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Review Paper

Chemotaxonomic study and botanical overview of some Ziziphus spp. in Sudan

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Abstract

The research aims to construct taxonomic keys for Ziziphus in Sudan, based on morphology and chemical components of fruits. This is by determination of the major chemical components of fruits of some species of the genus Ziziphus in the Sudan. The study also had surveyed the various uses of the genus in Sudanese rural areas. Plant materials were collected from different parts of Sudan, with special reference to the States of Gezira, Kordofan, Darfur and the White Nile. The collected plants were identified and synonymy was updated. Brief botanical species descriptions have been given with notes on geographical distribution and common uses. Fruits were chemically analyzed according to the methods adopted by the AOAC. These methods were used for the determination of the following contents: moisture, ash, crude, protein, fat, crude fiber, carbohydrates, reducing sugars, sucrose and the elements Ca, Mg, Fe, and P. High performance liquid chromatography (HPLC) was used for the determination of citric and ascorbic acid contents. Results of chemical data were analyzed using the variance ratio test (F-test) with a probability of 5%. The major findings of this study were that, five Ziziphus spp. were identified, namely: Z.spina-christi, Z.abyssinica, Z.mauritiana, Z.mucronata and Z.pubescens. Those species were multipurpose and have many uses such as food, medicine, rituals and often superstitions. Results of chemical analysis of three Ziziphus spp found a significant differences among the chemical components of Z.mauritiana, on one hand, and those of Z.spina-christi and Z.abyssinica on the other hand. These chemical variations could help to construct taxonomic keys for Ziziphus spp. in Sudan.

Keywords: *Ziziphus* spp, chemotaxonomic, botanical, Sudan.

Introduction

Indigenous fruit trees play a very important role in the livelihood of rural people, especially for those living in dry areas[1]. These fruits are important genetic resources in global efforts to maintain biodiversity[2], and improve the range of commodities available[3], withstand harsh conditions such as drought, heat and salinity stress[4].

Members of the genus *Ziziphus,* commonly referred to as jujubes, which are species of the genus *Ziziphus Tourn*. ex L., they belong to the family *Rhamnaceae*, named after the genus *Rhamnus*. The name *Ziziphus*is related to an Arabic word (zizoufo) used along the North African coast for *Z. lotus* (L.), Desf., but also related to the ancient Persian words *zizfum*or *zizafun* while ancient Greeks used the word *ziziphon* for the jujube.

There are two major domesticated jujubes namely: *Z.mauritiana* Lam., which is commonly known as the Indian jujube and *Z.jujube* Mill. which is commonly known as the Chinese or common jujube. There is a consensus that the genus contains about 135 species, while Liu and Cheng[5] suggested that there could be up to 70 species.

Sudan, as many other African countries, is endowed with a range of edapho-climatic conditions that favor the establishment of many plant species, such as *Z.spina-christi* and *Z.abyssinica* and *Z.mauritiana*, locally known as *Nabag* or *Nabak*. These are found to grow in central Sudan and the West States of the Sudan[6].

*Ziziphus*is is an important fruit species. The fleshy drupes of several *Ziziphus* species are rich in sugar and vitamins, and this fact has made *Ziziphus* spp. important fruit trees for many centuries. *Ziziphus* trees have a long tradition of selection and cultivation[7]. The fruits of all *Ziziphus* spp. are edible, dried or made into confectionary, or drunk as juice[8].

Some *Ziziphus* spp are used for the treatment of ulcers, wounds, eye diseases and bronchitis. Others used in medicine as an antiseptic, antifungal and anti-inflammatory agents, and for healing skin diseases such as dermatitis[9]. It also treat diarrhea, malaria, rheumatism and scorpion sting, heal swellings and treat intestinal spasms[10,11].

The seeds were rich in protein and the leaves in calcium, iron and magnesium[12]. The bitter-sweet pulp of fruit is dried and milled to produce a fine flour[13]. Fruits of *Z.mauritiana* have

higher contents of protein and vitamin C than apples[14]. It is processed in chips, paste, candy and preserved food[15].

Reasons for the complexity of the taxonomy of this genus are its wide geographical distribution and hybridization[16].

For the Muslims people seeds are used as rosaries, while for Christians the thorns are reported to have made up the crown of thorns of Jesus. In natural religions, the roots are used in superstitious practices[17]. Fruits and leaves of *Z.abyssinica. A. Rich.* are eagerly eaten by antelope, monkeys, baboons and especially by birds[18]. Fruits of *Z.mauritiana* Lam. are very nutritious and are usually eaten fresh[19,1].

Materials and methods

The plant material was collected from *Wadrawah* (Central Sudan), Darfur (Western Sudan) and Kordofan (Western Sudan), during the period December 2008 to March 2009.

A preliminary identification of the collected plant material was made using Andrews method[20]. The identification was later checked using the methods followed by EL Amin[21], El Awad[22], Von Maydell[23], and Sahni[24].

The identification was them confirmed by comparison with identified herbarium material from the herbaria of Faculty of Science, University of Khartoum; Medicinal and Aromatic Plant Research Institute (MAPRI), Khartoum, and Forest Research Centre at Soba, Khartoum.

Taxonomic keys have been constructed for the identified *Ziziphus* species based on morphological characters.

The fruit pulps and coats of the identified *Ziziphus* species were removed and the seeds were then manually separated from the pulp. All samples were milled, mixed, packaged in polyethylene bags and kept in a freezer. Then all parameters were calculated as percent %.

The moisture content, the total ash and the crude fiber content of each sample of *Ziziphus* spp. were determined according to AOAC[25]. The crude protein was determined by the semi-micro-kjeldahl method described by Pearson[26]. The fat content of *Ziziphus* spp. was determined according to the method of Pearson[26], by extracting the dry sample with petroleum ether at 60 – 80^{o}C in a continuous Soxhlet extraction apparatus.

Total carbohydrates were determined by Pomeran and Meloan[27] and calculated as percent by the following equation:
Carbohydrates % = 100 – (CP%+CF%+FC%+Ash%+MC%)

Where**:** CP = Crude protein; CF = Crude fiber; FC = Fat content; MC = Moisture content.

Reducing sugars and total sugars of *Ziziphus* spp. were determined by the modified Schneier method described in the ICUMSA[28] (extraction by Soxhlet Apparatus), in the titration, Fehling solution used as an indicator, the reducing sugars were calculated from the Lane-Eynon table[29]. Total sugars were also calculated from the invert sugar table. Sucrose content of *Ziziphus* spp. was determined by methods in ICUMSA[28] and calculated from the Lane-Eynon table according to the following equation:

Sucrose % = total sugars % - reducing sugars %.

For elemental analysis the plant samples were prepared according to the method described by Chapman and Prett[30] by placing the crucible in a muffle furnace (550^{o}C) for 2 hours. Iron, calcium, magnesium and phosphorus were determined according to the method described by Stewart[31], using an atomic absorption spectrophotometer (Perkin Elmer, model 2330). Different concentrations of standards were used. The organic acids content was determined by the High-Performance Liquid Chromatography (HPLC) according to the method described by Marsil *et al.*[32]. HPLC (Hewlett Packard 1050) ultraviolet/ visible detector column laypersil ODS sum (4.0×250mm) was used. Acentonitrile/water (15:25/V:UL were used as mobile phase). Analytical grade organic acids were used as standards. Grade acetonitrile was used in sample preparation.

Chemical data were analyzed using the variance ratio test (F-test) with a probability of 5% (P=0.05).

Results and discussion

Botanical overview: Botanical overview are given in the following subheadings:
Ziziphus species in Sudan: A field survey of the genus *Ziziphus* in the Sudan revealed that members of the genus occur in many parts of the Sudan but they are of wide occurrence in the States of Kordofan, Darfur and Gezira. Five species were identified namely: *Z.spina-christi, Z.abyssinica* and *Z.mauritiana*, *Z.mucronata* and *Z.pubescens*.

Members of the genus *Ziziphus,* commonly referred to as jujubes, belong to family *Rhamnaceae*. In this study, 5 *Ziziphus* spp. were identified namely: *Z.spina-christi*, *Z.abyssinica, Z.mauritiana*, *Z.mucronata and Z.pubescens*. The first three species of these had been further studied for chemical constituents.

The following is a brief outline for three of the *Ziziphus* spp. identified in this study.

***Z. spina-christi* (L.) Desf.:** Syn: *Z.africana* Mill.; *Z.amphibia* A. Chev.; Z.nabeca (Forssk.). Lam.; *Z.inermis* A. Chev.; *Z.sphaerocarpa* Tul.; *Z.spina-christi* Willd (as used by Polunin and Huxley)[33].

Vernacular names: Common English name: Christ's thorn. Common Arabic names: Nabaj, Nabak or Nabag (fruit), Sidir (tree).

Botanical description: Shrubs or small trees, 10-15m tall; Leaves ovate-lanceolate or ovate-elliptic, Fruits sub globose to globose drupes, yellow, reddish or red-brown, usually 2x1cm; fruit pulp astringent to taste. This species has two varieties: i. *var.spina-christi* (L.). Desf., ii. *var.* microphylla A. Rich.

Distribution: The tree is found in central Sudan.

Common Uses**:** Fruits are used as food especially by people in western and central Sudan. Fruits are used to treat some diseases like diarrhea, malaria and as an antispasmodic. The honey collected from the flowers is an excellent flavor.

Z.abyssinica A. Rich.: Syn. Z.abyssinica A. Rich.; *Z.atacorensis* A. Chev.; *Z.bagurmiae.* A. Chev.; Zxylopyrus Hochst. Ex A. Rich.

Vernacular names: Common English name: jujube. Common Arabic names: bemba, Nabaj.

Botanical description: Trees or shrub or climbers, 1.8-8m; Leaves ovate to broadly ovate, alternate, up to 8×4.7in, Flowers small, star-shaped, creamy to yellowish–green, with an unpleasant sharp smell. Fruit almost spherical, 2-3cm in diameter, shiny red or reddish brown when mature, smooth, containing 1 to 2 light brown glossy seeds inside the inner stone.

Distribution: In Darfur (J.Mara), Kordofan (Nuba Mts), Upper Nile, (Shambe), Bahr El Ghazal, Blue Nile (Roseires) and East Equatoria.

Common uses: The Fruit pulp is sweet and hence fruits are used to make porridge and meal in western Sudan. Fresh leaves are chewed and applied to wounds, boils and sores used to reduce pain. The root bark is used as an emetic, febrifuge and laxative. *Z.abyssinica* is a source of firewood and it is used in the production of charcoal.

***Z.mauritiana* Lam.:** Syn: *Z. jujube* (L.) Lam.; *Z. jujuba*(L.) Gaertn; *Z. tomentosa* Poir; *Z. rotunadata* DC.; *Z. aucheri* Boiss; *Z. insularis Smith*; *Z. sororia* Roem. and Schult.; and *Z. orthocantha* DC.

Vernacular names: Common English names: bear tree, ber, Chinese apple, Chinese date, Indian jujube, Indian cherry and dunks. Common Arabic names: nabak (fruit), Kanar Sidr (tree).

Botanical description: Shrubs, small or medium trees, 3-4 or 10-16m tall. Leaves elliptic to ovate or nearly orbicular. Fruits glabrous, globose or oval edible drupes, ca 2.1cm across and may reach 5×3cm; pulp acidic, sweet, yellow or sometimes reddish.

Distribution: Widespread on low lying ground near river beds or water depressions in the tall grass savanna.

Common uses**:** Fruits are eaten fresh or crushed in water forming a very popular cold drink.

The leaves are used as fodder for animals. Fruits are used internally in the treatment of a range of conditions including chronic fatigue and loss of appetite.

***Ziziphus* Tourn. ex L. Key to the species (based on morphology):** The following is a brief outline:

i. Unarmed trees or shrubs; bark rough; leaves elliptic or oblanceolate-elliptic, pubescent; fruit ellipsoid, yellow or red-yellow, up to 1cm across…… ……..*Z.pubescens.*

A. Armed trees or shrubs; bark scaly or deeply furrowed or deeply fissured; leaves mostly ovate or obovate or lanceolate, usually glabrous; fruit globose or spherical, reddish or reddish brown , more than 1cm across:

ii. Bark deeply furrowed; leaf-base asymmetrical; flowers with sharp unpleasant smell; fruit spherical, shiny red.…… *Z.abyssinica.*

B. Bark scaly or deeply fissured; leaf-base symmetrical; flowers not as above; fruit globose, reddish or reddish-brown:

iii. Bark scaly; sepals wooly above; fruit pulp astringent to taste ……..…..*Z.spina-christi* .

C. Bark deeply fissured; sepals not as above; fruit pulp sweet – acidic or bitter :

iv. Bark reddish or greyish–brown; fruit sweet–acidic, edible …….*Z.mauritiana.*

D. Bark dark-grey; fruit bitter, unedible…….*Z.mucronata.*

Chemical Studies: Chemical studies are given in the following subheadings:

The fruits of *Z.spina-christi* {from Kordofan}, *Z.abyssinica* {from Darfur} and *Z.mauritiana* {from Wadrawah} have been chemically analyzed for a number of chemical components. (Table-1, Figure-1 and Figure-2).

***Ziziphus* Tourn. ex L.: Key to the species (based on chemical components):** Moisture content 80%; fat 3%; total carbohydrates 8%; reducing sugars 6%; sucrose 2%; ascorbic acid 19%…….*Z.mauritiana*.

Moisture content ca 5.5%; fat < 1%; total carbohydrates > 82%; reducing sugars > 21%; sucrose > 19%; ascorbic acid 8-12%; or ascorbic acid < 9%..……..*Z.spina-christi*

Ascorbic acid > 12%...……..……...*Z.abyssinica*.

Significant differences were found among the three *Ziziphus* spp regarding all the analyzed parameters. *Z.mauritaina* had the highest ash content, and the highest moisture content (80.3%). The highest crude protein content was found in *Z.spina-christi* (4.1%) and the lowest was in *Z.mauritiana* (2.90%). The *Z.spina-christi* had the highest fat content (0.90%) whereas *Z.mauritiana* had the lowest (0.30%). The highest fiber content was found in *Z.mauritiana* (3.20%) and the lowest found in *Z.abyssinica*.

However, the ash content for all of the *Ziziphus* spp studied was higher than values reported by some authors such as Palmer[34], Duke[35], Morton[36] and Grosskinsky[37]. The ash content of *Z.spina-christi* (Kordofan) was close to values reported by other authors, Duke[35], Abdelgalil and Eljissry[38] and Abdelmuti[39] reported (4.4%), Nour *et .al.*[40] reported (3.5%).

Table-1. Summary of results of the chemical Analysis of fruits of *Z. spina-christi*, *Z.abyssinca* and *Z.mauritiona.*

Sample / Parameter	*Ziziphusspina-christi*, Kordofan	*Z.abyssinica*, Darfur	*Z.mauritiana*, Gezira
Ash %	4.3 g/100g	4.2 g/100g	5.1*g/100g
Moisture%	5.4g/100g	5.5 g/100g	80.3*g/100g
Crude protein%	4.1* g/100g	3.95 g/100g	2.9 g/100g
Fat %	0.9* g/100g	0.8 g/100g	0.3 g/100g
Fiber %	2.5 g/100g	2.4 g/100g	g 3.2* g/100g
Carbohydrates%	% 82.7 g/100g	83.00* g/100g	8.2 g/100g
Reducing sugars%	% 22.6* g/100g	21.5 g/100g	6.00 g/100g
Sucrose %	% 21.7* g/100g	19.4 g/100g	1.9 g/100g
Ca %	0.85 g/100g	1.25* g/100g	0.80 g/100g
Mg %	0.50* g/100g	0.45 g/100g	0.225 g/100g
Fe %	0.0377* g/100g	0.0192 g/100g	0.0242 g/100g
P %	0.28 g/100g	0.32 g/100g	0.44* g/100g
Ascorbic acid mg/100g	8.64mg/100g	12.24mg/100g	18.66mg/100g
Citric acid mg/100g	0.4133 mg/100g	0.6164*mg/100g	0.4203 g/100g

*Significant at P = 0.05.

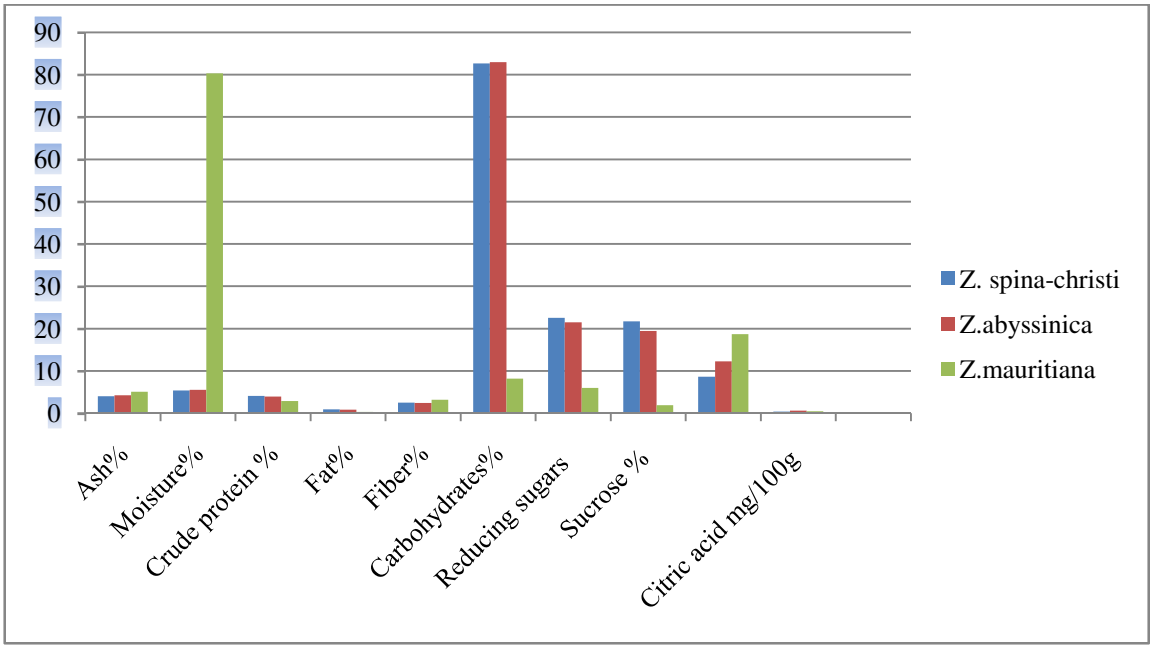

Figure-1: Summary of results of the chemical analysis of fruits of *Z.spina-christi*, *Z.abyssinca* and *Z.mauritiona.*

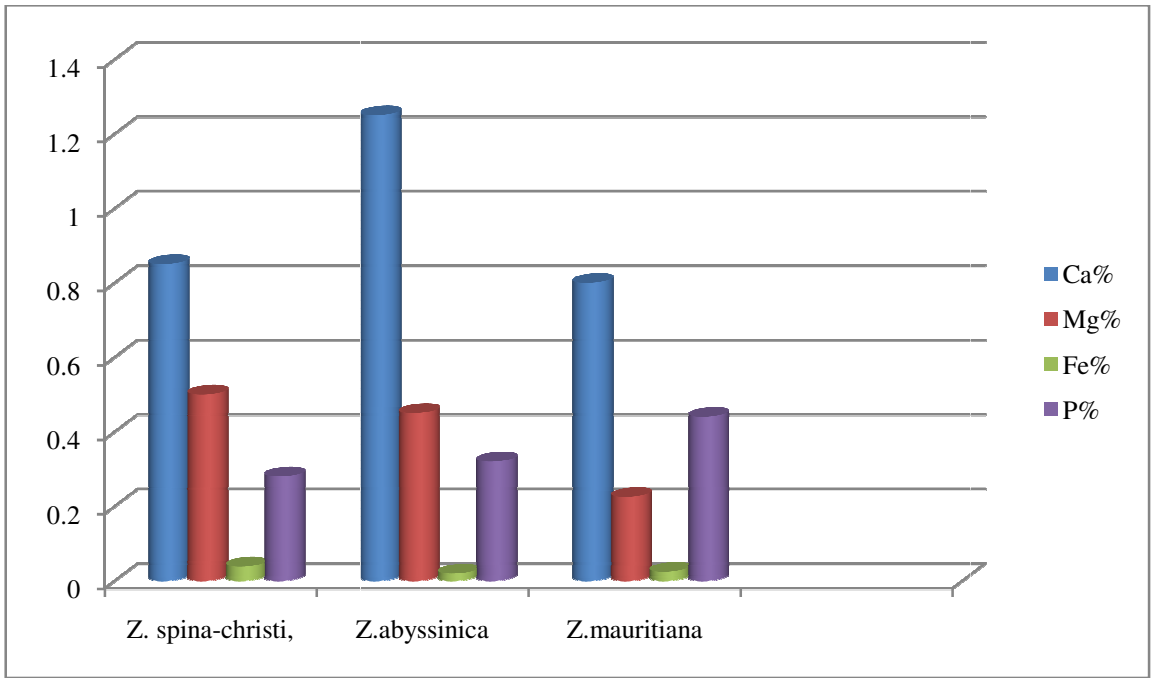

Figure-2: Mineral composition of fruits of *Z.spina-christi*, *Z.abyssinica* and *Z.mauritiana.*

The moisture contents of *Z.spina-christi* {Kordofan} (5.4%), and *Z.abyssinica* {Darfur} (5.5%), these results were comparable to that reported by Nour *et al.*[40] who found that *Z.spina-christi* had a moisture content of 6.00%. The moisture content of *Z.mauritiana,* {Gezira} was in agreement with those of Morton[36] and Pareek[41] who found values of 81%, and 81.6% - 83% respectively.

The crude protein content of *Z.spina-christi* {Kordofan} was in agreement with Duke[35] and Abdelgalil and Eljissry[38] who found values of 4.8% and 4.8 – 5.6% respectively. Other authors such as Abdelmuti[39] and Getachewo[42] reported lower values of 2.8% and 3.2% respectively for the crude protein of this species. *Z.abyssinica* Darfur had a crude protein of 3.95%, a value which was higher than values reported by Rittner and Reel[43] (1.4%) and Benhura and Katayi-Chedewe[44] (2.6%), respectively.

The fat content of *Z.spina-christi* Kordofan was similar to that reported by Duke[35], Berry *et al.*[45] and Abdelgalil and Eljissry[38] who each reported a value of 0.9%. This value is higher than that cited by Nour *et al.*[40] who found a value of 0.8%, but it is lower than that of Getachew[42] who found a value of 1.1%. The fat content of *Z.abyssinica* Darfur was 0.80%, this value was higher than values reported by Babayemi[46] and Russel[47] who reported values of 0.50% and 0.20%, respectively.

The fiber content of *Z.mauritiana* Gezira was in agreement with Duke[35] and Grossinky[37] who reported values of 4% and 3.4% respectively. However, the fiber content of this species was higher than results reported by Morton[36] and Li *et al.*[15] who found values of 1.2% and 2.8% respectively.

The highest value of Carbohydrates was found in *Z.abyssinica* (83.00%) and the lowest was found in *Z.mauritiana* (8.20%). The total sugars contents ranged from (7.9%) *Z.mauritiana* to 45.5% in *Z.spina-christi*. The *Z.spina-christi* had the highest content of reducing sugars (22.60%) while *Z.mauritiana* had the lowest (6.00%). The highest sucrose content was in *Z.spina-christi* (21.7%) whereas the lowest was in *Z.mauritian*.

The total carbohydrates of *Z.spina-christi* Kordofan (82.70%), were more or less comparable to those reported by Duke[35], Nour *et al.*[40] and Abdelmuti[39] and Getachew[42] who found values of 80.9%, 86.1%, 80.1%, and 80.7% respectively. The total carbohydrate of *Z.mauritiana* Gezira (8.2%) was comparable to that of Li *et al.*[15] who reported a value of 8.8% for this species.

The total sugars content for *Z.spina-christi* Kordofan was in agreement with results of Nour *et al.*[40] and Abdelmuti[39] who each found a value of 47.6% for this species. The total sugars content of *Z.mauritiana* Gezira (7.90%) is within the range 3 - 9.6% reported by Pareek[41]. The total reducing sugars content of *Z.spina-christi* Kordofan was in agreement with those of Nour *et al.*[40], Abdelgalil and Eljissry[38] and Abdelmuti[39].

The total reducing sugars content of *Z.mauritiana*, Gezira (9.7%) was comparable to values reported by Morton[36] (1.4 – 6.2%), Chovatia *et al.*[48] (5.1%), and Bal[49] (5.6%).

The sucrose content of *Z.spina-christi* Kordofan was comparable to values reported by Nour *et al.*[40], Abdelgalil and Eljissry[38] and Abdelmuti[39] who found values of 21.8%, 21.8% 21.8%, respectively. The sucrose content of *Z.amuritiana*, Gezira (1.9%) was lower than results of Morton[36] and Chovatia *et al.*[48] who found values of 15.4% and 5.7%, respectively.

Z.abyssinica had the highest Ca content (1.25%) whereas *Z.mauritiana* had the lowest (0.80%). *Z.spina-christi* had the highest Mg content (0.50%), whereas *Z.mauritiana* had the lowest (0.225%). The highest Fe content was in *Z.spina-christi* (0.377%) and the lowest was in *Z.abyssinica* (0.0192%). *Z.mauritiana* had the highest P content (0.44%), whereas *Z.spina-christi* had the lowest (0.28%).

This result of Ca content was in agreement with that of Nour *et al*.[40], Abdelgalil and Eljissry[38] and Abdelmuti[39] who found values of 1.4%, 0.61%, 0.6% and 0.6% respectively. However, this value was lower than findings reported by Duke[35], and Berry *et al*.[45] who reported values of 1.4%, and 1.4% respectively.

Furthermore, the Ca content of this species was much lower than results reported by Morton[36] and Grosskinsky[37] who found values of 2.5% and 2.1% respectively.

The Mg content of *Z.spina-christi* Kordofan was higher than that reported by Abdelgalil and Eljissry[38] and Abdelmuti[39] who found values of 0.12% and 0.10% respectively.

Z.mauritiana Wadrawah was found to contain 0.225% Mg, a finding that was in agreement with Grosskinsky[37] who found a value of 0.3%.

The Fe content of *Z.spina-christi* Kordofan was comparable to values reported by Duke[35], Berry *et al*.[45] and Abdelgalil and Eljissry[38] who reported values of 0.03%, 0.02%, and 0.03% respectively. However, the Fe content of *Z.mauritiana* Gezira was lower than that reported by Grosskinsky[37].

The P content *Z.spina-christi* Kordofan was the lowest compared to values reported by Nour *et al*.[40] who found a value of 1.06%. P content of *Z.mauritiana* Gezira (0.44%) was lower than that reported by Duke[35] who found a P content of 1.6%. However, this value agreed with values found by Morton[36] and Grosskinsky[37] (0.26% and 0.56%), respectively.

Two organic acids were found in the three *Ziziphus* spp. namely: ascorbic acid and citric acid. The highest content of ascorbic acid was found in *Z.mauritiana*, while the lowest was found in *Z.spina-christi*. The citric acid content of *Z.abyssinica* was (0.6164 mg/100g), while that of *Z.spina-christi* was 0.4133.

The ascorbic acid content of *Z.spina-christi* Kordofan was lower than values reported by Duke[35], Abdelgalil and Eljissry[38], Abdelmuti[39] and Getachwo[42] who found values of 30mg, 30 mg, 30mg and 35mg, respectively for the ascorbic acid of this species.

The ascorbic acid of *Z.mauritiana*, Gezira was 18.66mg, a value which was much lower than values reported by Bal and Mann[49], Duke[35], Morton[36] and Grosskinsky[37] who found values of 70-165mg/100g, 30mg, 66–76mg/100g, and 24mg/100g, respectively.

The citric acid content of *Z.mauritina* Gezira was found to be 0.4203 mg. This value lies within the range (0.2 – 1.1 mg) reported by Morton[36].

Conclusion

The differences of chemical components between the three studied *Ziziphus* species (*Z.spina-christi* *Z.abyssinica* *Z. mauritiana*) were almost significant. The most significant difference was among the chemical components of *Z.mauritiana*, on one hand, and those of *Z.spina-christi* and *Z.abyssinica* on the other hand. Results obtained by this study were not far from previous results obtained by other studies either those conducted in Sudan or elsewhere. This study provides additional data about the composition of the Indigenous plant in Sudan "*Nabag*", and can be used to enhance the utilization of the plant.

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