



**Research Paper**

**CHEMICAL COMPOSITION OF DIFFERENT SELECTED TRADITIONAL SUDANESE FRUITS AND DIFFERENT *Bifidobacterium infantis* 20088 FERMENTED BEVERAGES**

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**Abstract**

This study was carried out to determine chemical composition of six selected traditional Sudanese fruits [Aradaib (*Tamarindus indica*); Dom (*Hyphaene thebaica*); Gudaim (*Grewia tenax*); Gunguleiz (*Adansonia digitata*); Lalob (*Balanitesa gyptiaca*); and Nabak (*Ziziphusspina\_christi*)] to develop probiotic fermented beverages. The seeds were separated from fruits except for Gudaim. The collected pulps of or fruits were ground into powder to pass through 250 micron sieve. The results on chemical composition of the selected traditional Sudanese fruits showed that carbohydrates ranged from a minimum of 63.8% in gudaim to a maximum of 80.9% in nabak. Moreover gudaim and dom were the highest sugar and fiber content therefore, they were used for fermentation with *Bifidobacterium infantis* 20088. 10% beverages were prepared from dom and godaim powder .2.5 % (w/w) skim milk was supplemented to each formulation to provide the required nutrient for bacteria growth during the fermentation. After sterilization and cooling, pH of the mixture was inoculated with a 10% culture of *B. Infants* followed by incubation for 36 h at 37 °C. The results revealed that initial proteins of both fermented products increased due to fermentation. However, the rate of protein increase was only significant ( $p < 0.05$ ) in fermented dom. The total carbohydrate and ash of both fermented godaim and bom besides the fiber of fermented dom did not revealed any significant ( $P < 0.05$ ) changes when compared to their levels at maximum growth of *B. infantis*. Nevertheless, there was significant ( $P < 0.05$ ) fiber reduction in fermented godaim. Therefore godaim and dom are suitable carrier to deliver *bifidobacterium infantis* to consumer at the same time the fermented beverages provide other essential nutrients such as protein, ash and fiber.

Key words: Composition, Sudanese, traditional, fruits, Bifidobacterium, beverage.

**INTRODUCTION**

Probiotic bacterium is defined as a living microorganism which when consumed in sufficient number will improve health beyond inherent basic nutrition [1]. Strain of *Bifidbacterium*, *lactobacillus* and non pathogenic yeast such as *Saccharomyces boulardii* are principally used individually or in combination as probiotics [2]. However, most human origin probiotics are

fastidious when used alone, they are characterized by low growth capability in food mediums including the dairy, the main a recommended carrier of probiotics to human [3].

There has been a considerable interest in the use of prebiotic to enhance the survivability and colonization of probiotic bacteria that are added in food products [4]. Because of the difficulty in maintaining a probiotic in the gastrointestinal tract, significant research has been focused in discovering which prebiotic is most beneficial in increasing the level of bifidobacteria in the gastrointestinal tract [4].

Probiotics formulation with prebiotics is a promising approach known as synbiotics (presence of probiotics and prebiotics at the same time in a product); which effectively enhance their complementary technological and beneficial qualities [5]. This is because probiotic foods should contain a sufficient number of viable microorganisms, which are able to alter the microflora composition of the host through colonization to generate more beneficial effect [1].

In many Africa countries, prebiotic effects of traditional fruits remain largely untapped. Sudan is one of the important country in Africa which include different ecological zones rich in rational fruits such as Aradaib (*Tamarindus indica*) , Dom (*Hyphaene thebaica*) ,Gunguleiz (*Adansonia digitata*), Lalob (*Balanites aegyptiaca*) , Nabak (*Ziziphus spina-Christi*) ,Godim (*Grewia tenax*). However, the economic value of traditional fruits in Sudan is still low; their utilizations did not go beyond small scale fresh beverages. These traditional fruits are known to provide dietary fiber and essential nutrients such as vitamins, and energy for active working societies. In addition they through to be beneficial against several diseases; however, studies are lacking to prove useful information to interpret their therapeutic potential.

In this respect, fermentation of Sudanese traditional fruits with probiotics, will lead to develop fermented beverages with further enhanced value and improved therapeutic properties, and extended shelf life by natural process of fermentation. This is because; the main carriers of probiotics are the dairy based products. However dairy products are expensive and not consumed by a big slice of population. This has paved the way for researchers to introduce new carriers for probiotics, such as soymilk and malted rice [6, 7]. Therefore, this study was carried out to determine the chemical composition of selected traditional Sudanese fruits and the fermented probiotic beverages.

### Preparation of tradition fruits for analysis

Selected traditional Sudanese fruits were used including: Aradaib (*Tamarindus indica*); Dom (*Hyphaenethebaica*; Gudeim (*Grewia tenax*); Gunguleiz (*Adansonia digitata*); Lalob (*Balanitesa gyptiaca*); and Nabak (*Ziziphusspina christi*). The fruits were purchased from a local market in Khartoum town randomly without grouping according to their ripeness. The traditional fruits were cleaned by removing the dirt and foreign materials. The seeds were separated from the fruits by gently hammering the fruits of gunguleiz, Dom and Nabak. Moreover, that of gunguleiz seed was separated directly from its pulp. Aradaib and lalob, fruit were manual separated from their seed after peeling the hard skin which covers the fruit using a knife .While godaim fruits were used without separation of seed. The collected pulps of fruits were ground into powder to pass through 250 micron sieve.

### Chemical composition

#### Determination of moisture content

Moisture was determined using thermal drying method, 2g of each fruits were heated under careful specified conditions (104 °c for 6 hours), and the loss of the weight was taken as a measure of the moisture content.

#### Determination of fat

Fat content was determined according to the official method of [8]. A sample of 5g was weighed into an extraction thimble and covered with cotton, and then extracted with hexane. Then, the thimble containing the sample and a pre-dried weighed extraction flask containing about 100 ml hexanes were attached to the extraction unit. The extraction process was conducted for 16 hr. At the end of the extraction period, the flask was disconnected from the unit and the solvent

was evaporated. Later, the flask with the remaining crude hexane extract was put in an oven, cooled to room temperature reweighed and the dried extract was registered as fat content.

#### **Determination of protein**

Protein was determined by Kjeldhal method according to the [8].

#### **Determination of crude fiber**

About 2 g of a defatted sample was placed into a conical flask containing 200ml of H<sub>2</sub>SO<sub>4</sub> (0.26N). The flask was fitted to a condenser and allowed to boil for minutes. At the end of the digestion period, the flask was removed and the digest was filtered through a porcelain filter crucible (No.3). After that, the precipitate was repeatedly rinsed with distilled boiled water followed by boiling in 200 ml NaOH (0.23 N) solution for 30 min under reflux condenser and the precipitate was filtered, rinsed with hot distilled water, 20 ml ethyl alcohol (96%) and 20 ml diethyl ether. Finally, the crucible was dried at 105 °C until a constant weight was obtained and the difference in weight was considered as crude fiber.

#### **Determination of ash and minerals content**

Ash content was determined by dry method. The fruits were weighed into crucible dish, and the organic matter was burned in a muffle furnace. The dish containing the residue was cooled in a desiccators and the amount of total ash was determined by weighing the remaining residues. Three minerals (Ca, Mg, and Fe) were determined by atomic absorption (Shim-pack CLC- NH<sub>2</sub>, SHIMADZU, Japan).

#### **Calculation of carbohydrates**

Carbohydrates were calculated by difference according to the following:

Total carbohydrates =

100% - [Moisture (%) + Protein (%) + Fat (%) + Ash (%) + and fiber (%)].

#### **Determination of sugar**

Different sugars including glucose, galactose, raffinose, fructose, lactose, maltose and sucrose had been analyzed. Fruits samples were extracted into warm water (80-90 °C) the extract was filtered to remove solids, and then was injected into HPLC instrument.

#### **Statistical analysis**

One- way analysis of variance (ANOVA) was performed to examine significant differences between normally distributed data. Tukey' s-test was used to perform multiple comparisons between means. Probability level of less than 0.05 was considered significant (p<0.05). All data were analyzed using MINITAB statistical software [9].

## **RESULTS AND DISCUSSION**

### **Chemical composition of the selected traditional Sudanese fruits**

Data on chemical composition of traditional Sudanese fruits were given in table 1.

Referring to our results in table 1, all the fruits have low moisture content ranged from 3.13% in dom to 12.06% in lalob. There was no significant (p>0.05) different in moisture content of aradaib and lalob (this range of moisture is normal for traditional fruits which scripted by long shelf life).

However these data were far below the results presented by other author. Moisture range of 35.18- 57.41% for nabak was reported [10]. This result may represent the moisture of wet fresh nabak. Moisture of 22.32% for lalob was also reported [11]. In general, these variations might be due to the variety of fruits and stage of harvesting.

Lipids have at least two important functions in foods: physiological, and nutritional. Among the six studied traditional Sudanese fruits the fat levels were relatively low which ranged from 0.1% in gunguleiz to its highest level of 2.6% in nabak. This value were close to the results obtained by [12], which was 0.2% fat for gunguleiz. However [10] reported slightly higher range of fat in nabak (3.58-4.02%). Moreover, there was no significant (p>0.05) different in fat content of aradaib, dom, godaim and lalob .

Protein is one of the main groups of nutrients in food. Our result in table 1 showed that some traditional fruits have low protein content of 2.1% such as in gunguleiz up to a high level of

8.3% in godaïum. [12, 13] reported similar values of protein content for gunguleiz and godaïum which were 2.6% and 8.7 respectively. Moreover, the protein content of gunguleiz and godaïum was significantly ( $p < 0.05$ ) different. While there was no significant difference in protein contents of other traditional fruits including: aradaib, lalob, nabag and godaïum. .

Crude fiber is widely used to determined insoluble material in dilute acid and dilute alkali under specified conditions. Table 1 showed that dom has the highest fiber content of 20.13% followed by gudaim, gunguleiz, aradaib, nabak and the lowest fiber content was found in lalob. [12, 14, 11] reported more lower fiber content for gunguleiz (5.7%), aradaib (2.3\_3.4%) and lalob (1.7%) respectively. This variation could be dependent on specific variety of each traditional fruits. The fiber level significantly ( $p < 0.05$ ) differ among the tested traditional fruits; however there was no significant difference in fiber of aradaib and nabak.

Total ash content is a useful parameter of the nutritional value of some foods and feeds. Referring to the result presented in table 1. Ash content of aradaib and lalob did not differ significantly ( $p > 0.05$ ). The results reported by [14] was slightly lower, he found ash value of 2.9% in aradaib which was in contrast to our finding. While [11] found higher ash value of 4.8% in lalob; which almost similar to our findings. Referring to Table 1 gunguleiz has the highest value of 7.7% ash. This value of ash was higher than that obtained by [12], which was 5.3 %. Such variation in nutrients concentration might be related to species varies cultivation regions, storages conditions and harvesting stage, it may also be due to geographical and climatic difference at where these trees are grown.

Carbohydrates are the most abundant and widely distributed food component, carbohydrate include monosaccharide, oligosaccharides and polysaccharides. Our results in table 1 also showed that total carbohydrate for the six fruits were high and ranged from 63.85% in godaïum to a maximum of 80.9% in nabak. Referring to the same table we could recognized that there was no significant ( $p > 0.05$ ) different in carbohydrates content of aradaib, gunguleiz and lalob showing values of 74.6%, 74.8%, and 76.96%, respectively. However, results on godim was slightly outside the range reported by [13], which was (66-84%) for three different species of godium. However, our results on carbohydrate content of nabak was within the range reported by [10], which was 76.67 - 86.12 % carbohydrate for different species of nabak. In general this high carbohydrate content of the traditional fruits is of nutritional interest.

### Sugar content

The results of sugar content presented in table 2 revealed that rhamnose was detected in nabak, aradaib and lalob. The statistical analysis showed significant ( $p > 0.05$ ) rhamnose differences level among the selected traditional Sudanese fruits (Table 2). Lalob contained the highest rhamnose level as compared to nabak and aradaib. In addition, there was no significant ( $p > 0.05$ ) difference in fructose content of dom and gunguleiz, which were the lowest fructose among the tested traditional fruits. Significant levels of fructose were noticed in nabak and aradaib. However, the highest fructose level was in lalob (Table 2).

Glucose content differ significantly ( $p > 0.05$ ) among the different traditional Sudanese fruits (Table 2) the highest glucose level was 15.9 mg/g recorded for lalob followed by nabak, godaïum, aradaib, gunguleiz and then dom in decreasing order. [15] reported similar value for dom which was 3.2mg/g. Referring to our result in table 2 we can notice that sucrose content range from 32.4mg/g in nabak to 0.2mg/g in godaïum. On the other hand maltose was not detected in selected traditional Sudanese fruits except lalob; which was negligible.

### Minerals content

The minerals content including Ca, Mg, and Fe are differ significant ( $p > 0.05$ ) among the traditional Sudanese fruits (Table 3). Calcium was at its highest level in gunguleiz, this value was lower as compared to 6.6 mg/g reported by [12] for the same fruits. The lowest Ca level among the traditional tested fruits was recorded in lalob. Moreover aradaib and dom have a similar Ca levels. While the levels of Ca in godaïum and dom were not within the range reported by [13, 15], which was 2.7-7.9 mg/g of Ca in some species of godaïum and 2.5mg/g in dom.

Table 3 also showed that Mg and Fe levels were both significantly differ ( $p > 0.05$ ) among the different traditional fruits Mg was in range of 0.01-0.38 mg/g. [15] Observed higher value of 2.4 mg/g for Mg in dom.

Fe was in range of 0.27-2.20 mg/g. The value of Fe in gunguleiz was higher as compared to the findings by [12]. They reported a lower Fe content of 0.09 mg/g in gunguleiz. [15] also reported lower level of Fe in dom which was 0.05mg/g as compared to our findings in table 3.

### Chemical composition of beverages fermented with *B.infantis*

Tables 4 and 5 show the chemical composition of godaim and doum beverages fermented with *B.infantis* at initial (0h) and at maximum growth time.

Referring to tables 4 and 5, it's clear that initial protein of both fermented products increased due to fermentation. However, the rate of protein increase was only significant ( $p < 0.05$ ) in fermented doum. [16] reported similar results; they found an increase in crude protein during fermentation. In general, the increase in protein content could be attributed to either microbial growth and synthesis or loss of non- protein materials.

The initial fat level of both fermented products decreased by fermentation to a maximum growth of *B. infantis*. The reduction was very slight in fermented godaim (Table 4 and 5).

The total carbohydrate and ash of both fermented goudaim and bom besides the fiber of fermented dom did not revealed any significant ( $P < 0.05$ ) changes when compared to their levels at maximum growth of *B. infantis*. Nevertheless, there was significant ( $P < 0.05$ ) fiber reduction in fermented godaim indicating potential prebiotics effect of godaim fruit.

**Table1: Chemical composition (%) of selected traditional Sudanese fruits\***

Type of fruit	Moisture	Fat	Protein	Fiber	Ash	Carbohydrate s
Aradaib	11.07±0.33 <sub>a</sub>	1.40±0.42 <sub>b</sub>	3.2±0.42 <sub>b</sub>	6.38±0.60 <sub>d</sub>	3.37±0.00 <sub>c</sub>	74.60±0.57 <sub>b</sub>
Dom	3.13± 0.24 <sub>d</sub>	1.45±0.35 <sub>b</sub>	2.2±0.28 <sub>c</sub>	20.13±0.32 <sub>a</sub>	4.12±0.01 <sub>b</sub>	70.00±0. 00 <sub>c</sub>
Godaim	7.25±0.11 <sub>b</sub>	1.10±0.00 <sub>b</sub>	8.30±0.57 <sub>a</sub>	16.83±0.04 <sub>b</sub>	2.68±0.06 <sub>d</sub>	63.85±0.70 <sub>d</sub>
Gungulei z	5.73±0.03 <sub>c</sub>	0.10±0.00 <sub>c</sub>	2.05±0.1 <sub>c</sub>	9.83±0.74 <sub>c</sub>	7.72±0. 00 <sub>a</sub>	74.58±0.70 <sub>b</sub>
Lalob	12.06±0.05 <sub>a</sub>	1.20±0.00 <sub>b</sub>	4.00±0.28 <sub>b</sub>	2.15±0. 07 <sub>e</sub>	3.63±0.29 <sub>b</sub> <sub>c</sub>	76.96±0.69 <sub>b</sub>
Nabak	4.96±0.03 <sub>c</sub>	2.55±0.07 <sub>a</sub>	4.05±0.21 <sub>b</sub>	4.850±0.8 <sub>d</sub>	2.72±0.03 <sub>d</sub>	80.9±0.62 <sub>a</sub>

\*Values are mean± STD of replicate independent analysis

\*Different superscript letters in the same column indicate significant ( $P < 0.05$ ) differences between means

**Table 2: Sugar content (mg/g) of selected traditional Sudanese fruits**

Type of fruit	Rhamnose	Fructose	Glucose	Sucrose	Maltose
Nabk	4.03± 0.21 <sub>b</sub>	12.57 ± 0.31 <sub>c</sub>	14.3± 0.15 <sub>a</sub>	32.07 ± .35 <sub>a</sub>	-
Dom	-	2.67 ± 0.21 <sub>d</sub>	3.13 ± 0.06 <sub>c</sub>	23.30± .46 <sub>b</sub>	-
Godaim	-	16.43± 0.35 <sub>b</sub>	13.20±0.20 <sub>a</sub>	0.23± 0.06 <sub>f</sub>	-
Aradaib	0.45± 0.20 <sub>c</sub>	12.47± 0.31 <sub>c</sub>	7.17± 0.12 <sub>b</sub>	1.47± 0.06 <sub>e</sub>	-
Lalob	8.27 ±0.21 <sub>a</sub>	20.27± 0.21 <sub>a</sub>	15.73±0.15 <sub>a</sub>	6.83± 0.31 <sub>d</sub>	0.53±0.06
Gunguleiz	-	2.63± 0.12 <sub>d</sub>	3.17± 0.12 <sub>c</sub>	9.13 ± 0.06 <sub>c</sub>	-

\* Values are mean± STD of duplicate independent analysis

\*Different superscript letters in the same column indicate significant ( $P < 0.05$ ) differences between means



**Table 3: Minerals content (mg/g) of selected traditional Sudanese fruit**

Components (mg/g)	Ca	Mg	Fe
Aradub	0.40 ± 0.00 <sup>c</sup>	0.38 ± 0.02 <sup>a</sup>	2.20 ± 0.35 <sup>a</sup>
Dom	0.42 ± 0.01 <sup>c</sup>	0.19 ± 0.00 <sup>b</sup>	0.43 ± 0.06 <sup>b</sup>
Godaim	0.79 ± 0.00 <sup>b</sup>	0.15 ± 0.00 <sup>c</sup>	0.57 ± 0.58 <sup>b</sup>
Gunguleiz	1.48 ± 0.01 <sup>a</sup>	0.23 ± 0.00 <sup>b</sup>	0.57 ± 0.06 <sup>b</sup>
Lalob	0.13 ± 0.00 <sup>e</sup>	0.01 ± 0.00 <sup>d</sup>	0.27 ± 0.06 <sup>c</sup>
Nabak	0.70 ± 0.00 <sup>b</sup>	0.15 ± 0.00 <sup>c</sup>	0.47 ± 0.08 <sup>b</sup>

\* Values are mean ± STD of duplicate independent runs

\*Different superscript letters in the same column indicate significant (P < 0.05) differences between means

**Table 4: Chemical composition of fermented godaim beverages at initial and maximum growth time.**

Component (%)	Initial growth time	Maximum growth time
Oil content	1.85 ± 0.07 <sup>a</sup>	1.25 ± 0.07 <sup>a</sup>
Protein content	10.00 ± 0.28 <sup>a</sup>	11.15 ± 0.07 <sup>a</sup>
Ash content	8.45 ± 0.07 <sup>a</sup>	7.40 ± 0.47 <sup>a</sup>
Fiber content	6.53 ± 0.16 <sup>a</sup>	3.16 ± 2.60 <sup>b</sup>
Carbohydrates	76.83 ± 0.60 <sup>a</sup>	72.55 ± 3.01 <sup>a</sup>

\*Data calculated on dry weight basis

\* Values are mean ± STD of duplicate independent runs

\*Different superscript letters in the same row indicate significant (P < 0.05) differences between means

**Table 5: Chemical composition of fermented dom beverages at initial and maximum growth time**

Component (%)	Initial growth time	Maximum growth time
Oil content	1.20 0.14 <sup>a</sup>	0.55 0.07 <sup>b</sup>
Protein content	4.70 0.28 <sup>a</sup>	6.60 0.42 <sup>b</sup>
Ash content	5.35 0.60 <sup>a</sup>	6.60 0.42 <sup>a</sup>
Fiber content	8.05 1.77 <sup>a</sup>	10.73 0.02 <sup>a</sup>
Carbohydrates	78.39 ± 1.67 <sup>a</sup>	78.64 ± 1.00 <sup>a</sup>

\*Data calculated on dry weight basis

\* Values are mean ± STD of duplicate independent runs

\*Different superscript letters in the same row indicate significant (P < 0.05) differences between means

## CONCLUSION

Our investigation on chemical composition of selected traditional Sudanese fruits, indicate that traditional Sudanese fruits are rich in nutrients and they could be important contributors to improve the nutritional content of the rural and urban people in Sudan. Furthermore, these fruits can be considered as a source of carbohydrates supplements, they are consumed by active societies in urban areas. They provide enough energy for different activities. The fruits have high fiber content which play important role in human intestinal tract. Fermentation of the selected traditional Sudanese fruits beverages with *Bifidobacterium infantis* and its high survival during refrigeration storage of the fermented beverages potentially proved that doum and godaim could be a good alternative carrier for *B. infantis* as compared to fresh dairy milk which is prices high in today's food markets of Sudan. Further, the fruits are available, cheap and easy to deliver since they are dry.

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