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#### Chapter

# Kenaf (*Hibiscus cannabinus* L.) Seed Extract as a New Plant-Based Milk Alternative and Its Potential Food Uses

Roselina Karim, Nor Aini Mat Noh, Shafa'atu Giwa Ibrahim, Wan Zunairah Wan Ibadullah, Norhasnida Zawawi and Nazamid Saari

#### **Abstract**

Kenaf (Hibiscus cannabinus L.) seed is rich in protein, fat, fiber, and other essential nutrients. Kenaf seed comprises of high protein (22-31%) and oil (22–25%) contents which suggested its high potential food application. This chapter discusses the potential and early development of kenaf-based plant-milk and tofu. The step-by-step processes involved in preparation of kenaf-based milk and kenaf-based tofu at laboratory-scale are illustrated. Soaking conditions (temperature and time) of kenaf seed as pretreatment in preparation of kenaf seed milk were highlighted. Hydration of kenaf seed were found to be faster at elevated temperature, however higher soaking temperature and prolonged soaking time causes some losses of protein (%) and solid content (%) which are unfavorable for production of highly nutritious plant-based milk. Furthermore, in preparation of kenaf-based tofu, soaking temperature of seed also affected the properties of the tofu. As the soaking temperature was increased from 25–65°C, the yield, hardness, and chewiness of kenaf tofu decreased. It was recommended that soaking of kenaf seed at 25°C and the use of aluminum potassium salt at 1.00 g% as coagulant produces kenaf-based tofu with optimum quality.

**Keywords:** kenaf seed, soaking temperature, milky extract, hydration, physicochemical quality, chemical composition, kenaf-based tofu

#### 1. Introduction

Kenaf (*Hibiscus cannabinus* L.) is one of the most important fiber crops, belonging to the family of Malvaceae. Kenaf is cultivated in more than 20 countries where it is widely grown in China, India, and Thailand [1]. Kenaf is a useful multi-purpose crop with various industrial applications from paper to furniture and from biofuel to textile. Besides being a cordage crop it has substantial role in the construction and automotive industry. Kenaf plant has a significant economic value since all parts of the plant including the stem, leaves, flowers, and seeds can be used to the full advantage for mankind. In the recent

years, considerable attention and research has been carried out on the potential application of kenaf seeds in the food industry. Kenaf seed is reported as rich in fat, protein, dietary fibers, and a good source of raw material for production of edible oil, flour, protein concentrate, as well as potential ingredient for nutraceuticals and functional foods [2–4].

#### 2. Chemical composition

The proximate composition of different varieties of kenaf seed is presented in **Table 1**. The kenaf seed cultivars QP3, V36, and KB6 were cultivated in Malaysia while the seed cultivar C14 was cultivated in Korea, originated from Italy [2, 5, 6]. The seed varieties possess different chemical composition due to variation in agronomic conditions such as soil type, environmental variation, agriculture input (such as fertilizer application), planting season, maturity stage, harvesting period, and post-harvest treatments such as drying and storage conditions [2]. In general, kenaf seed is high in protein and fat content which comprises of 21.9–30.5% and 22.1–24.8%, respectively. Kenaf seed was also reported to have a considerable amount of carbohydrate (18.7–24.4%) and fibers (10.6–18.7%).

The fatty acid composition of kenaf milk was not affected by processing. Both the kenaf seed and the milk contain high concentration of hexadecanoic acid (palmitic acid), 9-octadecenoic acid (oleic acid) and 9,12-octadecadienoic acid (linoleic acid) (**Table 2**). These saturated and unsaturated fatty acids had been reported as the major fatty acids in other kenaf seed cultivars [7–9]. The kenaf seed and its milk were not significantly different in their saturated, mono- and poly-unsaturated fatty acids composition. Based on the 9-octadecenoic and 9,12-octadecadienoic acid contents, the oil from the kenaf seed and kenaf milk are less prone to rancidity and might be used as edible healthier oils. Oleic acid is suggested to show a protective effect on Alzheimer's disease and other neurological disorders. A study on mouse model supplemented with oleic acid and restricted cholesterol intake had reduced the Alzheimer's disease-type neuropathology [10]. Linoleic acid plays an important role in reducing the total cholesterol level. It was found that the presence of high linoleic acid (33.6%) in kenaf seed extract exhibited an anti-hypercholesterolemic effect [11]. The presence of these beneficial bioactive compounds suggested the potential nutraceutical properties of the kenaf seed and kenaf seed milk.

The amino acid profile of kenaf seed has not been well studied. The non-essential amino acids composition of kenaf seed were significantly higher than the kenaf milk except for the proline content (**Table 3**). Also, kenaf seed was significantly higher

Composition (%)	Kenaf QP3	Kenaf V36	Kenaf KB6	Kenaf C14
Moisture	8.5	8.4	9.0	8.3
Crude protein <sup>A</sup>	30.5	29.8	21.9	27.5
Crude fat	24.8	22.6	24.7	22.1
Crude fiber	12.5	11.5	18.7	10.6
Ash	4.5	4.5	6.2	5.8
Total carbohydrate <sup>B</sup>	19.2	23.2	18.7	24.4

<sup>&</sup>lt;sup>A</sup>Crude protein = N (%) × 6.25.

**Table 1.**Proximate composition of several kenaf seed varieties [2, 5, 6].

<sup>&</sup>lt;sup>B</sup>By difference.

Chemical name	Molecular formula	Kenaf seed area (%)	Kenaf milk area (%
Methyl tetradecanoate (15:0)	C <sub>15</sub> H <sub>30</sub> O <sub>2</sub>	$0.03 \pm 0.01^{b}$	$0.05 \pm 0.01^{a}$
Hexadecanoic acid (16:0)	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	26.71 ± 0.22 <sup>a</sup>	26.11 ± 0.57ª
9-Hexadecenoic acid (16:1)	C <sub>16</sub> H <sub>30</sub> O <sub>2</sub>	$0.40 \pm 0.02^{a}$	$0.28 \pm 0.01^{b}$
Heptadecanoic acid (17:0)	C <sub>17</sub> H <sub>34</sub> O <sub>2</sub>	0.04 ± 0.00 <sup>b</sup>	$0.05 \pm 0.00^{a}$
Octadecanoic acid (18:0)	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	3.17 ± 0.47 <sup>a</sup>	$2.94 \pm 0.05^{a}$
9-Octadecenoic acid (18:1)	C <sub>18</sub> H <sub>34</sub> O <sub>2</sub>	33.80 ± 2.39 <sup>a</sup>	40.41 ± 1.22 <sup>a</sup>
9,12-Octadecadienoic acid (18:2)	C <sub>18</sub> H <sub>32</sub> O <sub>2</sub>	30.59 ± 0.62 <sup>a</sup>	$28.62 \pm 0.86^{a}$
10-Nonadecenoic acid (20:1)	C <sub>20</sub> H <sub>38</sub> O <sub>2</sub>	$0.55 \pm 0.04^{b}$	$0.74 \pm 0.01^{a}$
Methyl 18-methylnonadecanoate (21:0)	$C_{21}H_{42}O_2$	$0.31 \pm 0.01^{a}$	$0.46 \pm 0.18^{a}$
Docosanoic acid (22:0)	$C_{22}H_{44}O_2$	$0.15 \pm 0.01^{a}$	$0.19 \pm 0.01^{a}$
Tetracosanoic acid (24:0)	$C_{24}H_{48}O_2$	$0.10 \pm 0.01^{a}$	$0.13 \pm 0.02^{a}$
Saturated		30.51 ± 0.73 <sup>a</sup>	29.93 ± 0.84 <sup>a</sup>
Monounsaturated		34.75 ± 2.45 <sup>a</sup>	41.12 ± 1.24 <sup>a</sup>
Polyunsaturated		$30.59 \pm 0.62^a$	28.62 ± 0.86 <sup>a</sup>

The values within the same row with different superscripts letters are significantly different at  $P \le 0.05$ .

**Table 2.**Composition and amount (%) of saturated and unsaturated fatty acids of kenaf seed KB6 variety.

Amino acids	Kenaf seed (mg/g)	Kenaf seed milk (mg/g)	
Asp	114.39 ± 4.29 <sup>a</sup>	94.21 ± 0.01 <sup>a</sup>	
Ser	87.97 ± 5.92 <sup>a</sup>	35.14 ± 0.01 <sup>b</sup>	
Glu	124.37 ± 2.87 <sup>a</sup>	58.77 ± 0.01°	
Arg	178.72 ± 0.89 <sup>a</sup>	70.79 ± 1.61 <sup>b</sup>	
Gly	62.21 ± 2.25 <sup>a</sup>	23.79 ± 0.01 <sup>b</sup>	
Ala	46.61 ± 4.33 <sup>a</sup>	14.70 ± 0.01 <sup>b</sup>	
Pro	35.69 ± 2.75 <sup>b</sup>	$54.31 \pm 0.01^{a}$	
Tyr	34.15 ± 0.46 <sup>a</sup>	11.04 ± 0.00 <sup>b</sup>	
Non-essential	684.11 <sup>a</sup>	362.75 <sup>b</sup>	
His	39.67 ± 2.35 <sup>a</sup>	17.79 ± 0.01 <sup>b</sup>	
Thr	54.65 ± 2.31 <sup>a</sup>	19.14 ± 0.01 <sup>b</sup>	
Lys	59.87 ± 1.47 <sup>b</sup>	$56.81 \pm 0.02^{b}$	
Val	64.11 ± 2.14 <sup>a</sup>	28.24 ± 0.01 <sup>c</sup>	
Met	28.09 ± 0.79 <sup>a</sup>	12.00 ± 0.00 <sup>b</sup>	
[le	54.07 ± 0.36 <sup>b</sup>	95.50 ± 1.35 <sup>a</sup>	
Leu	$91.32 \pm 0.62^{b}$ 129.59 ± 0.04 <sup>a</sup>		
Phe	$71.01 \pm 0.51^{b}$ 128.34 ± 0.04 <sup>a</sup>		
Essential	462.79 <sup>a</sup> 487.41 <sup>a</sup>		

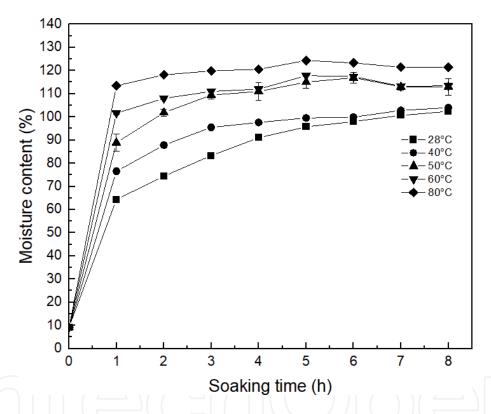
The values within the same row with different superscripts letters are significantly different at  $P \le 0.05$ .

**Table 3.** Amino acid profiles of KB6 kenaf seed and kenaf seed milk.

than the milk in terms of the essential amino acids such as histidine, threonine, valine and methionine. These indicated that the processes of extraction of the kenaf milk affected the amino acid content of the milk. However, the total essential amino acids of the seed and the milk were not significantly different.

#### 3. Seed hydration

Soaking is commonly used in processing of grains, to soften and hydrate the seed before proceeding to other stages such as cooking, extraction, fermentation, germination and malting [12]. In preparation of a plant-based milk alternative, an initial soaking of the plant seed is a prerequisite before extraction of the milk constituent by wet milling. Soaking of seeds reduced the hard-to-cook nature by softening the texture of the seeds which in turn facilitates subsequent processes and reduces the cooking time. Soaking is a batch process which can take an average



**Figure 1.**Moisture content (%) (d.b.) of kenaf seed soaked at different time and temperature.

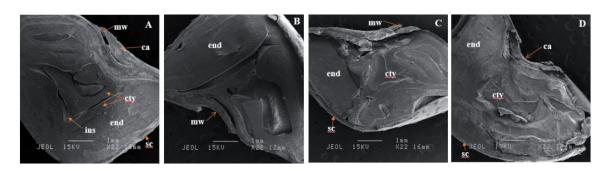


Figure 2. Effect of soaking temperatures on the scanning electron microscopy images of kenaf seeds. Note: A = unsoaked kenaf seed, B = kenaf seed soaked at 25°C, C = kenaf seed soaked at 45°C, D = kenaf seed soaked at 65°C, mw = major entrance of water, ca = caruncle, cty = cotyledon, end = endosperm, sc = seed coat, ins = intracellular spaces (adapted from [5]).

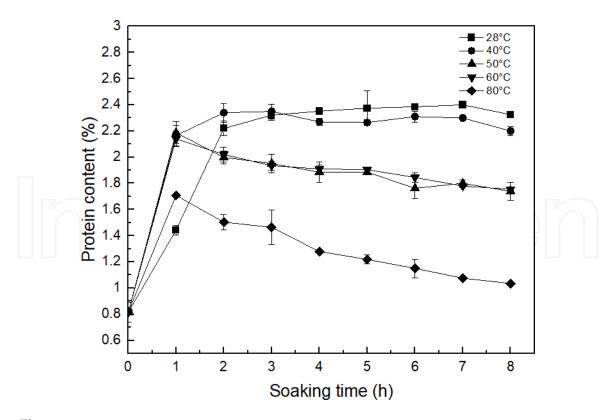
of 12 to 24 h at room temperature (25 + 2°C) and uses a substantial quantity of water. However, numerous ways to accelerate the process have been proposed which include soaking at elevated temperatures, high-power ultrasound, and high hydrostatic pressure [13].

Hydration at higher temperature is one of the most frequently used techniques in enhancing the hydration rate of plant seeds. **Figure 1** shows the water absorption rate of kenaf seed as a function of soaking temperature and time (unpublished data). It was observed that the highest water absorption rate of kenaf seed was recorded at the highest soaking temperature (80°C). Higher soaking temperature causes reduction in the soaking medium viscosity which improves the capillary flow and dilatation of the tissues and pores, hence increases the water absorption rate [14]. At the beginning of soaking, water absorption rate was high, and it gradually slowed down towards the end of soaking process. The former action was due to the predominant of capillarity flow of seed hydration mechanism while the latter was due to diffusion process [13]. The effectiveness of using higher temperature in increasing the absorption rate was also reported in other studies [15–18]. Further investigation on the effect of kenaf seed hydration towards its microstructure was studied using the scanning electron microscopy and is presented in Figure 2. It was observed that kenaf seed soaked at 25°C (Figure 2B) had a more visible intracellular spaces than the seed soaked at 45°C (**Figure 2C**), and 65°C (**Figure 2D**). The intracellular spaces inside the seeds soaked at 45°C and 65°C were filled with water and both the endosperm and cotyledon segment appeared expanded. Therefore, it is evident that hydration rate of seed soaked at higher temperature increased and causes further improvement in the capillarity flow and dilatation of the tissues and pores of kenaf seed.

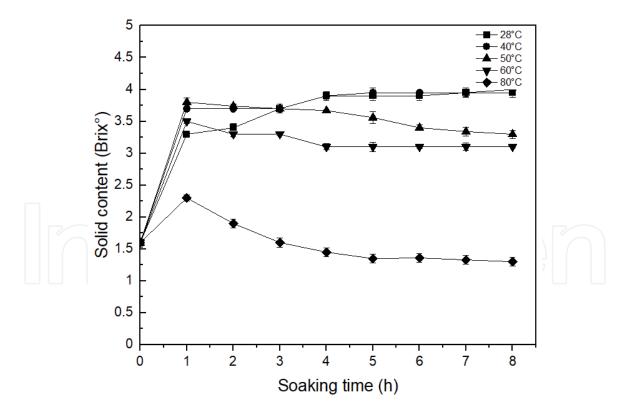
## 4. Effect of soaking condition on nutritional quality of kenaf seed milky extract

Although the use of high soaking temperature acts as a steeping factor for the soaking process, it generates a notable loss of solids from seeds such as protein (%) and solid content (%). Therefore, in preparation of kenaf seed milk, selection of suitable soaking conditions (temperature and time) need to be considered in minimizing the solid loss during soaking process.

Figures 3 and 4 show the effect of soaking temperature towards the subsequent protein content (%) and solid content (%) (expressed in °Brix) of the kenaf seed milky extract, respectively (unpublished data). From Figure 3, the protein content of kenaf seed milky extract were found to be increased with prolonged soaking time at lower soaking temperatures (28°C and 40°C) and decreased throughout the soaking period after the first hour of soaking at higher temperature (50°C, 60°C, 80°C). This result may be due to the release of some seed component including proteins into the soaking medium which is enhanced by the higher soaking temperature and time. Soaking at higher temperature causes the loose of seed coat structure and thus the seed storage proteins (7S and 11S proteins) that are protected by the seed coat were released into the soaking medium by the force of concentration difference [19]. Other than proteins, the leaching of the seed component may include carbohydrates [20]. The release of soluble solids from the seed were found to increase at higher soaking temperature due to the extended seed wall rupture. Based on Figure 4, the solid content of kenaf seed milky extract increased with longer soaking time of kenaf seed at lower temperatures (28°C and 40°C), and decreased at higher temperature (50°C, 60°C, 80°C) of soaking. The increased of solid loss upon higher temperature of soaking were also reported by other researchers [18, 20, 21].



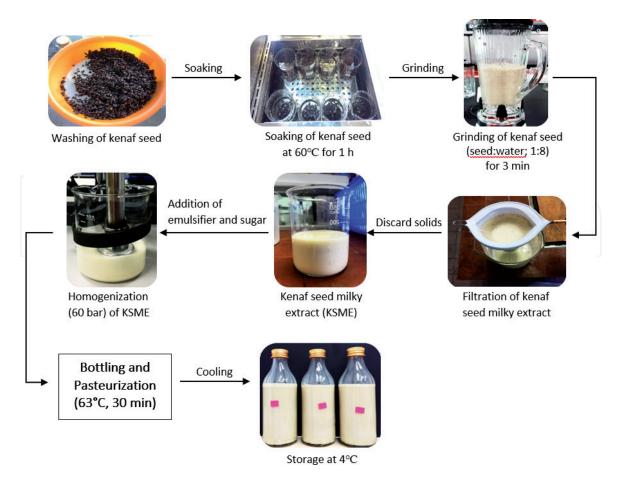
**Figure 3.**Protein content (%) of kenaf seed milky extract obtained from different soaking condition (time and temperature) of kenaf seed.



**Figure 4.**Solid content (°brix) of kenaf seed milky extract obtained using different soaking condition (time and temperature) of kenaf seed.

#### 5. Kenaf seed milk

**Figure 5** shows the procedures for the preparation of kenaf seed milk. Kenaf seed milk appears to be similar in appearance and texture to soymilk with creamy



**Figure 5.**Processes in preparation of kenaf seed milk.

Compositions	Kenaf seed milk	Soymilk [22, 23]	Almond milk [22, 23]	Hemp milk [24, 25]
Moisture (%)	91.04	88.12–91.00	72.00–93.40	91.60
Protein (%)	1.93–2.48	3.82–3.98	1.90–2.50	0.83-4.00
Fat content (%)	2.10–2.60	3.10-4.30	3.20–3.60	1.25–3.00
Carbohydrate (%)	1.82	4.64–4.92	4.30–4.70	2.50-20.00
Ash content (%)	3.11	0.84–0.81	0.09–3.04	0.47

**Table 4.**The proximate composition of kenaf seed milk (seed variety V36) and several commercially available plant-based milk alternatives.

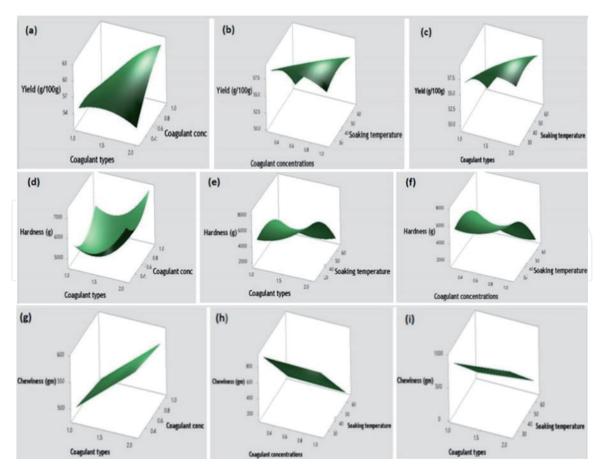
white in color. The taste of the unflavored kenaf seed milk is described as thin with a hint of earthy flavor. **Table 4** shows the proximate composition of kenaf milk in comparison to several commercially available plant-based milk alternatives. Kenaf seed milk contains 1.93–2.48% of protein and 2.10–2.60% fat which is comparable to almond milk in terms of the protein and fat contents, respectively.

## 6. Processing of value-added kenaf based tofu derived from kenaf seed milk

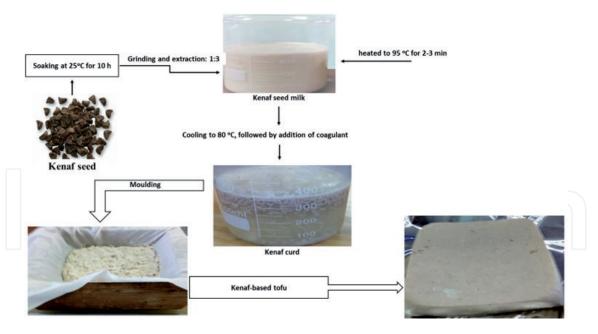
Tofu is a popular and important protein source in most countries such as Asian, Western and African countries [26, 27]. Over the years, soybean has been the most commonly used legume for tofu production. However, several researchers have

produced tofu from other grains such as peanut [28], lupin seed [29], sesame [30], chickpea and mung bean [31]; in most cases the selection of these grains were based on their high protein content and good functional properties. Kenaf seed have been reported to contain 21.4 to 30.5% protein on dry basis [6, 32] and protein has been known as one of the factors that affect tofu quality [29, 33, 34]. The general processing of soybean tofu involves soaking of the seed, grinding with water, boiling of the slurry, followed by separation of the milk from the residue (kenaf seed okara), cooking of the milk to 95°C, coagulating the cooked milk and molding [35]. Recently, we have produced kenaf-based tofu from kenaf seed milk and the effect of processing variables such as soaking temperature of kenaf seed, coagulant types and concentrations were investigated [5]. The outcomes of our research indicated that the yield, hardness, and chewiness of the kenaf-based tofu decreased as the soaking temperature increased from 25-65°C. Besides, the interaction among the processing variables such as coagulant type\*coagulant concentration, coagulant type\*soaking temperature and coagulant concentrations\*soaking temperature affected the yield, hardness, and chewiness of the kenaf-based tofu (Figure 6). It was discovered that soaking of kenaf seed at 25°C and using aluminum potassium salt as coagulant at concentration of 1.00 g% were recommended as the processing variables for optimum kenaf-based tofu production.

The procedure for kenaf-based tofu preparation slightly differs from the processing for soybean tofu whereby the kenaf seed slurry is not heated prior to extraction. Additionally, 1:5 to 1:8 soybean-to-water ratios were often used in the extraction of soybean milk for tofu production. However, in the case of kenaf-based



**Figure 6.**Surface plots of interactions of coagulant types, coagulant concentrations, and soaking temperature of seed on yield, hardness, and chewiness of kenaf-based tofu. Note: a, b, c = surface plots of yield; d, e, f = surface plots of hardness; g, h, i = surface plots of chewiness (adapted from [5]).



**Figure 7.**Procedures for production of kenaf seed tofu (adapted from [5]).

tofu it was discovered that the most suitable ratio of kenaf seed to water for the extraction of the milk during tofu production was 1:3, based on the lower total soluble solids content of kenaf milk (unpublished data). The procedure for the production of kenaf-based tofu is presented in **Figure 7**, the kenaf seed was soaked at 25°C for 10 h and the soaked seed was ground at low speed using 1:3 kenaf seed-to-water ratio for 2–3 min. The slurry obtained was then filtered using muslin cloth to separate the kenaf seed residue from the milk. The milk obtained (400 ml) from 100 g of kenaf seed was heated until the temperature of the milk reached 95°C and the temperature was hold at this temperature for 2–3 min. Then, the milk was allowed to cool to 80°C at room temperature (25 ± 2°C) and the specified coagulant was added to form the kenaf seed curd. The curd was then transferred to a wooden mold lined with muslin cloth and pressed with a load of 5 kg for 5 min to remove excess water. A solid curd-like product known as kenaf seed tofu is formed and is ready to be used or consumed.

#### 7. Other potential uses of kenaf seed

Several other potential food applications of kenaf seed have been postulated based on the functional characteristics of its protein concentrate [36]. The authors have proposed the use of kenaf seed as an ingredient in the production of vegetable-based protein substitute such as tempeh and texturized vegetable protein. Tempeh is a fermented vegetable meat substitute produced by inoculating pre-cooked legumes such as soybean, lentils and common bean with *Rhizopus oligosporus* [37, 38]. Similarly, pre-cooked kenaf seed can be fermented with *Rhizopus oligosporus* to produce kenaf-based tempeh based on its high protein content and emulsifying property (unpublished data). Additionally, kenaf seed protein concentrate has been reported to have high thermal stability [2]. Thus, protein concentrate from kenaf seed can be extruded into slices, crumbles, flakes and chunks to produce a meat-like chewy texture similar to texturized soy protein [39]. Also, kenaf seed has been used in the form of defatted meal or flour for the production of noodles and pasta [40].

#### 8. Conclusion

Kenaf seed is a highly nutritious plant seed that should be further exploited in innovation of non-conventional plant-based milk alternative and other food uses. In this chapter, the soaking conditions (temperature and time) of kenaf seed were studied. Higher soaking temperature and prolonged soaking time of kenaf seed were not recommended as they caused the loss of protein (%) and soluble solids (%) of the seed milky extract. Higher soaking temperature was also found to reduce the physical properties (yield, hardness, and chewiness) of kenaf-based tofu. It is recommended to use a lower temperature (25°C and 40°C) of soaking to preserve the nutritional value and quality of the subsequent products. Further and more thorough research ought to be done to produce a highly nutritious and highest quality of plant-based milk and tofu from kenaf seed that are comparable to the established soymilk and soy tofu, respectively.

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