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Comparative Study of the Physicochemical Properties of Zizyphus Spina and Moringa Oleifera Seed Oils

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Abstract: Oils of *Zyziphus* spina (*Z. spina*) and Moringa *oleifera* (*M. oleifera*) seeds were analyzed for physicochemical, phytochemical and lipid composition. *M. oleifera* seeds gave oil yield of 40.47% while *Z. Spina* seeds gave 30%. Refractive index, density and acid value (mg KOH g^{-1}) of *M. oleifera* oil were 1.471, 0.81, 1.16 while Z .spina had 1.441, 0.79, 7.57 respectively. Similarly, Iodine (Wijs), Saponification (mg KOH g^{-1}) and Peroxide (meq kg-1) values obtained for *M. oleifera* oil were 46.50, 166.77 and 2.16 whereas *Z. spina* had 86.40, 184.00 and traces respectively. The phytochemical analysis showed the presence of secondary metabolites such as saponin, flavonoids, terpenoid, sterol and balsam in M. *oleifera* and alkaloids, flavonoid, terpenoid, sterol and resin in *Z. spina*. Lipid composition showed the presence of neutral lipids, glycolipid and phospholipid in both *Z. spina* and *M. oleifera*.

Keywords: *Z. Spina*, *M. Oleifera*, Oil, Physico-Chemical and Phytochemical

1. Introduction

Plant seeds are important sources of edible oils and oils used as industrials raw materials as in the production of cosmetics, paint and biodiesel. The physicochemical characteristics of oils from different sources suggest it use either as edible oil or feed stock for industrial use. Consumers are having increased awareness on the health effect of oils; therefore the increased search for oils with considerable amount of monounsaturated and polyunsaturated fatty acids [1]. *Moringa oleifera* plant seeds are known to be good source of oil, it is a leafy plant belonging to the family *Moringaceae* species, native [2, 3]. Virtually all the parts of the plant have being reported to be useful for nutritional, industrial and medicinal purposes [4, 5, 6]. Studies have shown that the seeds of *M.oleifera* contain about 37- 40% oil also known as Ben oil or Behen oil and various result of the physicochemical study of the oil have being reported [7, 8]. *Ziziphus spina-christi* is an edible wild fruit which serves as food for very long time. It is a multipurpose tree species, and belongs to the family Rhaminaceae [9, 10]. It is a thorny plant with tiny fruits. It grows well in the Northern region of Nigeria and it is locally called 'Kurna'. The plant has been reported to possess unique nutritional and medicinal characteristics [11]. Report has shown the seed of *Z. spina* contains about 32% oil. This study was carried out to compares the physicochemical properties of *Z.spina-christi* seed oil with *M. oleifera* seed oil, for possible use as excipent in cosmetics or as nutraceuticles.

2. Materials and Methods

The fruits of *Zizyphus spina* were purchased from the

market in Jigawa State, North West Nigeria. After the removal of the pulp from the fruit, the pods were crushed to release the seeds. The seeds were then pulverized and stored in a polyethene bag. *Moringa oleifera* seeds were harvested from the plantation in Sheda Science and Technology Complex, Kwali Area Council Abuja, Nigeria. The dried seeds were remove from the pods and dehusked. They were further air- dried for a few days and pulverized. The oils from the two seeds were extracted separately using n-hexane as reported by [7].

3. Physico Chemical Properties of Oil

3.1. Analysis of Extracted Oil

The following parameters were determined: density, refractive index, iodine value, peroxide value, acidity, saponification value, and unsaponifiable matter of the extracted oil was carried out as described by [12] while the refractive index of the oils were determined on Abbey Refractometer.

3.2. Separation of Lipid Profile and Fatty Acid Distribution

The oils were separated into the different lipid classes (neutral lipids, glycolipids and phospholipids) on a 1 g scale using silica gel open column chromatography described in [1] and [13]; Neutral lipids, glycolipids, and phospholipid were eluted successively using hexane: ethyl acetate (9:1, v/v) as developing solvent for neutral lipid, chloroform: methanol: water (65:25:4 v/v), for glycosides and phospholipid. The eluted samples were identified using by placing the plates in an iodine tank for 5 minutes. Spots were outlined and the R_f (ratio to front) values were calculated and compared with those of the standards

3.3. Phytochemical Analysis of Oil Samples

The phytochemical analyses of the oil samples were determined as described in [14]. The presence or absence of the following plant secondary metabolites were determined alkaloids, phenols, sterols, terpenes, tannins, flavonoids, cardiac glycosides, saponins:

Phenols: Equal volumes of each extract and ferric chloride solution (which is prepared by dissolving 135.2g of $FeCl_3.6H_2O$ in distilled water containing 20 ml of concentrated HCl dilute to 1 litre) are added together. A deep bluish green precipitate indicates the presence of phenol.

3.3.1. Alkaloids

Each extract was added to 1% aqueous HCl over water bath and filtered. The filtrate was treated with (2g of Iodine in 6g of Potassium iodide in 100 ml of distilled water) .Formation brown or reddish brown precipitate indicates presence of alkaloids.

3.3.2. Steroids

Each Extract was added to 2ml acetic anhydride and 2ml H_2SO_4. Colour change from violet to blue or green indicates the presence of steroids.

3.3.3. Terpenes

Each Extract was added to 0.5ml acetic anhydride and few drops of concentrated H_2SO_4. A bluish green precipitate indicates the presence of terpenes.

3.3.4. Cardiac Glycosides

Extract was treated with 2ml glacial acetic acid with a drop of Ferric Chloride solution and underplayed with 1ml H_2SO_4. A browning at the interface indicates the presence of cardiac glycosides.

3.3.5. Tannins

Each Extract was boiled in 20ml water and filtered. A few drops of 0.1% Ferric Chloride solution were added. Brownish green or blue-black color indicates the presence of Tannins.

3.3.6. Flavonoids

5ml Ammonium solution was added to aqueous filtrate of each extract and then few drops of concentrated H_2SO_4. Yellow coloration indicates the presence of Flavonoids.

3.3.7. Saponins

1g each extract was boiled with 5ml distilled water and filtered. 3ml distilled water was added to the filtrate and shaken vigorously for 5 minutes. Persistent frothing on warming indicates the presence of Saponins.

3.3.8. Triterpenoids

Crude extract was mixed with chloroform and few drops of conc. H_2SO_4 was added, shaken and allowed to stand for some time. The formation of a yellow colored layer indicates the presence of triterpenoids.

3.3.9. Glycosides

5ml H_2SO_4 was added to each of the test extract in a boiling tube. The mixture was heated in boiling water for 15minutes. Fehling's solution A and B was added and the resulting mixture was heated to boiling. A brick red precipitates indicates the presence of glycosides.

3.3.10. Carbohydrates

5ml of the equal mixture of both fehling's solutions A and B was added to 2ml of test extract in a boiling tube, this was heated for 2 minutes. A brick red precipitates indicates a positive result.

3.3.11. Phlobatanins

A few drops of 1% HCl was added to 1ml of test extract and was boiled. A reddish precipitates indicates the presence of phlobatanins.

3.3.12. Resins

2ml of test extract plus an equal volume of acetic anhydride solution with drops of conc. H_2SO_4 gives a colophony resins, a violet color indicates the presence of resins.

3.3.13. Balsams

3 drops of alcoholic $FeCl_3$ was added to 4ml of extract which was warmed. A dark green coloration indicates the presence of balsams.

3.3.14. Volatile Oil

A small quantity of the test extract was shaken with dilute NaOH and 0.1M HCl. The formation of a white precipitate indicates a positive result.

4. Result and Discussion

The oil yield from *M. oleifera* seeds was 41.47%. The *M. oleifera* oil was light golden yellow in color, liquid at room temperature with palatable flavor while *Z. spina* is bright brownish yellow, semi-solid at room temperature with a yield of 30.0 %. The physico-chemical properties of the oils revealed a significant difference between *M. oleifera* oil and *Z. spina* oil in terms of their saponification, peroxide, iodine and acid value. *M. oleifera* oil had lower values for most of the properties compare to *Z. spina* except for peroxide value (Table 1). Their refractive index and density are close. Phytochemical analysis shows the presence of flavonoid, sterol and terpenoid in both *M. oleifera* and Z. *Spina* while balsam and saponin was found in *M. oleifera* alone and *Z. spina* showed the presence of alkaloid and resin (Table 2). The lipid distribution and classes showed the presence of neutral lipid, glycolipid and phospholipid (Table 3).

Table 1 shows the physicochemical result of *Ziziphu*s *spina* seed oil and *Moringa oleifera* seed oil. The result for both oils falls within the standard range for the oil. The acid value of *Z. spina* is 7.57mg KOH g^{-1} while that of *M. oleifera* is 1.16mg KOH g^{-1}. Acid value is an important index of the physicochemical property of oil which is used for to indicate the quality, age, edibility and suitability for use such as paint, it also used to measure the extent to which the triglycerides in oil has been decomposed by lipase and other physical factors such as heat and light. The observed result for *Z. spina* contrasts the value reported by [9] which is 1.19 mg KOH g^{-1}. The high acid value observed for *Z. spina* in this work may be a result of other external factors. The result for *M. oleifera* is comparable with the report of [8].

The saponification value for the oils is 184.00mg KOH g^{-1} and 166.77mg KOH g^{-1} for *Z. spina* and *M. oleifera* oil respectively. The higher saponification value for *Z. spina* is an indication that it will be better oil for soap making.

The moisture/volatile compound content of *Z. spina* is shown to be 18.44%. The expected moisture/volatile matter content of oil is expected to be about 0.2% [12]. This is high could make the oil more susceptible to oxidation [15].

The iodine value for *Z. spina* (86.64 wijs) is slightly lower than 88.0 wijs that reported by [16] and 117.38 wij's reported by [9]. The iodine value is the measure of the level of unsaturation and could also be used to quantify the amount of double bonds present in the oil which reflects the susceptibility of oil to oxidation. The observed result in this study suggests *Z. spina* oil is susceptible to oxidation than *M. oleifera* oil which recorded iodine value as 46.56 wijs.

The peroxide value is a measure of rancidity of the oil; this was present in a trace amount in *Z. spina*. This could be as result of the oil being freshly extracted and analyzed immediately without any appreciable oxidation taking place. The peroxide value for the *M. oleifera* (2.16meq/Kg) is within the standard value. The other physical properties results which include density, refractive index is also shown in Table 1. Some of the chemical properties reported in this study are closely related to the result presented by [9] 2016 for *Z. spina* and [8] *M. oleifera.*

Table 1. *Physicochemical result of Zyziphus spina and Moringa oleifera.*

Parameter	Z. spina (found)	M. oleifera(found)
Acid value mg KOH g^{-1}	7.57±1.5	1.16±0.13
Saponification value mg KOH g^{-1}	184.00±0.5	166.77±0.15
Unsaponifiable matter g Kg^{-1}	6.70±0.5	1.40±0.12
Moisture content/volatile matter %	18.44±0.10	0.890±0.10
Iodine wijs	86.64± 3.52	46.56±4.25
Peroxide value meq Kg^{-1}	Traces	0.8±0.13
Density g cm^{-3}	0.79± 0.1	0.81±0.1
Refractive index	1.44±0.1	1.43±0.1
Free fatty acids mg KOH g^{-1}	3.79±0.75	0.58±0.065
Color	Brownish yellow	Yellow

Each Value is ± S.E

The phytochemical results of *Z. spina* and *M. oleifera* oils are presented in Table 2. There are at least fourteen classes of secondary metabolites (chemical compounds) from fruits and vegetables that exert biological activities and can potentially be used to promote human health. These include alkaloids, amines, cyanogenic glycosides, diterpenes, flavonoids, glucosinolates, monoterpenes, non-protein aminoacids, phenylpropanes, polyacetylenes, polyketides, sesquiterpenes, tetraterpenes, triterpenes, saponins and steroids [17]. Flavonoids, terpenoid, steroid were detected in oil samples. The biological functions of flavonoids include protection against allergies, inflammation, free radicals scavenging, platelets aggregation, microbes, ulcers, hepatoxins, viruses and tumours [18]. The presence of steroids in the oils is of importance and interest in medicine due to their relationship with compounds as sex hormones [19]. *Z. spina* contains both alkaloids and saponins which are known to exhibit medicinal, physiological activity and can be seen as a potential source of useful drugs [20]. Many pure isolated alkaloids and their synthetic derivatives are used as basic

medicinal agents for their analgesic, antispasmodic and bactericidal effects [21, 22]. The presence of alkaloid in the oil suggests a possible medicinal use of the oil.

Table 2. *Phytochemical result of Z. spina and M. oleifera.*

Parameter	Z. spina	M. oleifera
Alkaloid	+	-
Carbohydrate	-	-
Saponin	-	+
Flavonoid	+	+
Tannin	-	-
Terpenoid	+	+
Phenol	-	-
Sterol	+	+
Glycoside	-	-
Cardic glycosides	-	-
Resin	+	-
Tritepenoid	-	-
Balsam	-	+
Volatile oils	-	-
Phlobatannin	-	-

The lipid profile and fatty acid distribution results show that both oils contain phospholipids, glycolipids and neutral lipids. Phospholipids have been reported to be an important component of cell membrane fluidity functionality, the neutral lipids serve as store of energy and constituents of membrane structure [23]. The presence of the lipid shows that the oils of *M. oleifera* and *Z. spina* will be good source of energy. Tsaknis et al 1999 [8] reported that the oil of *M. oleifera* contain high level of monounsaturated and saturated fatty acids and could be suitable source of oil for human consumption.

Table 3. *Lipid classes and Fatty acids distribution.*

Sample	Phospholipid	Glycolipids	Neutral
Z.spina	Phosphotidylethanol amine R_f value 0.7662	Synthetic cardidipin and phosphoglycerol; R_f value 0.2923 and 0.6615	Free fatty acid and dihosphatidyl glycerol; R_f value 0.3906 and 0.9531
M.oleifera	Free fatty acid R_f value 0.4545	Lyso-phosphootidylcholin and phosphotidylglycerol R_f value 0.2462 and 0.6615	Free fatty acid and dihosphatidyl glycerol R_f value 0.3906 and 0.9531

5. Conclusion

The observations made in the cause of this work suggest that the oil of *Z. spina* has potentials that could be explored for use as either industrial feed stock or other purposes. Further studies will be carried out to determine the effect of degumming on the physicochemical properties of oil sample.

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