



## Physicochemical properties, phenolic acids and volatile compounds of oil extracted from dry alhydwan (*Boerhavia elegana Choisy*) seeds

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**SUMMARY:** In this study, the chemical composition, physicochemical properties, phenolic acids and volatile compounds of alhydwan (*Boerhavia elegana Choisy*) seed oil were evaluated. The crude oil content was 11.49%, ash 6.88%, moisture 6.12%, protein content 14.60%, total carbohydrate 24.77% and fiber 36.13%. The oil contain a high quantity of unsaturated fatty acids (74.63 mg·100 g<sup>-1</sup>) with oleic (C18:1) (57.77%), palmitic (C16:0) (18.65%) and linoleic (C18:2) (12.88%) acids as the most abundant. The relative density was 0.88 and the iodine value 105.59. The color analysis showed a value of 28.33 Y+1.43 R. The oil also had a high relative oxidative stability. The tocol composition showed that  $\alpha$ -tocotrienol,  $\gamma$ -tocopherol and  $\gamma$ -tocotrienol were in a higher concentration than the rest. Seven phenolic acids (caffeic, vanillic, galic, *p*-coumaric, ascorbic, cinnamic and ferulic) were detected, with ascorbic acid as the predominant one (5.44 mg·100 g<sup>-1</sup>). In relation to the volatile composition, 48 compounds were found with Z-10-Pentadecen-1-ol (56.73%); Hexadecenoic acid, Z-11- (18.52%); 9,12-Octadecadienoic acid (Z,Z)- (3.93%) and 9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-1-(hydroxymethyl) ethyl ester (3.04%) as the most abundant. These findings demonstrated the potential of alhydwan seeds to be used as a good source of quality edible oil.

**KEYWORDS:** *Alhydwan*; *Boerhavia elegana Choisy*; *Novel food*; *Phenolic acids*; *Physicochemical properties*; *Volatile compounds*

**RESUMEN:** *Propiedades físico-químicas, ácidos fenólicos y compuestos volátiles del aceite extraído de semillas de alhydwan (Boerhavia elegana Choisy).* En este estudio se ha determinado la composición química, las propiedades físico-químicas, ácidos fenólicos y compuestos volátiles de aceites de semillas de alhydwan (*Boerhavia elegana Choisy*). Las semillas contenían un 11.49% de aceite, 6.88% de cenizas, 6.12% de humedad, 14.60% de proteínas, 24.77% de carbohidratos totales y 36.13% de fibra. El aceite contiene 74,63 mg·100 g<sup>-1</sup> de ácidos grasos insaturados, con oleico (C18: 1) (57,77%), palmítico (C16: 0) (18,65%) y linoleico (C18: 2) (12,88%) como los más abundantes. La densidad relativa fue de 0,88 y el índice de yodo de 105,59. El análisis del color mostró un valor de 28.33 Y+1,43 R. El aceite también mostró tener una alta estabilidad oxidativa relativa. La determinación de la composición de tocols mostró que  $\alpha$ -tocotrienol,  $\gamma$ -tocoferol y  $\gamma$ -tocotrienol están presentes en mayor concentración que el resto. Se detectaron siete ácidos fenólicos (cafeico, vanílicico, galico, *p*-cumárico, ascórbico, cinámico y ferúlico), siendo el ácido ascórbico el mayoritario (5,44 mg·100 g<sup>-1</sup>). En la determinación de volátiles, se encontraron 48 componentes, con Z-10-Pentadecen-1-ol (56,73%); ácido hexadecenoico, Z-11- (18,52%); ácido 9,12-octadecadienoico (Z, Z) - (3,93%) y ácido 9,12-octadecadienoico (Z, Z) -, éster 2-hidroxi-1- (hidroximetil) etil (3,04%) como mayoritarios. Estos resultados demostraron que las semillas de alhydwan tiene un gran potencial para ser utilizadas como una buena fuente de aceite comestible de calidad.

**PALABRAS CLAVE:** *Ácidos fenólicos; Alhydwan; Boerhavia elegana Choisy; Compuestos volátiles; Nuevo alimento; Propiedades físico-químicas*

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## 1. INTRODUCTION

Due to the ever-increasing demand for and price of edible oils, a number of explorations are being made to discover more edible oils, more especially those extracted from plant sources (Sbih *et al.*, 2013). The global supply available from conventional animal fat and vegetable oils is not sufficient to meet all the energy requirements without compromising the demand for food. Much attention however, is put on plant seed sources that have been proven to have healthy and nutritional benefits to consumers in one way or another (Su *et al.*, 2014). In recent years there has been an increased important exploitation of promising plant species as a source of dietary or specialty oils (Jelassi *et al.*, 2014). Several of these plants contain considerable quantities of oils, desirable fatty acids and/or a high proportion of nutritionally, medicinally or industrially important materials (Bowen and Clandinin, 2005). Nevertheless, with regards to the ever-growing world population, the demand for high-quality seed oils continues to grow. In order to meet this demand, there is a need to increase the production of the major oil crops and to diversify the sources through increasing and improving the production of minor oil crops (Mulatu *et al.*, 2011). Vegetable oils are utilized in many foods and many other industrial processes. Of great importance are those with higher relative content of minor lipid components for human health (Jelassi *et al.*, 2014). The world consumption is dominated by palm, soybean, rapeseed and sunflower oils with 38.1, 35.7, 17.8 and 18.2 million tones consumed per year, respectively (American Soybean Association, 2007) with world interest still on the recovery and exploitation of oils from more natural plant resources (Jelassi *et al.*, 2014). Alhydwan (*Boerhavia elegana Choisy*) is one of the edible herbaceous plants, a type of *Boerhavia* that belongs to the Nynctaginaceae family, commonly available in South Yemen (Boulous, 1988). *Boerhavia* is a genus of 40 species, almost all of which are widely distributed in tropical and sub-tropical areas of Asia, Africa, America and Australia (Chaudhary *et al.*, 2011). Alhydwan is cultivated only in Yemen and is not well known even in its neighboring countries. It has a long history of uses by indigenous and tribal people there in the traditional cuisine of as one of the staple ingredients in the manufacture of porridge, desserts and

savory products. It is also eaten as a supplement mixed in bread and cakes, and is characterized by good flavor (Al-Farga *et al.*, 2014). To our knowledge so far, there is no scientific report on physicochemical properties, chemical composition, phenolic acids and volatile compounds of oil extracted from the alhydwan seeds. The present study therefore, aimed at examining these particular aspects. The knowledge of these properties may lead to the innovative use of alhydwan seed oil for food and other industrial purposes.

## 2. MATERIALS AND METHODS

### 2.1. Material and chemicals

Dried alhydwan seeds were brought from a local farm in Wad Hadramout City, Yemen in June of 2014 after harvesting, and transported to the Functional Ingredients and Healthy Foods Laboratory of Jiangnan University, Wuxi city, China. Seeds were milled using a laboratory scale hammer mill (Tianjin Taisite Instrument Co., Ltd., Tianjin, China). Flour was then screened (80 mesh), packed in polyethylene bags, and stored in a refrigerator at 5 °C until use. Standard tocopherols were purchased from Sigma Aldrich, Shanghai, China. All other chemicals were of analytical grade.

### 2.2. Oil extraction

The seeds (100 g) were mixed with 1 L of n-hexane using a shaker (IKA® RW 20 digital, UK-plug) at a rate of 440 rpm for 4 h. The mixture was then centrifuged for 20 min at 4 °C. The mixture was then filtered and the supernatant was recovered. The extraction process was repeated. The oil trapped in the supernatant was then recovered by evaporating off the solvent using a rotary evaporator (Model N-1, Eyela; Tokyo Rikakikal, Japan). The remaining solvent was removed under a laboratory fume hood for 30 min at 37 °C. The extracted oil was drained under a nitrogen stream and was then stored in a refrigerator at 4 °C for further analysis.

### 2.3. Proximate composition analysis of alhydwan seeds

The following analyses were carried out: Nitrogen contents were determined using a micro-Kjeldahl method. Moisture content was determined

by oven-drying at 60 °C to constant weight. Crude lipids were estimated by employing the Soxhlet apparatus method using n-hexane as solvent. The standard methods of The Association of Official Analytical Chemists (AOAC, 2000) were employed for the estimation of the proximate composition of seed flour. Crude fiber contents were estimated as outlined in the AOAC Method 962.09 (AOAC, 2000). Ash content was determined by ashing the sample at 550 °C to constant weight. Iron content was determined using a UV-Visible spectrophotometer (TECHCOM Co., Shanghai, China) at 480 nm (AOAC, 1995). Magnesium was determined according to the method described by Ranganna (1986). The blue color that developed was read at 650 nm in a UV-Visible spectrophotometer and expressed as magnesium mg·100 g<sup>-1</sup> meal. Other minerals were determined by atomic absorption spectroscopy (Shimadzu AA 6701F, Atomic Absorption Flame Emission Spectrophotometer equipped with a hollow cathode lamp).

#### 2.4. Total energy (Caloric value)

Energy was calculated according to the method of (Osborne, D.R. and Voogt, P. 1978), using the Atwater factor. 1 g of fat provides (9 K calories), 1 g of protein provides (4 K calories) and 1 g of carbohydrates provides (4 K calories).

#### 2.5. Chemical analysis of the oil

The American Oil Chemist's Society (1997) methods were used for the estimation of free fatty acids (method Ca 5a-4), peroxide value (method Cd 8-53), saponification value (method Cd 3-25), unsaponifiable matter (method Ca 6a-40), and specific gravity (using a 10 mL pycnometer at 25 °C). The refractive index was determined using an Abbe refractometer at 25 °C. The iodine value was calculated following the procedures of Kyriakidis and Katsiloulis (2000). Specific absorption values  $k_{232}$  and  $k_{270}$  were estimated using a UV spectrophotometer.

#### 2.6. Determination of the oil's fatty acid composition

Fatty acid methyl esters (FAME) were prepared by methylation of the total lipids according to the method described by Azhari *et al.* (2014). One  $\mu$ L of FAME sample was analyzed by gas chromatography (GC) (Shimadzu GC-2010, Series PEG30 M, Japan) equipped with a flame ionization detector. GC separation was conducted on a capillary column (PEG30 M; 30 m×0.32 mm×0.50  $\mu$ m). The carrier gas was nitrogen and the column flow rate was 0.8 mL·min<sup>-1</sup>. The chromatographic analysis was done at 190 °C oven temperature for 1 min and then increased to 230 °C at a rate of 3 °C·min<sup>-1</sup> and maintained at 230 °C for 10 min. The injector

and detector temperatures were 240 and 250 °C, respectively. The peaks were estimated on the chromatogram according to retention time from analyzed standard samples. Finally, fatty acid contents were calculated as percentages (%).

#### 2.7. Color parameters

The color of the alhydwan oil was measured with a Hunter Lab digital colorimeter (TC-PIIG system, Beijing Optical Instrument Co. Ltd., Beijing, China) as R Value on the red slide, Y value on the yellow slide and the values were recorded. A cylindrical plastic dish (58 mm in diameter and 15 mm in depth) containing the same content of sample was placed at the light port (50 mm in diameter).

#### 2.8. Oxidative stability determination

The oxidative stability was determined with the 743 Rancimat apparatus (Metrohm Co., Basel, Switzerland). The oxidative-induction time (OIT) was determined using 3.5 g of oil. The temperature was set at 100 °C, and the purified air flow at a rate of 10 L·h<sup>-1</sup>. During the oxidation process, volatile acids were formed in the distilled water where the conductivity was measured. The induction period was defined as the time necessary to reach the inflection point of the conductivity curve.

#### 2.9. Tocol composition

The tocol composition was determined according to norm ISO 9936 by HPLC (Agilent 1100, CA, USA), consisting of a G1354 quaternary pump, a G1313A standard auto sampler, a G1321A fluorescence detector set at ( $\lambda$  excitation=295 nm, and  $\lambda$  emission=330 nm) and chemstation software. The seed oil 0.5 g was diluted with 5 mL n-hexane and 5  $\mu$ L samples were automatically injected into a normal phase column (Pinnacle II silica) (150 mm×3.2 mm×3  $\mu$ m) with hexane/isopropanol (99.5/0.5, v/v) as a mobile phase. The system was operated isocratically at a flow rate of 0.5 mL·min<sup>-1</sup>. The separations were carried out at 30 °C. The mixed tocopherol standards in a hexane solution (2 mg·mL<sup>-1</sup>) were prepared from the standard compounds:  $\alpha$ -,  $\beta$ -,  $\gamma$ -, and the  $\delta$ -tocopherols (Sigma Aldrich, Shanghai, China). The tocotrienol peaks of *B. elegans Choisy* seed oil were identified by comparison with tocol chromatograms of coconut oil and palm oil obtained under similar analytical conditions.

#### 2.10. Determination of sterols

The sterol components of alhydwan seed oil were determined by derivatization with N-methyl-N-trimethyl-silyl-heptafluorobutyramid as silylation agent. The assignments of the peaks were made using

the retention times of the individual sterols and calculation of the relative retention times in relation to betulin according to internal standard (ISO/FIDS, 1999). In brief, 250 mg of oil were saponified with a solution of ethanolic potassium hydroxide by boiling under reflux. The unsaponifiable matter was extracted by solid-phase extraction on an aluminum oxide column on which fatty acid anions were retained and sterols passed through. The sterol fraction from the unsaponifiable matter was separated by thin-layer chromatography (TLC) on 20×20 cm silica gel, 0.25 mm layer thickness using hexane/diethyl ether 1:1 (v/v) as developing solvent was extracted again from the TLC substrate. The sterol fraction was detected by gas liquid chromatography using an internal standard (betulin). Unclear cases were detected by GC mass only lastly, other parameters were: hydrogen, which was the carrier gas; split ratio, 1:20; injection and detection temperature were established at 320 °C; oven temperature was programmed between 245–260 °C at 5 °C·min<sup>-1</sup>.

### 2.11. Identification and quantification of phenolic acids by HPLC–MS/MS

The HPLC–MS were performed with an Agilent 1100 LC system consisting of a degasser, a binary pump, an auto sampler, and a column heater (chromatographic separation, a Knauer C18 column (250 mm×4.6 mm, 5 µm). CHEMCAD 6.3, chemical process simulation software designed by chemstation, Inc, was used to obtain data acquisition and mass spectrometric evaluation. The column outlet was coupled to an Agilent MSD Ion Trap XCT mass spectrometer equipped with an ESI ion source. The column set at 40 °C was first held at 90% solvent A (1% acetic acid in water) and 10% solvent B (1% acetic acid in methanol), followed by a step gradient from 10% B to 20% B in 4 min, and a second step gradient from 20% B to 100% B in 20 min. Thereafter, it was held for 6 minutes with 100% B. Finally, the elution was obtained from 100% B to 20% B for 6 min. The flow rate was 400 µL·min<sup>-1</sup> and the injection volume was 10 µL of the mixture that contains 900 µL of ethyl acetate and 100 µL of alhydwan seeds oil. The following parameters were used in all stages of the experiment MS, that is, for electro spray ionization with negative ion polarity: the drying gas flow at 10 L·min<sup>-1</sup>, the nebulizer pressure at 40 psi, the drying temperature at 350 °C and the capillary voltage was set at 1.6 kV. The scan speed was 26,000 mZ<sup>-1</sup> s<sup>-1</sup> (Ultra Scan Mode), the fragmentation time was 30 ms, whereas the maximum accumulation time was 50 ms. The phenolic compounds were identified using a combination of high performance liquid chromatography (HPLC, Agilent 1100) for diode array detection and liquid chromatography with electro spray ionization mass spectrometry (ESI-LC-MS). Ultraviolet (UV)

spectra and mass spectra were compared to the spectra of authentic standards available.

### 2.12. Determination of volatile compounds

The volatile compounds were separated on a CP-Sil-8CB (Varian, Walnut Creek, CA, USA) fused silica capillary column (30 m length, 0.25 mm, id, and 0.25 µm film thicknesses) using a Varian model 3800 gas chromatography. The splitless mode injector was maintained at 220 °C and the flame ionization detector (FID) at 250 °C, with a capillary column DB WAX (30 m×0.25 µm, J and W Scientific, Folsom, CA, USA). The oven temperature was set at 40 °C, held for 3 min, ramped up to 100 °C at the rate of 6 °C·min<sup>-1</sup> and then to 230 °C at 10 °C·min<sup>-1</sup>. The constant column flow was 0.9 mL·min<sup>-1</sup>. Mass spectra was obtained in the Electron Impact (EI+) mode with an energy voltage of 70eV; the mass range was 33 to 450 m/z. volatile compound identification were conducted by matching their mass spectra of standard compounds found in the Wiley 130 K and national institute of standards and technology (NIST) 98 library of MS spectra and based on their retention indices.

## 3. RESULTS AND DISCUSSION

### 3.1. Chemical analysis of powdered alhydwan seeds

The chemical composition of alhydwan seeds is shown in Table 1. Fiber, fat, protein, carbohydrates, and ash were 36.13%, 11.49%, 14.60%, 24.77% and 6.88% on dry weight, respectively, and the moisture content was 6.12%. The high level of fiber makes alhydwan seeds convenient for food industry applications, and they could be a source of dietary fiber for animals. Since, alhydwan seed contains a good level of oil it is considered to be a good source for application in functional foods. The seeds are good sources for lipids, which contain fatty acids that play a very important role in the human body (Saidu and Jideobi, 2009). They facilitate the absorption of fat-soluble vitamins such as vitamins A and E (Osborne and Voogt, 1978). The variations in the oil content can be related to differences in plant species, climate cultivation, isolation method used and ripening level (Nyam *et al.*, 2009). Alhydwan seeds were also found to contain high levels of minerals (Table 1). Calcium was the most abundant element (655 mg·100 g<sup>-1</sup>), whereas copper was found at the lowest level.

### 3.2. Total Energy (Caloric Value)

The total energy content of the alhydwan seed is recorded in Table 1, where the calorific value has been calculated for fat, carbohydrates, and protein at 1.03, 0.99 and 0.58 kcal·g<sup>-1</sup>, respectively, with fat providing the highest amount of energy. From these



TABLE 1. Proximate composition and mineral contents of alhydwan *B. Elegana Choisy* (% w/w on dry basis)<sup>a</sup>

Chemical Components	Kcal·g <sup>-1</sup>	
<b>Proximate composition (%)</b>		
Moisture	6.12±0.11	–
Crude protein	14.60±0.54	0.58
Carbohydrates	24.77±0.35	0.99
Crude fat	11.49±0.81	1.03
Fiber	36.13±0.33	–
Ash	6.88±0.16	–
<b>Minerals content (mg·100 g<sup>-1</sup>)</b>		
Zinc (Zn)	1.72±1.44	
Iron (Fe)	3.83±1.52	
Copper (Cu)	0.44±0.62	
Manganese (Mn)	0.73±0.72	
Potassium (K)	595.00±1.52	
Sodium (Na)	26.15±0.71	
Magnesium (Mg)	109.50±0.54	
Calcium (Ca)	655.00±0.14	

<sup>a</sup>All values given are means of three determinations means ± standard deviation.

data, alhydwan seeds can be considered good sources of calories. It is well known that a calorie is a measurement of energy, the energy can be used immediately or stored for later use, and foods have calories. That is, foods supply the body with energy, which is released when foods are broken down during digestion. Thus, energy enables the cells to carry out all of their functions, including building protein and others substances needed by the body (Duyff and Ada, 2011).

### 3.3. Chemical analysis of the oil

Table 2 presents the chemical properties of *B. elegana Choisy* seed oil. The oil was in liquid state at room temperature, an indication of polyunsaturated fatty acids. The iodine value was relatively high compared to other oils suggesting that the seed oil has good edible oil quality (Eromosele *et al.*, 1997). The values of peroxide (3.35 meq O<sub>2</sub>·kg<sup>-1</sup> oil), free fatty acid composition (1.44%) and *p*-Anisidine (3.11) were very low indicating that the oil can be stored for a long period without deterioration (Ojeh, 1981). The high saponification value determined, 182.88 mg of KOH·g<sup>-1</sup> of oil, suggested a high content of low molecular weight triacylglycerols (Nehdi *et al.*, 2010). It was close to that of raspberry seed oil (Oomah *et al.*, 2000). Iodine value is the measurement of the degree of unsaturation of the oil. The iodine value for Alhydwan seed oil was 105.59; this result was found similar to the found by Roselle and Bittermelon (Nyam *et al.*, 2009). The iodine values of these seed

TABLE 2. Physicochemical properties of alhydwan seed oil

Parameters	Values
Physical state at room temperature	Liquid
Free fatty acids (oleic acid %)	1.44±0.06
Iodine value	105.59
Peroxide value (meq O <sub>2</sub> ·kg <sup>-1</sup> oil)	3.35±0.20
k <sub>232</sub>	3.03±0.18
k <sub>270</sub>	1.51±0.08
Color	28.33 Y+1.43 R
Index of refraction (25 °C)	1.43±0.06
Relative density (25 °C)	0.88±0.03
Unsaponifiable matter (%)	1.19±0.11
Saponification value (mg KOH·g <sup>-1</sup> oil)	182.88±0.11
<i>p</i> -Anisidine value	3.11±0.17
Oil stability index (h)	16.82±0.09

Values are means±SD of three determinations.

R value on the red slide, Y value on the yellow slide.

Iodine value was calculated following Kyriakidis and Katsiloulis (2000).

oils are situated inside the interval range of the value as mentioned by Tan *et al.* (2002).

### 3.4. Fatty acid composition

The fatty acid composition of alhydwan seed oil is recorded in Table 3. Among the current nine fatty acids, five were unsaturated. The most abundant fatty acids were oleic (C18:1) (57.77%), palmitic (C16:0) (18.65%) and linoleic (C18:2) (12.88%). Therefore, alhydwan seed oil was not similar to sunflower oil, which contains a lower content of linoleic acid, but a higher content of oleic acid (El-Mallah *et al.*, 1999). It is worth mentioning that the high content of oleic acid makes alhydwan seed oil a potential source of oil for patients with arteriosclerosis and essential hypertension. This means that the consumption of alhydwan seed oil may offer health benefits (Oomah *et al.*, 2000), making it a potential alternative natural oil to replace or combine with other edible oils.

### 3.5. Oxidative stability

The results from the Rancimat test are presented in Table 2. The oxidative stability of the alhydwan oil was relatively high (16.82 h). This value may be due to the high composition of oleic acid, which has only one double bond in the chain and the corresponding ester undergoes oxidation at a slower rate than the polyunsaturated esters (da Silva *et al.*, 2014). A linear regression on the basis of the ratio of linoleic acid and the contents of tocopherols and phenols in virgin olive oil and a good correlation with the oxidative stability measured by Rancimat have been reported (Aparicio *et al.*, 1999).

TABLE 3. Fatty acid composition of alhydwan ( $\text{g}\cdot 100\text{ g}^{-1}$  total fatty acid)<sup>a</sup>

Fatty acid	Composition
$\Sigma$ SFA	22.2
Myristic acid (C14:0)	0.2 $\pm$ 0.01
Palmitic acid (C16:0)	18.6 $\pm$ 0.03
Stearic acid (C18:0)	2.1 $\pm$ 0.06
Arachidic acid (C20:0)	1.3 $\pm$ 0.04
$\Sigma$ UFA	74.6
Palmitoleic acid (C16:1)	0.16 $\pm$ 0.02
Oleic acid (C18:1)	57.8 $\pm$ 0.4
Linoleic acid (C18:2)	12.9 $\pm$ 0.09
Linolenic acid (C18:3)	2.3 $\pm$ 0.05
Eicosenoic acid (C20:1)	1.5 $\pm$ 0.03
$\Sigma$ MUFA	59.4
$\Sigma$ PUFA	15.2
Ratio SFA/UFA	3.9

<sup>a</sup>All values given are means of three determinations means  $\pm$  standard deviation.

SFA- saturated fatty acids; MUFA- monounsaturated fatty acids; PUFA- polyunsaturated fatty acids; UFA- unsaturated fatty acids.

### 3.6. Color parameters

The CIE Lab coordinate values ( $L^*$ ,  $a^*$ ,  $b^*$ ) of alhydwan seed oil were 65.44, 1.11 and 28.33, respectively. These indicate the presence of yellow pigments such as carotenoid compounds. These values are close to those reported on other vegetable oils such as palm, soybean, sunflower, olive, and corn, which range from 63.4 to 69.5, 3.8 to 4.4 and 9.2 to 10.4, respectively (Hsu and Yu, 2002).

### 3.7. Tocol composition

The tocopherol and tocotrienol composition of alhydwan seed oil are shown in Table 4. The seed oil showed high amounts of  $\alpha$ -tocotrienol ( $\alpha$ -T3),  $\gamma$ -tocopherol ( $\gamma$ -T), and  $\gamma$ -tocotrienol ( $\gamma$ -T3). The major tocols were the  $\alpha$ -T3, which was 68% of total tocols. Fatnassi *et al.* (2009) reported that  $\alpha$ -tocopherol is useful to human nutrition due to its higher biological activity than other tocopherols. The total tocopherol content in *B. elegans Choisy* seed oil was 53.20  $\text{mg}\cdot 100\text{ g}^{-1}$  oil, which is very close to that of palm seed oil (51.54  $\text{mg}\cdot 100\text{ g}^{-1}$ ) reported by Nehdi *et al.* (2010). The results strongly suggest that the *B. elegans Choisy* seed oil is a rich source of  $\alpha$ -tocotrienol ( $\alpha$ -T3), also similar to palm oil (Al-Saqer *et al.*, 2004). It should be noted that tocopherols and tocotrienols have antioxidant properties and they are active as vitamin E, whose deficiency affects hemolysis in humans and nervous system development in children (Mohamed *et al.*, 2007).

TABLE 4. Tocols and sterols of alhydwan seed oil

Compound	$\text{mg}\cdot 100\text{ g}^{-1}$ oil
<b>Tocols</b>	
$\alpha$ -Tocopherol	0.92 $\pm$ 0.09
$\beta$ -Tocopherol	8.22 $\pm$ 0.13
$\gamma$ -Tocopherol	1.03 $\pm$ 0.08
$\delta$ -Tocopherol	2.02 $\pm$ 0.11
$\alpha$ -Tocotrienol	38.02 $\pm$ 0.10
$\gamma$ -Tocotrienol	5.33 $\pm$ 0.06
Total tocols	55.54 $\pm$ 0.09
<b>Sterols</b>	
Stigmasterol	2.26 $\pm$ 0.40
Campesterol	26.24 $\pm$ 0.70
$\Delta$ 7-Avenasterol	4.02 $\pm$ 0.77
$\Delta$ 7-Stigmastadienol	2.66 $\pm$ 0.10
$\Delta$ 5-Avenasterol	24.44 $\pm$ 0.3
$\Delta$ 5,24-Stigmastadienol	14.25 $\pm$ 0.50
$\beta$ -Sitosterol	231.11 $\pm$ 1.02
Cholesterol	1.92 $\pm$ 0.3
Total sterols	306.90

All determinations were carried out in triplicate and mean value  $\pm$  standard deviation.

### 3.8. Sterols

The sterol contents of this crude oil are presented in Table 4. It was found that alhydwan seeds mainly contained  $\Delta$ 5-avenasterol,  $\beta$ -sitosterol, campesterol,  $\Delta$ 5,24-stigmastadienol,  $\Delta$ 7-stigmastadienol,  $\Delta$ 7-avenasterol, stigmasterol and cholesterol. But the predominant one was  $\beta$ -sitosterol (231.11  $\text{mg}\cdot 100\text{ g}^{-1}$  oil) and is similar to palm seed oils (Nehdi *et al.*, 2010).

### 3.9. Phenolic acids

It is well known that the phenolic compounds are minor oil constituents which play a crucial role in the flavor and on the resistance against oxidation and shelf life because of their chemical characteristics. Table 5,

TABLE 5. Phenolic acids ( $\text{mg}\cdot 100\text{ g}^{-1}$ ) composition of Alhydwan (*Boerhavia elegans Choisy*) seed oil

Phenolic acids	Composition
<i>p</i> -coumaric	4.1 $\pm$ 0.15
caffeic	1.2 $\pm$ 0.33
vanillic	2.3 $\pm$ 0.23
galic	1.6 $\pm$ 0.45
ascorbic	5.44 $\pm$ 0.11
cinnamic	3.4 $\pm$ 0.56
ferulic	2.6 $\pm$ 0.29

Values are means $\pm$ SD of three determinations.

TABLE 6. Headspace volatile components of alhydwan seed oil, retention time and peak area percent (%) and retention time (min)

Serial	Components	Rt* (min)	Peak area (%)
1	Heptane	3.21	0.05
2	Octane, 2,7-dimethyl-	10.29	0.05
3	1,6-Anhydro- $\alpha$ -D-talopyranose	17.09	0.20
4	1,6-Anhydro- $\alpha$ -D-galactofuranose	19.22	0.06
5	4-(1-Hydroperoxy-2,2-dimethyl-6-methylene-cyclohexyl)-pent-3-en-2-one	19.51	0.19
6	Heptanoic acid, anhydride	19.60	0.12
7	1s,4R,7R,11R-1,3,4,7-Tetramethyltricyclo[5.3.1.0(4,11)]undec-2-en-8-one	19.73	0.37
8	Dodecane, 2,7,10-trimethyl-	20.01	0.11
9	Phenol, 2,4-bis(1,1-dimethylethyl)-	20.31	0.12
10	<b>Butylated Hydroxytoluene</b>	20.40	2.72
11	Dodecanoic acid	21.33	0.35
12	Diethyl Phthalate	21.95	0.09
13	2,6-di-tert-Butyl-4-(dimethylaminomethyl)phenol	23.86	0.15
14	Tetradecanal	24.03	0.22
15	Tetradecanoic acid	24.66	0.05
16	Tetradecanamide	24.72	0.07
17	3,5-di-tert-Butyl-4-hydroxybenzaldehyde	24.84	0.05
18	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	25.70	0.13
19	Pentadecanoic acid	25.91	0.06
20	1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester	26.06	0.11
21	Silane, trichlorooctadecyl-	26.66	0.35
22	<b>Hexadecenoic acid, Z-11-</b>	26.81	18.52
23	n-Hexadecanoic acid	27.11	0.09
24	n-Hexadecanoic acid	27.40	0.09
25	Z-8-Methyl-9-tetradecenoic acid	27.79	0.15
26	Heptadecanoic acid	27.97	0.06
27	Hexadecenenitrile	28.19	0.05
28	<b>Z-10-Pentadecen-1-ol</b>	28.29	56.73
29	<b>9,12-Octadecadienoic acid (Z,Z)-</b>	28.82	3.93
30	Octadecanoic acid	28.98	0.43
31	9-Octadecenamide, (Z)-	29.16	0.39
32	Acetic acid, octadecyl ester	29.34	0.94
33	Pyrrolidine, 1-(1-oxo-7,10-hexadecadienyl)-	30.55	0.52
34	Erucic acid	30.84	0.06
35	E-8-Methyl-9-tetradecen-1-ol acetate	30.95	1.04
36	<b>9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester</b>	31.10	3.04
37	9-Octadecenamide, (Z)-	31.17	0.07
38	Hexadecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester	31.34	0.50
39	Tetradecanamide	31.47	0.51
40	Diisooctyladipate	31.61	0.17
41	13-Tertadecen-1-ol acetate	31.75	0.17
42	1-Heneicosyl formate	33.07	0.11
43	Pentacosane	33.20	0.80
44	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester	33.42	0.14
45	1,2-Benzenedicarboxylic acid, diisooctyl ester	34.22	0.13
46	Docosanoic acid	34.47	1.34
47	9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester	37.58	0.52
48	9-Octadecenoic acid (Z)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester	37.64	0.05

Major volatile (&gt;2.0%) bold; \*RT: Retention times.

shows the phenolic acids with their HPLC retention times and concentrations. In alhydwan seed oil, seven phenolic acids were identified, namely ascorbic acid (the most abundant), *p*-coumaric acid, cinnamic acid, ferulic acid, vanillic acid, galic acid and caffeic acid. This is a good indication that the phenolics of this plant are important components as the *Boerhavia elegans Choisy* seeds possessed high phenolic content. Therefore, Alhydwan (*B. elegans Choisy*) seeds can be used as a good source of natural antioxidants which can have an invaluable pharmacological effect (Alfarga *et al.*, 2014).

### 3.10. Volatile compounds

The forty-eight volatile compounds found in alhydwan seed oil are shown in Table 6. The peak area percentage was used to indicate the relative concentration of each compound. The main compounds identified based on relative contents were; Z-10-Pentadecen-1-ol (56.73%); Hexadecenoic acid, Z-11-(18.52%); 9,12-Octadecadienoic acid (Z,Z)-(3.93%); 2-hydroxy-1-(hydroxymethyl)ethyl ester (3.04%) and Docosanoic acid (1.34%). Volatile compounds play a prominent role as flavoring agents in the food industry and they are responsible for a plant's distinctive scent or taste (Bruneton, 1999). Flavor volatiles are derived from an array of nutrients, including carotenoids, amino acids, and fatty acids (Goff and Harry, 2006). Alhydwan seeds oil can be considered a good source of volatile compounds, which can be used as an addition to several food or beverages to give them better taste and flavor.

## 4. CONCLUSION

The results of this study have revealed that alhydwan seed oil has a number of compounds which makes it a commendable food oil. Comparatively, it has shown some similarity with palm seed oil. The high contents of monounsaturated fatty acids, the liquid state at room temperature and the many volatile compounds present make it potentially healthy and a good organoleptic oil for human consumption. Unfortunately, many countries are not even familiar with the alhydwan plant, much less consider its potential of being a source of good oil. This study is the first carried out on the alhydwan seed grown in Yemen, opening the way for further studies on these seeds.

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