



## CHARACTERISTICS AND CHEMICAL COMPOSITION OF *Solanum elaeagnifolium* SEED OIL

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### ABSTRACT

The physico-chemical characteristics and the fatty acid, sterol and triacylglycerol compositions of oil extracted from *Solanum elaeagnifolium* seeds were determined in this study. The percentage yield of the oil was calculated as 3%. The saponification value (226.15mg KOH g<sup>-1</sup>), iodine value (149.1 g/100 g of oil), peroxide value (61.87 mequiv.O<sub>2</sub>/kg) and free fatty acid (13.9 mg KOH g<sup>-1</sup>) were determined to assess the quality of the oil. The physico-chemical characterization showed that *Solanum elaeagnifolium* seed oil is unsaturated semi-drying oil, with high saponification and acidic values. The oil contains high levels of Linoleic acid (C<sub>18:2</sub>, 67.59%), oleic acid (C<sub>18:1</sub>, 16.70%) and palmitic acid (C<sub>16:0</sub>, 8.52%). LLL (35.38%), OLL (18.91%), PLL (16.52%) and PLO (13.81%) were the abundant triacylglycerols representing more than 84% of the seed oil (L: linoleic, O: oleic, P: palmitic, S: stearic). The sterol marker, β-sitosterol, accounted for 69.66% of the total sterol content in the seed oil and is followed by campesterol (11.51%), stigmasterol (7.64%) and Δ<sup>5</sup>-avenasterol (5.37%).

**Keywords:** *solanum elaeagnifolium*, seed oil, physico-chemical, chemical composition.

### 1. INTRODUCTION

Vegetable oils are important not only for nutritional purposes, but also as raw materials for a wide range of industrial products which includes fuels, skin care products, high pressure lubricants and alkyd resins for paint (Ibemesi, 1993; Foidl *et al.*, 1996). These applications require extensive studies on the physico-chemical properties of oils in order to ascertain their suitability as raw materials.

As apart of our investigation into some medicinal plants known in Tunisia (Koubaa and Damak, 2003; Miladi *et al.*, 2008; Damak *et al.*, 2011; Kadri *et al.*, 2011), we report below the study of the *Solanum elaeagnifolium* seed oil.

The genus *Solanum* belonging to Solanaceae family (subfamily: curvembryea) (Nicolas *et al.*, 1849; Cauvet, 1864), consists of approximately 2000 species (Vander Burgt and Van Mdenbach de Rooy, 1996). The specimen *Solanum elaeagnifolium*, commonly named *silverleaf nightshade* or *trompillo* (Mellado *et al.*, 2008) is a perennial shrub, is widely distributed in Asia, Africa, Australia, and tropical and subtropical America (Chiale *et al.*, 1991; Boyd *et al.*, 1984). It is native to countries bordering the Mediterranean Sea. No report indicates its uses in traditional, but relatively few phytochemical analyses have been published (Chiale *et al.*, 1991).

To our knowledge, the physico-chemical characterization of the oil produced from the seeds of *Solanum elaeagnifolium* has not been reported. This investigation was undertaken to determine the physico-chemical properties, and the fatty acid, triacylglycerol and sterol composition of this oil.

### 2. MATERIALS AND METHODS

#### 2.1. Plant material

*Solanum elaeagnifolium* was collected in February 2009 at Monastir, Tunisia. It was identified by Pr. M. Om Zin (Institut Supérieur Agronomique de Chott Meriem, Sousse, Tunisie). Voucher specimen (N°LCSN112) was deposited at the Natural Product Chemistry Laboratory, Faculty of Sciences of Sfax, University of Sfax, Tunisia.

The fruit (the berries) is an irregularly dehiscent berry, initially spherical, green (with white patches) and fleshy, drying and becoming yellow to orange (approximately 0.3 g weight and 10-15 mm of diameter) at maturity. A single plant generally produces 40-60 fruits, each containing 60-120 seeds, smooth, flat, greenish-brown, 2-3 mm in diameter, closely resembling those of tomatoes (OEPP/EPP0, 2007).

#### 2.2. Lipid extraction

Mature seeds were first ground by a grinder to fine powder. The extracted with 500 ml hexane using a soxhlet extraction apparatus for 20 h. The solvent was removed via a rotatory vacuum distillation at 40-50°C.

The residue was weighed and stored at -20°C until it was analyzed. The weight of the oil extracted from 850 g of the seed powder was determined to calculate the lipid content. The result was expressed as the lipid percentage in the dry seed powder.

#### 2.3. Analytical methods

##### 2.3.1. Chemical analysis of the oil

Some physico-chemical indices of the oil were determined. The following were evaluated according to the methods listed in the ISO (International Organization



for Standardization) and in IOC (International Oil Council).

**Table-1.** Norm of chemical parameters of *Solanum elaeagnifolium*.

Parameter	Norm
Saponification value (mg KOH/g oil)	ISO 3657, 1988
Peroxyde value (meq/Kg)	ISO 3960, 1998
Acid value (mg KOH <sup>-1</sup> )	ISO 660, 1996
Unsaponifiable matter	ISO 3596, 1996
Iodine value (%)	ISO 3961, 1996

#### (a) Fatty acid composition

The fatty acid methyl esters were prepared by adding 5mL of methylation reagent: anhydrous methanol-hexane-concentrated sulphuric acid ( $\rho = 1.84$ ) in the ratio 75:25:1 (V/V/V) to 0.1g of oil. The mixture was refluxed for 30 min, followed by adding 10 mL of distilled water and 10 mL of petroleum ether according to the procedure reported by IOC (COI/T.20/Doc.N<sup>o</sup>.10, 2001). After stirring and waiting until the phase separation, the fatty acid methyl esters were extracted twice times with 20 mL of ether and concentrated using a current of nitrogen.

In order to determine the fatty acid composition, One microliter of the FAMES was injected onto a 6890N Network GC System (Agilent Technologies) equipped with a flame ionization detector (FID) and a polar capillary column (HP- cyanoproyl polysiloxane cyanoproyl polysiloxane, 0.25 mm internal diameter, and 50 m in length and 0.25  $\mu$ m film in thickness) to obtain individual peaks of fatty acid methyl esters. A flow rate of 1 mL/min of helium as a carrier gas was used. The injection port was heated at 220°C. The detector temperature was set at 250°C. The column was maintained at an initial temperature of 170°C to final temperature of 225°C.

The relative percentage of the fatty acid was calculated on the basis of the peak area of a fatty acid species to the total peak area of all the fatty acids in the oil sample. Fatty acid methyl esters peak identification was accomplished by comparing the retention times of peaks with those of pure standards purchased from Sigma and analyzed under the same conditions.

#### (b) Sterol composition

Unsaponifiable matter of *Solanum elaeagnifolium* seed oil was extracted and determined in the oil sample according to IOC (COI/T.20/Doc.N<sup>o</sup>.10, 2001). One gram of oil was refluxed with 10 ml of 10% ethanolic potassium hydroxide for 1h. The reaction mixture was diluted with 10 ml of distilled water and the unsaponifiable matter was extracted three times with 20mL portion of ether. The ether extract was dried over anhydrous sodium sulphate and evaporated. The sterol fractions obtained by TLC separation from the unsaponifiable of the oil was derived with silanizing mixture, (pyridine-hexamethyl disilazane-

trimethyl chlorosilane 9:3:1, v/v/v) in the ratio of 50  $\mu$ L for every mg of sterols.

Further determination of the sterol analysis was carried out on an Agilent Technologies (7890 A) CPG equipped with a HP-5 column (30m  $\times$  0.32 mm  $\times$  0.25mm i.d) under the following operating condition: column temperature 267°C, detector temperature 290°C, injector temperature 280°C, carrier gas: helium (1mL min<sup>-1</sup>).

#### (c) Triacylglycerol composition

The analysis of triglycerides was performed according to IUPAC 2.324 method. In fact, a 5% solution of the samples to be analyzed was prepared by weighing 0.5 g of the oil sample into a 10 mL graduated flask and making up to 10 ml with acetone.

The triacylglycerols (TAGs) profile was obtained by a reverse phase high performance liquid chromatography (HPLC) (HP 1100, Agilent Technology) equipped with a differential refractometer detector, using a C18 column (250 mm, 4.6 mm, 5  $\mu$ m particle size). The eluent used was a mixture of acetonitrile/acetone (50/50, v/v) at a flow rate of 1.5 mL/min and an injection volume of 20  $\mu$ L of the sample prepared as indicated above.

The identified TAGs of seed oil were concluded by comparing the retention time of standard TAGs peak. It was assumed that the sum of the areas of the peaks corresponding to the various TAGs was equal to 100%, and the relative percentage of each TAGs was calculated.

### 2.3.2 Physical analysis of the oil

#### (a) Specific extinction K232 and K270

Extinction coefficients were determined from absorbance at 232 and 270 nm, respectively, with UV spectrophotometer (UNICO 2800 UV visible) using a 1% solution of oil in cyclohexane and a path length of 1 cm; in according to IOC (COI/T.20/Doc.N<sup>o</sup>.19/Rév.1, 2001).

#### (b) Carotenoids and chlorophylls determination

The chlorophyll content was determined by the method reported by (Wolff, 1968) and the carotenoid content was determined by the method reported by (Poiana *et al.*, 2001).

#### (c) Extraction and analysis of total phenolic content

The total phenolic content (TPC) of the seed oil of *Solanum elaeagnifolium* was estimated by a colorimetric assay, according to the method described by (Singleton *et al.*, 1965) with some modifications. Briefly, 1 mL of oil (1mg/ml) was mixed with 1mL of Folin-Ciocalteu reagent. After 3 min, 1 mL of saturated Na<sub>2</sub>CO<sub>3</sub> solution was added to the mixture followed by the addition of 7 mL of distilled water. The mixture was kept in the dark for 90 min, after which the absorbance was read at 725 nm (JENWAY UV-6320 D). The total phenol contents (TPC) was determined using a standard curve obtained with gallic acid (0.01-0.4 mM). The estimation of the phenolic compound was carried out in triplicate. The



result was mean  $\pm$  standard deviation and expressed as milligram of gallic acid equivalent/g of extract (GAEs).

### 3. RESULTS AND DISCUSSIONS

#### 3.1. The physico-chemical properties of seed oil

The physico-chemical properties of the seed oil of *Solanum elaeagnifolium* are shown in Table-2.

**Table-2.** Physico-chemical properties of *Solanum elaeagnifolium* seed oil.

Parameter	<i>Solanum elaeagnifolium</i>
Yield (%)	2.95 $\pm$ 0.35
Acid value (mg KOH g <sup>-1</sup> )	13.9 $\pm$ 0.5
Iodine number (g/100 g of oil)	149.1 $\pm$ 0.7
Peroxide value (mequiv.O <sub>2</sub> /kg)	61.87 $\pm$ 0.41
Saponification value (mg KOH g <sup>-1</sup> )	226.15 $\pm$ 1.16
Unsaponifiable matter (%)	3.51 $\pm$ 0.05
Refractive index of (20 °C)	1.476 $\pm$ 0.002
Carotene (ppm)	0.95 $\pm$ 0.002
Chlorophyll (ppm)	1.58 $\pm$ 0.02
E <sub>232</sub>	11.30 $\pm$ 0.04
E <sub>270</sub>	2.10 $\pm$ 0.02
total phenolic content (TPC)	0.25 $\pm$ 0.03
State at ambient temperature	Liquid
Color	Green

#### 3.2. Fatty acid composition

The fatty acid composition of *Solanum elaeagnifolium* seed oil is presented in Table-3. The most important acids were linoleic C<sub>18:2</sub> (67.59%), oleic C<sub>18:1</sub> (16.70%), palmitic C<sub>16:0</sub> (8.52%), stearic C<sub>18:0</sub> (3.65%) and myristic C<sub>14:0</sub> (1.36%), which together compose about 97.8% of the total fatty acids? The *S. elaeagnifolium* seed oil can be regarded as linoleic-oleic oil. In fact it contains a high amount of linoleic acid (67.59%), which makes it especially prone to oxidation.

*S. elaeagnifolium* seed oil was characterized by a polyunsaturated/ saturated (P/S) ratio of 6.19. This value is in correlation with the refractive index. A high ratio of P/S is regarded favorably in the reduction of serum cholesterol and atherosclerosis and the prevention of heart diseases (Oomah *et al.*, 2000, 2002).

**Table-3.** Fatty acid compositions of *Solanum elaeagnifolium* seed oil (%).

Fatty acid	Carbon length	Composition (%)
Saturated		
Myristic	C14:0	1.36 $\pm$ 0.03
Palmitic	C16:0	8.52 $\pm$ 0.10
Margaric	C17:0	0.10 $\pm$ 0.02
Stearic	C18:0	3.65 $\pm$ 0.08
Arachidic	C20:0	0.16 $\pm$ 0.05
Behenic	C22:0	0.19 $\pm$ 0.05
<b>Monounsaturated</b>		
Palmetoleic	C16:1	0.50 $\pm$ 0.08
Margaroleic	C17:1	0.03 $\pm$ 0.01
Oleic	C18:1	16.70 $\pm$ 0.21
Eicosenoic	C20:1	0.17 $\pm$ 0.02
<b>Polyunsaturated</b>		
Linoleic	C18:2	67.59 $\pm$ 0.42
Linolenic	C18:3	0.96 $\pm$ 0.03
SAFA	-	13.98 $\pm$ 0.33
MUFA	-	17.40 $\pm$ 0.32
PUFA	-	68.55 $\pm$ 0.45
P/S	-	6.19

SAFA: Saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids.

#### 3.3. Sterol composition

The most studied fraction of the unsaponifiable matter is that of sterols, which is frequently analyzed for tracking commercial frauds (Lerker and Rodriguez-Estrada, 2000). This fraction has been considered as the major unsaponifiable fraction in many oils. The composition of the sterol fraction is shown in Table-4. The sterol fraction of the *S. elaeagnifolium* seed oil consisted mainly of  $\beta$ -sitosterol, campesterol, stigmasterol and  $\Delta_5$ -avenasterol, among which  $\beta$ -sitosterol was the most predominant (69% which), accompanied with minute amounts of cholesterol and  $\Delta_7$ -stigmastanol. The sterol composition of the seed oil of *S. elaeagnifolium* is comparable with literature (Zygdlo, 1994).

**Table-4.** Sterol compositions of *Solanum elaeagnifolium*.

Sterol	%	Quantity (ppm)
Cholesterol	2.43	169.65 ± 0.33
Brassicasterol	0.25	14.65 ± 0.10
Campesterol	11.51	814.53 ± 0.72
Stigmasterol	7.64	519.10 ± 0.48
Clerosterol	0.74	37.28 ± 0.14
$\beta$ -sitosterol	69.66	4858.34 ± 2.50
$\Delta$ -5-Avenasterol	5.37	380.46 ± 0.30
$\Delta$ -5-24 Stigmastadienol	0.54	21.85 ± 0.09
$\Delta$ -7- Stigmastenol	1.51	120.03 ± 0.18
$\Delta$ -7- Avenasterol	0.33	15.69 ± 0.09
Sterols Totaux	-	6952

### 3.4. Triacylglycerol composition

The distribution of triacylglycerols (TAGs), with the equivalent carbon number (ECN) is given in Table-5. According to the results, this oil contained twelve triacylglycerols (from ECN 42 to ECN 50). Considering the fatty acid composition, the major constituent was (LLL) followed by (OLL), (PLL), (POL) and (OOL). The triacylglycerol composition of *S. elaeagnifolium* reflects a close relationship between the fatty acid and triacylglycerol content of the oil.

**Table-5.** Triacylglycerol compositions (%) of *Solanum elaeagnifolium* seed oil.

Triglycéríde	ECN	%
LLL	42	35.38
LLO	44	18.91
PLL	44	16.52
LOO	46	6.17
POL	46	13.81
PLP	46	0.69
OOO	48	1.88
SLO + POO	48	4.62
POP	48	0.71
PPS	50	0.48
SOO	50	0.62
SLS + POS	50	0.21

L, linoleic acid; O, oleic acid; P, palmitic acid; S, stearic acid; ECN, equivalent carbon number.

### 4. CONCLUSIONS

This study shows that *Solanum elaeagnifolium* seed oil contains a large amount of unsaponifiable matter. The physico-chemical characteristics and fatty acid

composition suggested that *Solanum elaeagnifolium* seed oil could be successfully used for making soap, hair shampoo and alkyl resin.

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### REFERENCES

- Boyd JW, Murray DS and Tyrl RJ. 1984. Silver leaf nightshade, *Solanum elaeagnifolium*, origin, distribution and relation to man. *Economic Botany*. 38: 210 -217.
- Cauvet D. 1864. Des Solanées. 1<sup>st</sup> Ed. G. Silbermann, California. p. 75.
- Chiale CA, Cabrera JL and Juliani HR. 1991. Kaempferol 3-(6''-Czs-cinnamoylglucoside) from *Solanum elaeagnifolium*. *Phytochem*. 30: 1042-1043.
- Damak N, Allouche N, Hamdi B, Litaudon M and Damak M. 2011. Two new epimers of secoiridoid from olive mill wastewater. *Nat prod res*. In press.
- OEPP/EPP. 2007. *Solanum elaeagnifolium*. Data sheets on quarantine pests. EPPO Bull. 37: 236-245. <http://www.eppo.org>, (08.04.2010).
- Foidl N, Foidl G, Sanchez M, Mittelbach M and Hackel S. 1996. *Jatropha curcas* L. as a source of production of biofuel in Nicaragua. *Biores. Technol*. 58: 77-82.
- Ibemesi JA. 1993. Potential of melon seed oil in development of alkyl resin. *N. J. Sci*. 27: 299-304.
- Kadri A, Charsalla N, Damak M and Gdoua R. 2011. Chemical composition and in vitro antioxidant properties of essential oil of *Ricinus communis*. *J. Med. Plant Res*. 5(8): 1466-1470.



Koubaa I, Damak M. 2003. A new dilignan from *Cynara cardunculus*. *Fitoterapia*. 74: 18 -22.

Lercker G and Rodriguez-Estrada MT. 2000. Chromatographic analysis of unsaponifiable compounds of olive oils and fat-containing food. *J. Chromatogr.* 881: 105-129.

Mellado M, Garcia JE, Arévalo JR and Pittroff W. 2008. Replacement value of *Solanum elaeagnifolium* for alfalfa hay offered to growing goats. *Arid Environments*. 72: 2034-2039.

Miladi S, Jarraya R and Damak M. 2008. Lipid Composition and antioxidant Activities of *Daucus maritimus* Seeds. *J. Applied Sci.*

Nicolas JB, Gaston and Guibourt GP. 1849. *Histoire naturelle des drogues simples*. 4<sup>th</sup> Ed. J.-B. Baillière, Pais.

Oomah DB, Ladet S, Godfrey VD, Liang J and Giarard B. 2000. Characteristics of raspberry (*Rubus idaeus* L.) seed oil. *J. Food Chem.* 69: 187-193.

Oomah BD, Busson M, Godfrey DV and Drover JCG. 2002. Characteristics of hemp (*Cannabis sativa* L.) seed oil. *Food Chem.* 76: 33-43.

Poiana M, Mincione A, Giuffrè A M and Mincione B. 2001. Ricerche sugli oli di oliva monovarietali. Nota XII, Contributo alla caratterizzazione dell'olio estratto dalle olive della cv Picholine coltivata in provincia di Reggio Calabria. *Rivista Italiana Delle Sostanze Grasse*. LXXVIII: pp. 571-592.

Rude LL, Kelly K, Sawyer JK, Shah R and Wilso MD. 1998. Dietary monounsaturated fatty acids promote aortic atherosclerosis in LDL receptor-null ApoB100-overexpressing transgenic mice.

Ruggeri S, Cappelloni M, Gambelli L, Nicoli S and Carnovale E. 1998. Chemical composition and nutritive value of nuts grown in Italy. *J. Food Sci.* 10: 243-252.

Singleton VL and Rossi J A Jr. 1965. *Am. J. Enol. Viticult.* 16: 144.

vander Burgt XM and van Medenbach de Rooy JM. 1996. *The biodiversity of African plants*. Springer. p. 861.

Wolff JP. 1968. *Manuel d'analyse des corps gras*. Azoulay Ed., Paris, (France). pp. 197-199.

Zygdlo JA. 1994. A comparative study of sterols in oil seeds of *solanum* species. *Phytochem.* 35: 163-167.