Proximate Composition of Moringa Oleifera Lam. from different Regions in Sudan

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Abstract:

Moringa oleifera Lam. (Moringaceae) is a very useful tree in tropical countries. In folklore and ayurvedic all parts of the tree used in different healing procedures for different diseases. The aim of this work was to contribute to our knowledge of the proximate analysis of M. oleifera (leaves) in Sudan. Proximate analyzes of M. oleifera from different regions in Sudan (Khartoum, Omdurman, Atbara and a private farm located in Sababi area Khartoum North, Sudan). The percentage of moisture, ash, Acid insoluble ashes, fibre, fat, Carbohydrate and protein in leaves of M. oleifera from Khartoum, Omdurman, Atbara and Sababi in Sudan was found to be 6.29 to 8.40, 7.14 to 9.12, 0.35 to 0.65, 9.01 to 10, 7.00 to 9.05, 36.28 to 43.90 and 24.47 to 30.05 %, respectively. The higher moisture overall, the contents of moisture, Acid insoluble ashes and fibre were found to be higher in the samples of leaves of Atbara region while lowest in Sababi region. These present results indicated that the leaves of M. oleifera analyzed from different regions, although having considerably different contribution of proximate parameters, can be explored as a potential source of valuable nutrients.

Key Words: Proximate Analysis, Moringa oleifera (leaves), Sudan

Introduction:

Moringa oleifera is one of the species of family Moringaceae. It is useful for healing as well as for curing of human diseases because of the presence of phytochemical constituents that produce a definite physiological action on the human body. Therefore, medicinal plants come into preparation of various drugs singly or in combination or even are used as the source of raw material for the other medicines (Hannah and Krishnakumari, 2015). In the Sudan, the rising cost of the protein rich feeds has encouraged search for protein sources to formulate adequate-least-cost diets for broiler which can satisfy the bird's requirements for maintenance and production. The incorporation of protein from leaf sources in diets for broilers is fast gaining grounds because of its availability, abundance and relatively reduced cost (El-Tazi, 2014). Moringa oleifera Lam.

(M. oleifera) is belonging to family Moringaceae (Sankhalkar and Vernekar, 2016). It is a fast-growing tropical edible tree (Elsayed et al., 2016). This plant is utilized from centuries and prescribed extensively in traditional medicine; it was mentioned in ancient Egyptian, Romans and Greeks (Raj et al., 2016). It is now distributed and cultivated as a crop in so many African, Asian, Latin America and Caribbean countries (Mridha, 2015). This plant is consumed as a popular food in some countries; it has high nutritional value, being a good source for proteins, vitamins and minerals, so it is used to treat malnutrition in rural regions (Gopalakrishnan et al., 2016). Leaves of M. oleifera have many phytochemical secondary metabolites of great pharmacological properties, such as alkaloids, flavonoids, saponins (Leone et al., 2015). All these metabolites were found to have antimicrobial

properties (Bindhu and Umadevi, 2015). Many medicinal uses were also reported, various parts of this plant employed as anti-inflammatory, anti-hypertensive, antioxidant, hepato-protective, anti-diabetic and antimicrobial (Arise et al., 2016). In Sudan, the rural citizens are traditionally use the powdered seeds of M. oleifera for purifying the drinking water, during this application a decrease in total bacterial count of this water was observed (Abdallah, 2016). Plants proximate analysis gives valuable information and help to access the quality of the sample. It provides information on moisture content, ash content, volatile matter, content, ash, fixed carbon etc. Ash is the inorganic residue remaining after water and organic matter has been removed by heating, which provides a measure of the total amount of minerals within the drug. Minerals are not destroyed by heating and they have a low volatility as compared to other food components. Total ash may vary within wide limits for specimen of genuine drugs due to variable natural or physiological ash. Ashes give us an idea of the mineral matter contained in a plant. Measuring it is important, because mineral matter may be the cause of a pharmacological effect (Grammelis et al., 2016). The aim of this study was to determine the proximate analysis of Moringa oleifera leaves.

Materials and Methods:

Plant Material:

M. oleifera (leaves) were collected during November to December 2016 from different regions in Sudan (Khartoum, Omdurman, Atbara and a private farm located in Sababi area Khartoum North, Sudan). They were identified and authenticating by the taxonomist Medicinal and Aromatic Plants and Traditional Medicine Research Institute, Khartoum, Sudan. Washed, shade, dried, powdered and ground with a pestle and mortar. The plant material was transferred to pre-weighed plastic containers and their weights were recorded at 100g.

Proximate Analysis:

Determination of moisture content:

The dry Moringa leaves were analyzed for moisture content using the methods described by the Food Chemical Codex (Codex, 1996). Each powdered sample was carefully weighed to (1.000 ± 0.001g) and placed in a pre- weighed foil envelope, pre-marked with the sample code. Each envelope was tightly sealed to avoid spillage and sample weight was recorded. The envelopes were placed in an oven at 80-90 oC for 5 days after which they were removed from the oven and immediately placed in a desiccator for 10 mins until the weight was constant, and cooled to room temperature. Subsequently, the samples were removed, the final masses recorded and the percent of moisture of each sample was calculated. This was done in triplicate.

Determination of Ash Content: Total Ashes and Acid Insoluble Ashes:

The method of analysis used was based upon prior work conducted in Moringa (Koroch et al., 2007). The crucibles were dried in the furnace at 400-600 oC for 5 hrs, cooled to room temperature and weighed. Powdered Moringa samples (2.000 ± 0.001 g) were transferred to each crucible, placed in a furnace and ignited at 650 oC for 5 hrs. After the crucibles were cooled to room temperature, the crucible (containing the sample) was removed from the furnace and placed in the desiccator for 10-20 mins. The weight of ash was recorded, and the ash percent calculated from the initial sample weight. To the weighed ashes, 25 mL of 2.7N HCl was added and boiled for 10-15 mins, the crucibles were allowed to cool, and the solution was filtered through ashless filter paper. The insoluble matter collected in the filter paper was washed with distilled water and after complete drainage, folded neatly inside its respective crucible and reheated in the furnace at 650 oC for 5 hrs. When the furnace cooled to room temperature, the crucibles were removed and placed in a desiccator for 10 mins. Each crucible was weighed, and the insoluble matter and percentage of acid insoluble ash of each sample was calculated. This was done in triplicate.

Determination of Fat Content:

Fat content was determined according to the method of AOAC (Williams, 1984) using soxhlet apparatus as follows: An empty clean and dry exhaustion flask was weighed, about 2g of sample was weighed and placed in a clean extraction thimble and covered with cotton wool. The thimble was placed in an extractor. Extraction was carried-out for 6-8 hours with petroleum ether. The heat was regulated to obtain least one siphoning per hour. The residual ether was dried by evaporation. The extraction was placed in an oven till it dried completely and then cooled in a desiccator and weighed. The fat content was calculated using the following equation:

$$FC\,(\%) = \frac{(W_2 - W_1)}{W} \times 100$$

Where: W= weight of sample.   W1= weight of extraction flask.   W2= weight of extraction flask with fat.

Determination of crude fiber contents:

Crude fibre was determined according to the method of AOAC, (Williams, 1984), using fibertic system 1010 heat extractor as follows: 1 g of defatted sample was accurately weighed into the fibre crucible, then transferred to the fibertic system 1010 heat extractor. The sample then digested with preheated H_2SO_4 (0.26 N) for 30 minutes. After washing the sample twice with hot water, the digestion was repeated using preheating KOH (0.23N) for 30 minutes. Again, the sample was washed with hot water. The crucible with sample was transferred to an oven adjusted to 1050 C and left over-night, then the sample was removed, allowed cool in a desiccator and weighed. Finally, the sample was ashed in the muffle- furnace at 5500 C for at least 3 hours allowed to cool and reweighed. The crude fibre (C.F. %) was calculated using the following equation:

$$C.F \% = \frac{(W_1 - W_2)}{Wt.\ S} \times 100$$

Where: W 1 = weight of crucible with sample before ashing. W 2 = weight of crucible with sample after ashing. Wt. S = weight of sample.

Determination of crude protein:

The crude protein was determined by using the micro-kjeldahal method according to AOAC (Williams, 1984) as follows:

a) **Digestion:** 2g of the sample was weighed and placed in small digestion flask (50ml), about 0.4 catalyst mixture (96% anhydrous sodium sulphate and 3.5% copper sulphate) was added. 3.5 ml of a proximately 98% v/w of H_2SO_4 was added. The content of the flask was heated on an electrical heater for 2 hr till the colour changes to blue- green. The tubes were removed from the digester and allowed to cool.

b) **Distillation:** The digested sample was transferred to the distillation unit and 20ml of 40% sodium hydroxide was added. The ammonia was received in 100ml conical flask containing 10ml of 2% boric acid plus 3-4 drops of methyl-red indicator. The distillation was continued until the volume reached 50 ml.

c) **Titration:** The content of the flask was titrated against 0.02 N HCl. The titration reading was recorded.

The crude protein was calculated using the following equation:

$$CP \ (\%) = \frac{(T-B) \times N \times 14 \quad 100 \times 6.25}{(St-B) \times Wt.\ S \times \quad 1000}$$

Where: T = Titration reading. B = Blank titration reading. St = Standard titration reading. N = HCl normality. Wt S = Sample weight. 1000 = to convert to mg.

Determination of carbohydrates (CHO):

The determination of Carbohydrates was calculated as follows:

CHO = 100 – (MC + AC +FC +FC +CP).
Where: C = Crude. M = Moisture. A = Ash. F = fat. F = fibre. P = Protein.
Statistical analysis:
All the results will be expressed as mean ± SEM Statistical analysis for all the assays results will be done using Microsoft Excel Program (2016).

Results and Discussion:

Proximate Analysis:
Proximate analysis is the description of proximate composition and nutritive value of foods and feeds (Sidhu et al., 2000). Protein, fiber and minerals are essential to life. The present work deals with the estimation of moisture, ash, crude fiber and crude protein contents of leave of M. oleifera harvested from three different regions in Sudan. The percentage of moisture, ash, Acid insoluble ashes, fibre, fat, Carbohydrate and protein in leaves of M. oleifera from Khartoum, Omdurman, Atbara and Sababi in Sudan was found to be 6.29 to 8.40, 7.14 to 9.12, 0.35 to 0.65, 9.01 to 10, 7.00 to 9.05, 36.28 to 43.90 and 24.47 to 30.05 %, respectively. The higher moisture Overall, the contents of moisture, Acid insoluble ashes and fibre were found to be higher in the samples of leaves of Atbara while lowest in Sababi region, show in Table (1) and Figure (9).

Determination of moisture content: The moisture content of M. oleifera leaves among the different regions (Khartoum, Omdurman, Sababi and Atbara) was found to be not significantly different ($P > 0.05$) ranged from (6.38 to 8.40 %). The moisture in leaves from Atbara was found to not significantly different (0.05) (8.31 ± 0.02) was higher than Omdurman (7.29 ± 0.02), Khartoum (6.70 ± 0.05) and Sababi (6.38 ± 0.03), show in Table (1) and Figures (2 & 9).

Determination of ash:
Ash contents of leaves of M. oleifera from different regions of Sudan were determined on the dried weight basis and the results reported as mean ± SD of triplicate experiments shown in Table (1) and Figures (3 & 9). The values of ash in M. oleifera leaves from the four regions (Khartoum, Omdurman, Sababi and Atbara) were statistically significantly different ($P < 0.05$) and were found range from 7.14 to 9.12 %. However, ash contents of leaves

Khartoum region was higher (9.12 ± 0.03) as compared to Atbara, Sababi and Omdurman regions.

Determination of acid insoluble ashes:

Acid insoluble ashes M. oleifera leaves from different regions of Sudan were determined on the dried weight basis and the results were reported as mean ± SD of triplicate experiments shown in Table (1) and Figures (4 & 9). The value of Acid insoluble ashes in the of M. oleifera leaves from was found to be significantly different among the four regions (Khartoum, Omdurman, Sababi and Atbara) ($P < 0.05$) and was to be found 0.35 to 0.65 %. The ash contents of leaves of Atbara region was higher (0.65 ± 0.02) as compared to Khartoum, Sababi and Omdurman regions.

Crude protein contents:

The amount of crude protein in leaves of M. oleifera from different regions of Sudan as given in Table (1) and Figure (5 & 9), was noted to be 24.47 to 30.05%. The overall region based variation of protein content in leaves of M. oleifera was found to be significantly different among the four regions ($P < 0.05$): Sababi samples > Atbara samples > Khartoum samples > Omdurman samples that might be attributed to the variable agroclimatic and geographical conditions of these regions.

Crude fiber contents: The value of crude fiber in the leaves of M. oleifera from four different regions (Khartoum, Omdurman, Sababi and Atbara) of Sudan was found to be 9.01 to 10 %. The contents of fibre in leaves of Khartoum and Sababi region were higher (10 ± 0.05) than those from other two regions studied. However statistically, there was no significant different among the samples show in Table (1) and Figure (6 & 9).

Crude fat and ash contents:

Fat contents of leaves of M. oleifera from different regions of Sudan were determined on the dried weight basis and the results reported as mean ± SD of triplicate experiments show in Table (1) and Figure (7 & 9). The value of fat in the leaves of M. oleifera from four regions (Khartoum, Omdurman, Sababi and Atbara) of Sudan was found to be 7.00 to 9.05 % which showed no statistical significance among the samples. Moreover, the ash content among the different samples was significantly different ($P < 0.05$). Moreover, the ash contents of leaves Sababi region was higher (9.12 ± 0.03) as compared to Atbara, Omdurman and Khartoum regions.

Carbohydrate contents:

The amount of Carbohydrate in leaves of M. oleifera from different regions of Sudan as given in Table (1) and Figure (8 & 9), was significantly different among the regions and ranging have 36.28 to 43.90%. The overall region based variation of Carbohydrate in leaves of M. oleifera was found to be: Omdurman samples > Khartoum samples > Sababi sampels > Atbara samples that might be attributed to the variable agroclimatic and geographical conditions of these regions. The Means of proximate composition of leaves of M. oliefera from different regions in Sudan. The percentage of moisture, ash, Acid insoluble ashes, fibre, fat, Carbohydrate and protein in leaves of M. oleifera from Sudan was found to be 7.19 ± 0.02, 8.0 ± 0.07, 0.46 ± 0.08, 9.54, 8.01 ±0.79, 39.52 ± 0.92 and 27.90 ± 0.41 % respectively, as given in Table (1) and Figure (9).

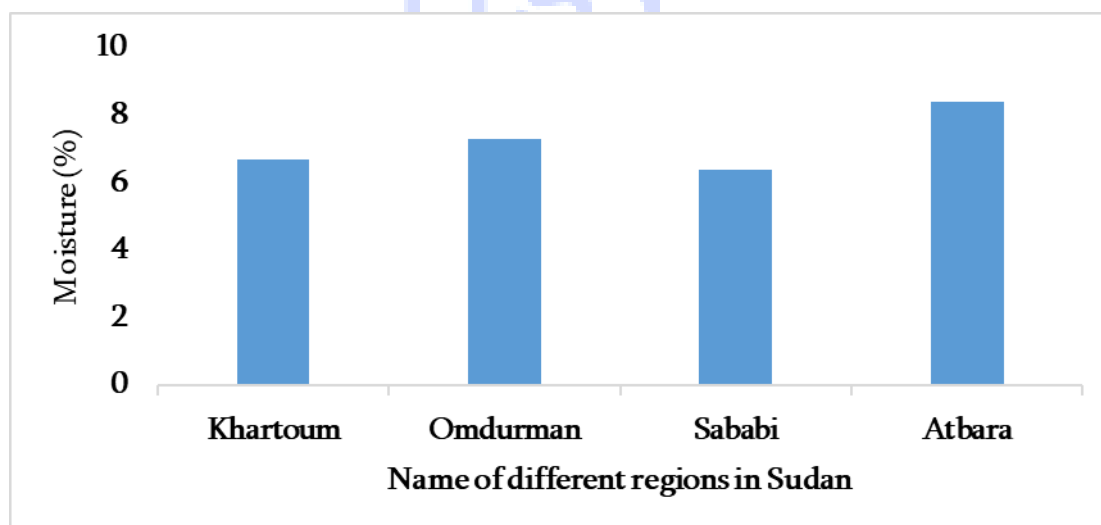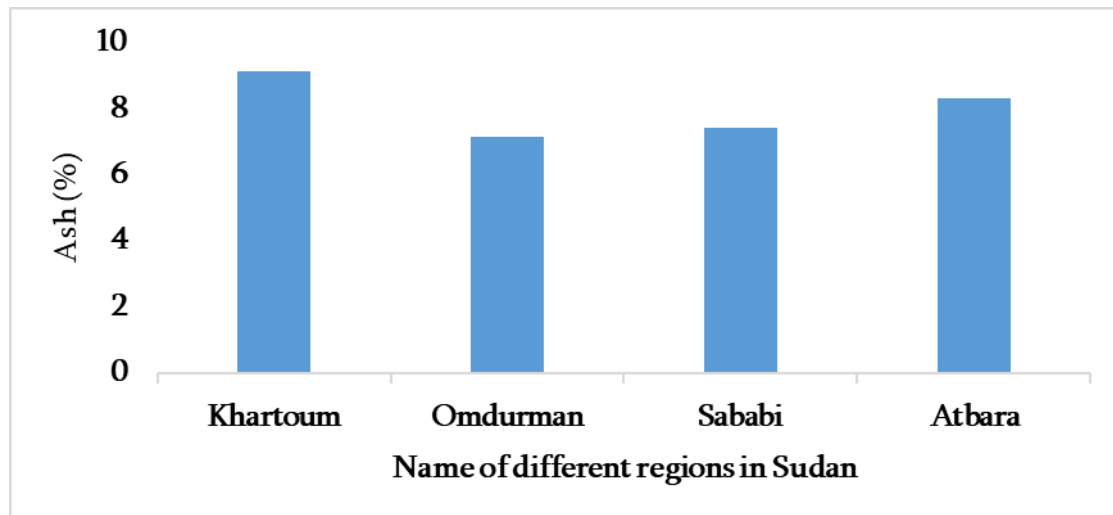**Figure 1:** Moisture (%) of leaves of M. oliefera from different regions in Sudan

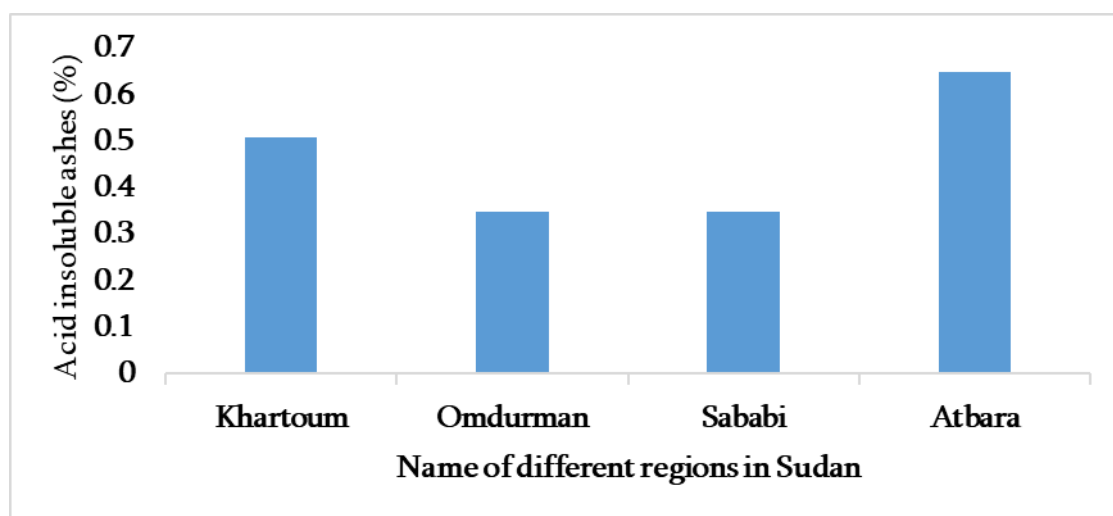**Figure 2:** Ash (%) of leaves of M. oliefera from different regions in Sudan

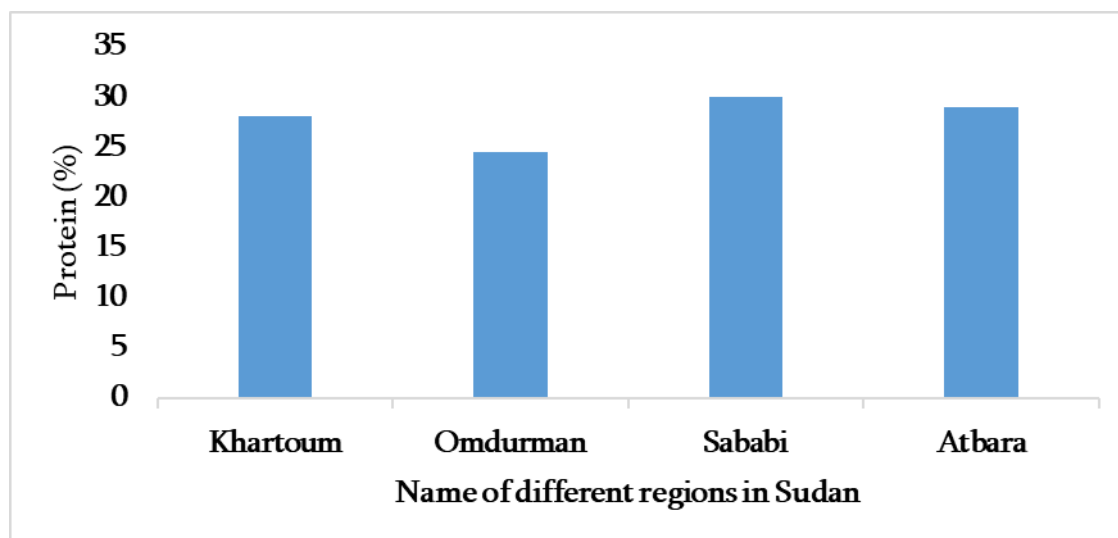**Figure 3:** Acid insoluble ashes (%) of leaves of M. oliefera from different regions in Sudan

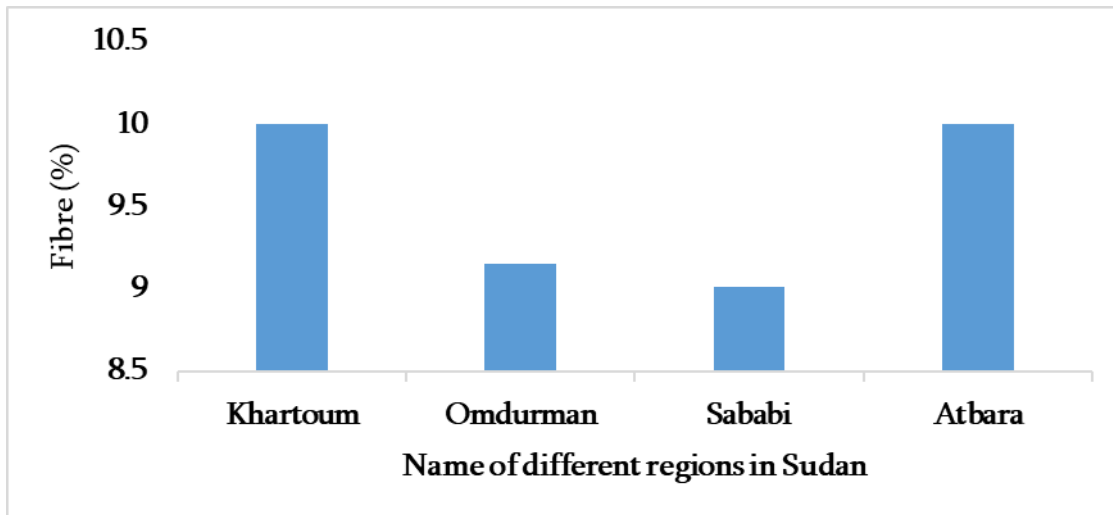**Figure 4:** Protein (%) of leaves of M. oliefera from different regions in Sudan

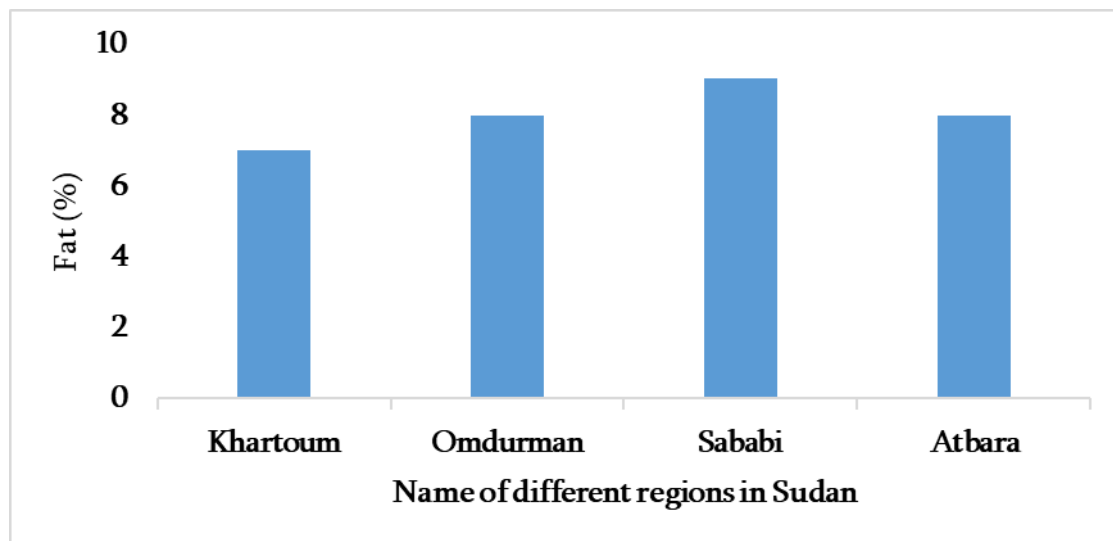**Figure 5:** Fibre (%) of leaves of M. oliefera from different regions in Sudan

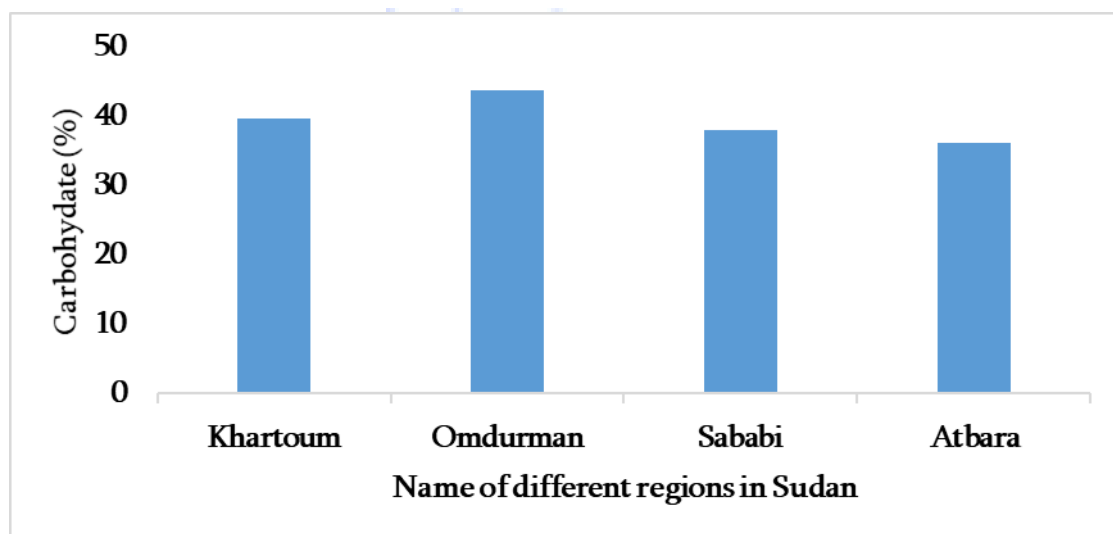**Figure 6:** Fat (%) of leaves of M. oliefera from different regions in Sudan

Figure 7: Carbohydrate (%) of leaves of M. oliefera from different regions in Sudan
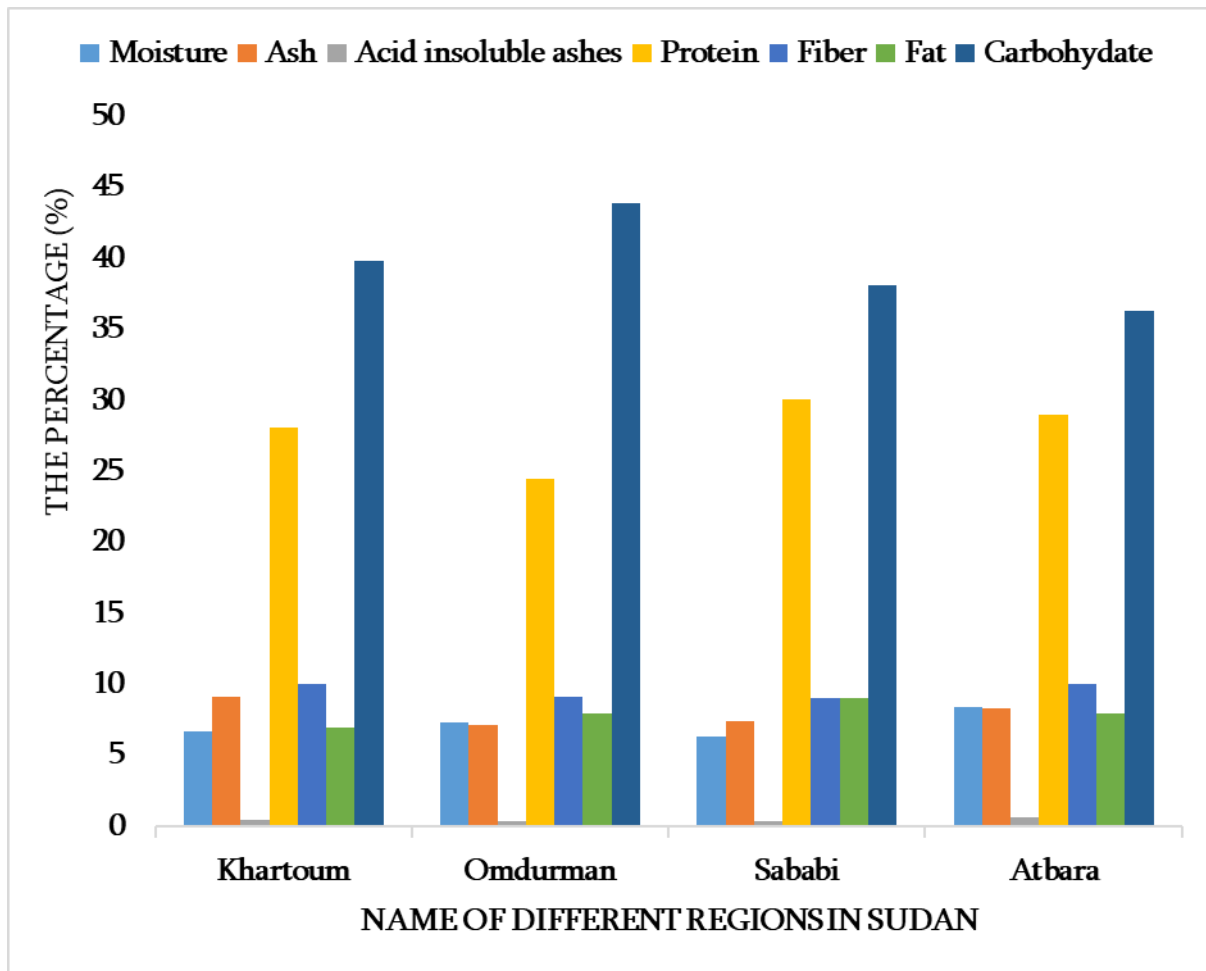
Figure 8: Comparison of proximate composition of leaves of M. oliefera from different regions in Sudan
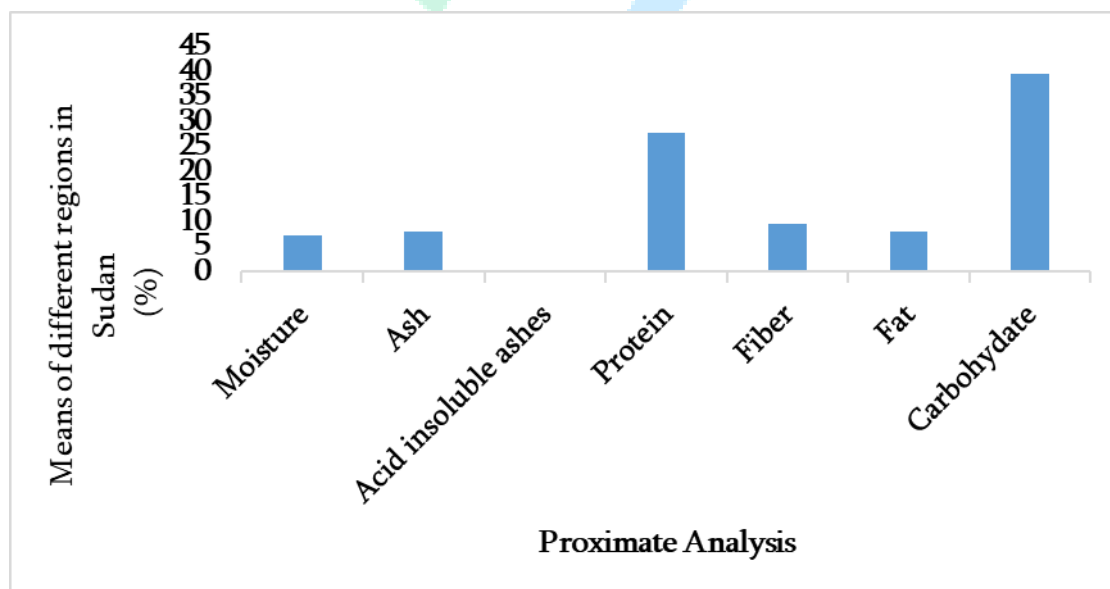

Figure 9: Means of proximate composition of leaves of M. oliefera from different regions in Sudan

Table 1: Comparison of proximate composition of leaves of M. oliefera from different regions in Sudan:

Content	Khartoum	Omdurman	Sababi	Atbara	Means ± SE
Moisture	6.70 ± 0.05	7.29 ± 0.02	6.38 ± 0.03	8.40 ± 0.02	7.19 ± 0.02
Ash	9.12 ± 0.03	7.14 ± 0.06	7.43 ± 0.01	8.32 ± 0.04	8.0 ± 0.07
Acid insoluble ashes	0.51 ± 0.01	0.35 ± 0.04	0.35 ± 0.07	0.65 ± 0.02	0.46 ± 0.08
Protein	28.1 ± 0.09	24.47 ± 0.03	30.05 ± 0.07	29 ± 0.02	27.90 ± 0.41
Fibre	10 ± 0.05	9.15 ± 0.01	9.01 ± 0.01	10 ± 0.05	9.54 ± 0.40
Fat	7.0 ± 0.02	8.0 ± 0.06	9.05 ± 0.03	8.0 ± 0.01	8.01 ± 0.79
Carbohydrate	39.8 ± 0.04	43.90 ± 0.08	38.08 ± 0.04	36.28 ± 0.06	39.52 ± 0.92

Key: Mean ± SE.

Conclusion:

The nutritional parameters explored in leaves of M. oliefera from different regions of Sudan (Khartoum, Omdurman, Sababi and Atbara) showed that leaves of this species can be potentially used as valuable ingredients for human nutrition. M. oliefera leaves are rich source of protein and carbohydrates with well balance of essential aminoacids. They can thus constitute a good source of many nutrients for people of less developed country which cannot find meat sufficiently every day. Overall, proximate parameters of M. oliefera varied slightly in relation to the parts tested while greatly among the regions studied.

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