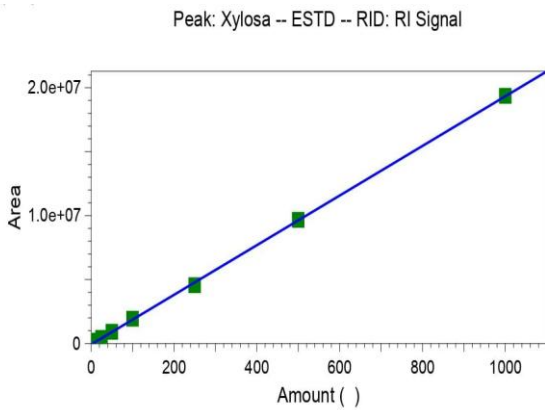
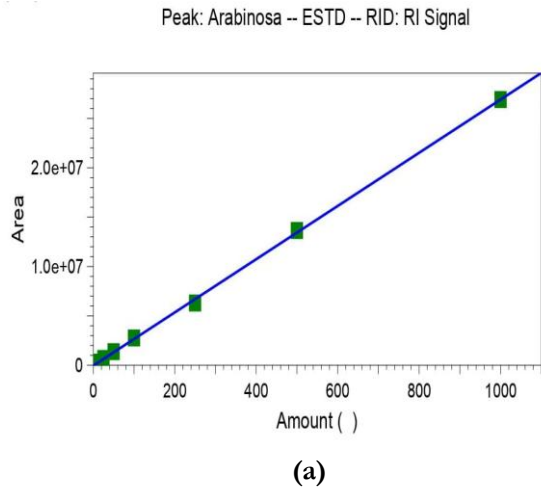
Note: The same letter notation indicates no significant difference ( $\alpha = 0.05$ ).

Based on fig. 5, it can be seen that the total content of crude arabinoxylan starch at extraction

temperatures of 40°C and 45°C had no significant difference and was significantly different at extraction temperature treatment of 50°C. The total crude arabinoxylan starch content averaged 1.80–2.84%. This result is inconsistent with the research of [6] who extracted arabinoxylan with water solvent from wheat flour and rye flour, with undetectable starch levels in both samples. Although starch residues were initially removed by incubation with the  $\alpha$ -amylase enzyme, it is possible that some residues may remain. As stated by [6] that foreign components such as protein and starch cannot be completely removed by enzymatic treatment during purification and still remain in the sample. According to Rosenthal *et al.*, (1996) pre-enzymatic treatment in an extraction process is an effective method for releasing active compounds from the membranes and cell walls of the material [24].

### 3.5 Crude Arabinoxylan Monosaccharides

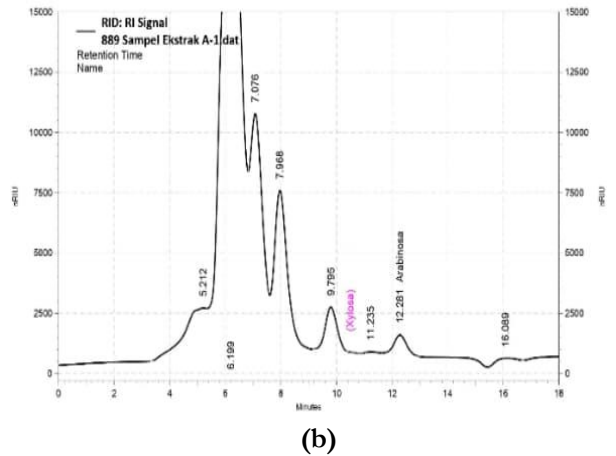
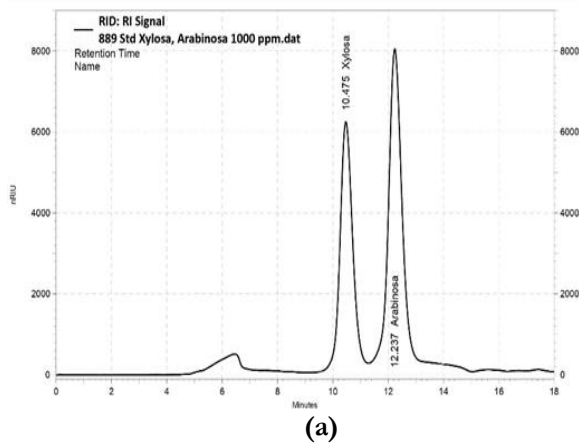
Arabinoxylan is a co-polymer compound composed of arabinose and xylose. Both compounds belong to the pentose monosaccharide group, consisting of five carbon atoms, one aldehyde group, and four alcohol groups. Identification test for arabinose and xylose monosaccharides using high-performance liquid chromatography (HPLC) instruments. Determining carbohydrate analysis using chromatography methods is considered more efficient and has high sensitivity. Calibration curves of arabinose and xylose can be seen in fig. 6. The type of column used in HPLC in this study was Metacharb H plus, mobile phase H<sub>2</sub>O with flow rate 0.5 ml/min, RID detector. The choice of mobile phase or water eluent is because deionized water has the same polarity as carbohydrates [23].



(b)

Fig 6. Standard Calibration Curve (a) arabinose (b) xylose

The temperature used in the column was 70°C and the amount of sample injected was 20µL. The area under the curve (AUC) of each concentration of the series solution at the maximum wave phase was determined to obtain a linear regression equation by entering the obtained AUC data versus the concentration data of the series solution [26]. If the R value is close to 1, the linearity parameter is met. The curve calibration of the arabinose standard obtained a regression equation  $y = 26954.8x - 37216.6$  with a correlation coefficient value of  $r^2 = 0.999615$  (fig. 6.a). Meanwhile, the standard calibration curve for xylose obtained the regression equation  $y = 19405.0x - 74026.2$  with a correlation coefficient value of  $r^2 = 0.999799$  (fig. 6.b). Based on fig. 6, the standard R values of arabinose and xylose are close to 1, So in this study the linearity parameters have been met.



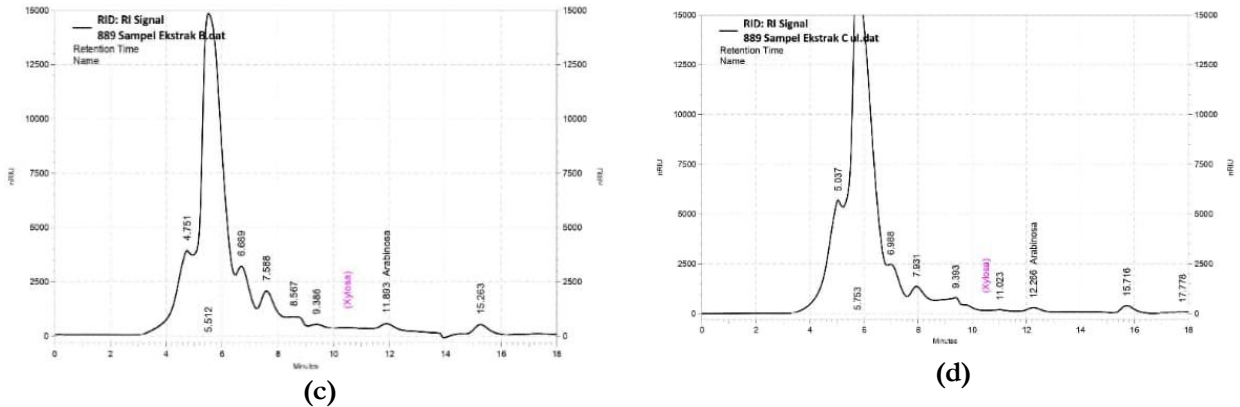


Fig 7. Chromatogram (a) Std Xylose, Arabinose 1000 ppm (b) sample extraction temperature 40°C (c) sample extraction temperature 45°C (d) sample extraction temperature 50°C.

Qualitative testing is carried out by equating the retention time of each peak from the sample chromatogram corresponds to the retention time of the standard compound [23]. Based on Fig. 7.a, standard arabinose has a retention time of 12.237 minutes and standard xylose at 10.475 minutes. Furthermore, the extract extracted from breadfruit flour at a temperature of 40°C was detected to contain arabinose with a retention time of 12.281 minutes (Fig. 7.b). The results of the extraction at a temperature of 45°C contained arabinose with a retention time of 11.893 minutes (Fig. 7.c) and the results of the extraction at a temperature of 50°C contained arabinose at a retention time of 12.266 minutes (Fig. 7.d).

Table 1. Crude Monosaccharide Analysis Results Arabinoxyylan

Sample	Arabinos e %b/b	D (+) Xylose %b/b
Extraction temperature 40°C	1,04	Not detected
Extraction temperature 45°C	0,30	Not detected
Extraction temperature 50°C	0,22	Not detected

Based on Table 1, it can be seen that the temperature treatment in the extraction process with water solvent produces an average arabinose

value between 0.22 - 1.04% b/b. The highest arabinose value was obtained at an extraction temperature of 40°C, namely 1.04%. While the lowest arabinose value was obtained at a temperature treatment of 50°C, namely 0.22%. The arabinose value obtained was much lower than the study of Buksa on 2016 [6] who extracted arabinoxyylan from wheat with water solvent, namely 35.8%. In this study, arabinoxyylan calculations were not carried out because xylose was not detected. The low arabinose value and the undetectable xylose value are thought to be due to the Maillard process occurring in the drying process. This is because, arabinose is classified as a reducing sugar [2]. Furthermore, the presence of a free aldehyde group causes this monomer to act as a reducing sugar [22]. The Maillard reaction is a non-enzymatic browning reaction between reducing sugars and amino acids that occurs during processing thermally processed food [7]. The extraction temperature treatment of 50°C produced a higher yield value compared to other temperature treatments (fig. 1). Higher yields require a longer drying process, resulting in more Maillard processes occurring at a temperature of 50°C.

The extraction of raw material is also a factor influencing the low monosaccharide value produced. This study used commercial breadfruit flour. Commercial breadfruit flour is made using ripe or nearly ripe breadfruit. Meanwhile, according to Considine [8] the fiber content in young breadfruit is higher than that in mature

breadfruit. Arabinoxylan is included in the fiber, related to the age of the breadfruit used in extraction, it affects the resulting monosaccharide value. In addition, in commercial breadfruit flour, the skin of the breadfruit is not included in the production of breadfruit flour. According to Bender *et al.*, [5] most arabinoxylan is found in plant cell walls in a complex matrix with cell wall components. Therefore, the monosaccharide value produced in this study is lower.

#### 4. Conclusion

This study demonstrated that extraction temperature significantly affects the physicochemical and compositional properties of crude arabinoxylan from breadfruit. Increasing the temperature from 40°C to 50°C increased yield (up to 4.1%) but reduced arabinose content due to possible degradation. Water and ash content exhibited distinct responses, with the lowest water content (6.42%) and highest ash content (2.97%) observed at 45°C, suggesting improved structural compactness and mineral retention at this temperature. Overall, 45°C was identified as the optimal extraction condition, balancing yield, purity, and stability of crude arabinoxylan. These findings indicate that temperature optimization is critical in maximizing the recovery and functional quality of breadfruit-derived arabinoxylan, providing a foundation for further refinement and potential food applications

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