SEED-OIL CONTENT AND FATTY ACID COMPOSITION OF SEED-OIL OF *THESPESIA POPULNEA* (L.) SOL. EX. CORR. GROWING IN SALINE SOILS IN KARACHI

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ABSTRACT

Seed oil content and fatty acid composition of the seed-oil was determined from *T. populnea* (L.) Sol. Ex. Corr. plants growing in saline soils (EC_e: 1.56-43.9 $dS.m^{-1}$). The seed oil content varied from 18.2 to 21.77% (mean: 19.81 ± 0.36%). Against the variation of salinity by 68.4%, the seed oil content varied merely by 4.83%. Twelve fatty acids were identified in *T. populnea* seed oil. Linoleic (37.27 ± 0.46 %), Palmitic (31.96 ± 0.24 %), and Oleic acid (17.73 ± 0.40%) were the major fatty acids. Over the ECe range of 1.56 to 43.03 $dS.m^{-1}$, the variation in fatty acid composition of seed oil was too small to make any significant difference in the oil quality as was evident from the compositional similarity of c 97% on an average amongst the oil samples extracted from the seeds of various trees. Saturated fatty acids content was 36.4% and unsaturated fatty acid content 63.6%. Oleic / linoleic acid ratio was oleic / linoleic acid ratio in sunflower with increase in salinity. No such increase in oleic / Linoleic acid ratio in *T. populnea* seeds didn't increase with salinity and remained almost constant - around 0.48. The saponification value of the oil was 204.

Key Words: *Thespesia populnea* (L.) Sol Ex. Corr., Seed oil, Fatty acid composition, Soil salinity, Foliar Na concentration.

INTRODUCTION

Oil crops are one of the important cash crops and are worth billion of rupees a year trade. In most of the developing countries oil production is not sufficient to meet requirements and these countries including Pakistan spent a large amount of money for oil import. Seventy five per cent of the World oil production comes from four main crops, Soybean, oil palm, rapeseed and sunflower (Murphy, 1996). Vegetable oil is mainly used for edible commodities and only a small percentage is used for the manufacture of industrial chemicals. The energy crisis through out the world has led the attention of world scientists towards the new oil crops for their possible use for edible purpose or fuel production. It is important that instead of relying on the small number of major crops, new oil crops should be explored and introduced to increase oil productivity (Murphy, 1996). Hamed *et al.* (2004) have described the fatty acid composition of *Nelumbo nucifera* seed oil and indicated to its potentiality as food in humans. Abid *et al.* (2007) described physicochemical characteristics and fatty acid composition of seed and pulp oil of *Hippophae rhamnoides*, a miracle plant, of Chitral and Northern areas of Pakistan and found its oil potentially suitable for edible purpose. The seeds of *Salvadora persica*, a known halophyte, is reported to contain 43.28 – 44.36 % oil of potential industrial use while growing in saline (EC: 24.0 ± 5.32 $dS.m^{-1}$; pH 7.4 ± 0.12) soil and 44.78 – 45.5 5 oil when growing in alkali soil (EC: 6.23 ± $dS.m^{-1}$; pH 9.23 ± 0.36; SAR, 42.71 ± 5.64) (Reddy *et al.*, 2008). The oil contains Lauric acid predominantly so rated for industrial use.

Salinity has affected the crop production of agricultural lands. Conventional agricultural crops have limited salt tolerance and the yield decreases as the salinity increases. There are many halophytic plants which contain oil and could potentially be used for human consumption. Weber *et al.* (2007) have reported that seeds of several native halophytes such as *Arthrocnemum indicum, Alhagi maurorum, Cressa cretica, Halopyrum mucronatum, Haloxylon stocksii* and *Suaeda fruticosa* have varying oil content (22 - 25%) with percentage of unsaturated fatty acids quite high (65-74%) although lower than the unsaturated fatty acid content in conventional vegetable oils such as soybean (84.6%), olive oil (85.8%), safflower (91.1%), and canola oil (92.8%).

Thespesia populnea (L.) Sol. Ex. Corr. is a coastal plant and a mangrove associate in vegetation of Samartan mangroves forests, Sarawak, Malaysia (Ashton *et al.*, 2002). It is highly salt tolerant plant (Aronson, 1989). In Pakistan, it is confined to coastal regions particularly Karachi – cultivated in parks or along roadside as ornamental or a shade plant (Abedin, 1979). The seeds of this plant are known to contain oil which is not exploited commercially. The increasing demand for food, energy and other plant products make it imperative to introduce new

plant species capable of growing on marginal land and water resources. The oil content and fatty acid composition of *T. populnea* seeds collected from trees growing under saline conditions around Karachi is, therefore, investigated in this paper.

MATERIALS AND METHODS

Description of the area

The climate of Karachi is of BWh type and bio-climate type as determined by Holdridge's system falls in the category of Tropical Bush Formation (Qadir *et al*., 1966). The rainfall is irregular and averages below 200mm mostly received in summer (Khan *et al*., 2000). Annual evapo-transpiration is 1750 mm (Zubenok, 1977). Solar radiation is maximum in summer months of May and June and substantially lower in winter months (Ahmad *et. al*., 1991).

Collection of Plant Material

The A sizeable number of mature capsules from seven trees of *T. populnea* grown in saline soils or raised under saline water irrigation were collected from various areas of Karachi (Hawkes Bay, North Nazimabad, Boat Basin and Karsaz areas) and stored in laboratory at room temperature till further analysis.

Soil Analysis

The surface and subsurface (0-30 and 30-60 cm deep, respectively) soil samples collected from underneath the sample plants were analyzed for their salinity status, pH and cationic content (Na and K) as per standard methods described in Richards (1954) using saturated soil extract.

Seed Oil Extraction

Seed-oil extraction was undertaken for seed lots of seven *T. populnea* plants (K1, K2, N1, B1, B2, B5, HB1) growing in soils of differential salinity (28.7, 43.93, 10.2, 25.7, 23.53, 11.2 and 1.56 dS.m-1, respectively). One hundred gram seeds from each seed lot were crushed and then subjected to extraction via Soxhlet extractor fitted with a 250 ml round bottom flask and a condenser. The extraction was done on a water bath for 7-8 hrs with 150 ml of n-hexane. The solvent was removed under reduced pressure. The oil was dried by anhydrous sodium sulfate, filtered and kept in separate sealed bottle at low temperature (0-40^{o}C) before use. (Khatri *et. al*, 1995; Ahmad *et. al.*, 1996*).*

Seed Oil Extraction and Analysis of Fatty Acid Composition

Oil from seeds of seven *T. populnea* plants from Hawkes Bay, Nazimabad, Boat Basin and Karsaz areas growing in soils of differential salinity (28.7, 43.93, 10.2, 25.7, 23.53, 11.2 and 1.56 dS.m-1, respectively) was extracted with n-hexane using Soxhlet extractor. Fatty acid composition was determined by using CALRUS model GC-500 (Perkins Elmer Company) gas chromatograph. Standard methods of AOCS (2003) were employed in the analysis. *T. populnea* seed oil (250 mg) was trans-estrified with 0.5% sodium methoxide in methanol (50 ml) under reflux. After 45 minutes, the solution was diluted with distilled water (25 ml) and extracted with hexane **(**3*20 ml). The combined extract was washed with H_2O (4*20 ml), dried over anhydrous sodium sulphate and the solvent removed under reduced pressure using a rotary evaporator. The methyl esters were diluted in hexane to obtain a solution for GC- analysis) (Conway *et al*., 1985; Fatima *et. al.,* 1992). The chemical composition of fatty acid methyl esters was accomplished with a Perkin Elmer gas chromatograph model CLARUS-500 fitted with a polar capillary column Sp-2330 (60 x 0.25 mm x 0.20 µm, film thickness) and a flame ionization detector. Oxygen free nitrogen was used as a carrier gas at a flow rate of 2.5ml/min. Other conditions were as follows: initial oven temperature, 70^{o}C for 5 minutes; ramp rate, 10^{o}C/min till 180^{o}C; ramp rate, 3^{o}C final temperature, 220^{o}C; injector temperature, 250^{o}C; detector temperature, 275^{o}C. A sample volume 0.5 µL was injected (splitless). Fatty acid methyl esters were identified by comparing their relative and absolute retention times to those of authentic standards of fatty acid methyl esters purchased from Supelco and Sigma-Aldrich Co. A built-in data-handling program, provided by the manufacture of the gas chromatograph, did the quantification. The analyses were performed in triplicate.

Saponification value

Five gram of the oil sample was taken in a quick fit conical flask and 50 ml of alcoholic KOH was added to it by pipette. The mixture was refluxed for one hour on water bath till complete saponification. The condenser was washed and rinsed with 10 ml distilled water. The flask was detached from the condenser and cooled. This was titrated with 0.5N HCl using Phenolphthalein an indicator to a colorless end point. The blank was run under the same conditions simultaneously. The saponification value was calculated as:

SV = (B-S) * 28.05/ weight of the sample,

Where B is the volume of 0.5N HCl used for blank and S is the volume of 0.5N HCl used for sample. All chemicals and solvents (analytical grade) used were from E. Merck or Sigma-Aldrich. Fatty acid methyl esters were purchased from Supelco (Belle forte, PA) and Sigma-Aldrich Co. (St. Louis, MO).

RESULTS AND OBSERVATION

Salinity Status

The soil associated with the sample trees ranged from Sandy Loam to Clay loam through sandy loam to silt loam in texture. The soil was differentially saline ranging from 1.56 – 43.93 $dS.m^{-1}$ and basic in reaction (7.3 – 8.7). The surface layer of the soil was generally more saline than the lower layers. It is a common feature in arid regions. The soil had K in low concentrations (Table 1).

Table1. Soil characteristics associated with the *Thespesia populnea* trees growing in the saline-arid environment of Karachi. ECe, pH and cations are based on saturated soil extract.

Soil Sample	Locality of sample *T. populnea* trees						
	HB	N1	B1	B2	B5	K1	K2
ECe: $dS.m^{-1}$							
A**	2.2	11.9	47.00	34.0	8.90	26.70	72.50
B	1.6	10.5	16.60	27.2	13.50	31.70	28.50
C	0.9	8.10	13.50	9.4	11.20	26.90	30.80
Mean	1.56	10.20	25.7	23.53	11.20	28.7	43.93
pH							
A**	8.55	6.25	8.75	8.75	8.55	8.4	7.65
B	8.45	7.4	8.6	8.5	8.45	8.25	7.60
C	8.10	8.25	8.8	8.7	8.35	7.95	8.05
Mean	8.37	7.3	8.7	8.65	8.45	8.2	7.80
Cations (meq / l) (0 – 60 cm profile)							
Sodium	-	54.35	304.3	391.3	391.3	336.96	380.43
Potassium	-	8.33	5.13	30.13	89.7	14.8	-
Soil Texture (0-60 cm profile)***							
0-60 cm profile	SL	L	SL – CLL	SL – CLL	STL-CLL	L	L

*, Locality - HB, Hawkes Bay sample 1; N1, North Nazimabad sample, B1,B2, B5, Boat Basin samples and K1-K2, Karsaz samples. **, A, Surface sample; B, 30 cm deep; C, 60 cm deep; ***, L, Loam; SL, Sandy Loam; STL, Silt Loam; CLL, Clay Loam.

Oil contents of the seeds

The oil of *Thespesia populnea* seeds is dark red in colour, syrupy in flow and possesses faint agreeable odour. The seed oil content varied from 18.2 to 21.07 % (mean = 19.81 ± 0.362 %) of the air-dried seed mass with as little variation as 4.8% amongst the seed samples analyzed (Table 2). Our results on oil content of seeds are in close agreement with the earlier studies. Menon (1910) reported oil contents in *T. populnea* seeds to be 19.6% of the whole seed mass and Subbaram (1954) reported similar results i.e., "20 % of the seed is oil" in *T. populnea* when extracted with petrol". Family Malvaceae on the basis of analysis of its 10 species is reported to contain seed oil around 14.0 % of seeds on dry weight basis (Levin, 1974). Sawan *et al.,* (2006) have reported cotton seeds to contain 19.6% oil. The seed oil content of *T. populnea* seeds is, therefore, comparable to that in cotton seeds.

Table 2. Percent yield of seed oil of *T. populnea* growing in saline environment of Karachi.

S. No.	Sample	Plant Locality*	Soil salinity (0 - 60 cm) Seed-oil Yield (%)	pH ECe: $dS.m^{-1}$
1.	K1	18.74	28.7	8.2
2.	K2	19.29	43.9	7.8
3.	N1	18.72	10.2	7.3
4.	B1	20.42	25.7	8.7
5.	B2	19.67	23.5	8.65
6.	B5	21.07	11.2	8.45
7.	HB	20.77	1.56	8.37
	Mean:	19.81 ± 0.362; CV: 4.83 %	20.68 ± 5.35, CV: 68.44%	8.37 ± 0.19

Saponification value of *T. populnea* seed -oil = 204

*, Locality – HB, Hawkes Bay sample; N1, Nazimabad samples; B1, B2, and B5, Boat Basin samples and K1-K2, Karsaz samples

. The saponification number of *T. populnea* seed oil was found to be 204 which is little higher than that of cotton seed oil (193 – 195), peanut (190 – 196), Soybean (193), olive oil (185 -196) and *Moringa olifera* (181.4,), comparable to palm oil (191 – 205) but lower than that of coconut (246 – 260) and palm kernel oil (242 – 246) (Lewkowitsch, 1922; Anwar and Rashid, 2007). Low saponification value of *T. populnea* seed oil as compared to coconut and palm kernel oil may be due the absence of Lauric acid and very low concentration of Myristic acid in *Thespesia* oil. The large proportion of these two fatty acids in coconut and palm kernel oil is considered to be reason of high saponification of these oils. Saponification value is considered to be an indicator of the usefulness of the oil in sense that larger the saponification value, more soluble is the soap that may be made from the oil, if the oil is moisture free and unsaponifiable matter in it are negligible (Lewkowitsch, 1922).

Fatty Acid Composition

Twelve fatty acids were identified in *T. populnea* seed oil of which Linoleic and Palmitic acids were the major fatty acids occupying a proportion of 37.27 and 31.96%, respectively (Table 3). Oleic acid was the third important fatty acid (17.73% on average basis). Proportion of several fatty acids identified viz. Myristic, Palmitoleic, Arachidic, Bahenic, Lignoceric acid, etc. was much low (less than 1%) except Elaidic acid being around 7%. Stearic acid was 2.95% of the total fatty acid contents. The proportion of total unsaturated fatty acids was 63.3% as against 35.3% saturated fatty acids. The variation of fatty acid concentration (in terms of coefficient of variation) among the source trees was generally low in case of fatty acids synthesized in higher concentration e.g., Palmitic, Myristic, oleic, and Linoleic acids (Cvar: 2.02 – 7.3%), moderate in Palmitoleic, Cis-10-Heptadecanoic acid (C17:0), Stearic, Elaidic, and Bahenic acids (Cvar: 8.6 – 28.0 %) and high in arachidic, lignoceric and cis-4,7,10,13,16,19-Docosahexaenoic acids (C22:6) (Cvar - 39.36 – 45.28%) synthesize in lower concentrations. Larger the concentration of a fatty acid, lower the variation in its concentration among trees (Fig. 1). The concentration of ω3 fatty acid, represented by cis-4,7,10,13,16,19 – docosahexanoic acid (C22:6), was low and ranged from 0.12 to 0.31g per 100g fatty acids in the seeds (mean = 0.167 ± 0.0273 %).

Table 3.Fatty acid composition of the seed oil of *T. populnea* trees growing in differentially salinity-affected soils of Karachi.

	FATTY ACID CONTENT (g / 100g Fatty acids) Plant Code								
Fatty acid	K1	K2	N1	B1	B2	B5	HB	Mean ± SE,	Cvar (%)
Myristic acid (C14: 0)	0.65	0.70	0.69	0.60	0.64	0.58	0.60	0.637 ± 0.0176	7.30
Palmitic acid (C16: 0)	32.24	32.90	32.57	31.77	31.09	31.76	31.38	31.959 ± 0.244	2.02
Palmitoleic acid (C16: 0)	0.69	0.38	0.40	0.67	0.51	0.85	0.67	0.596 ± 0.0649	28.80
Cis-10-Heptadecanoic acid (C17: 0)	0.77	0.86	0.81	1.00	0.88	0.93	0.87	0.874 ± 0.0285	8.63
Stearic acid (C18:0)	2.82	3.06	3.44	2.66	3.27	2.86	2.54	2.950 ± 0.123	11.00
Elaidic acid (C18:1t)	6.47	7.10	6.51	7.14	6.31	7.62	8.01	7.02 ± 0.240	9.06
Oleic acid (C18: 1c)	16.53	16.45	17.36	19.42	17.41	17.88	16.59	17.732 ± 0.397	5.93
Linoleic acid (C18: 2)	38.23	37.62	37.16	35.48	38.54	35.71	38.13	37.267 ± 0.464	3.29
Arachidic acid (C20:0)	0.77	0.50	0.38	0.31	0.36	0.31	0.33	0.423 ± 0.063	39.36
Bahenic acid (C22: 0)	0.10	0.12	0.15	0.18	0.15	0.12	0.11	0.133 ± 0.0106	21.14
Lignoceric acid (C24: 0)	0.12	0.09	0.11	0.27	0.13	0.12	0.10	0.134 ± 0.0232	45.77
Cis-4,7,10,13,16,19 -Docosahexaenoic acid (C22:6)	0.16	0.12	0.31	0.14	0.18	0.18	0.08	0.167 ± 0.0273	43.28
Total amount of saturated fatty acids	36.70	37.37	37.34	35.99	35.61	35.75	35.06	35.26 ± 0.338	2.53
Total amount of monounsaturated fatty acids	24.49	24.79	25.08	28.23	25.11	27.28	26.14	25.87 ± 0.532	5.44
Total amount of polyunsaturated fatty acids	38.39	37.74	37.47	35.62	38.72	35.89	38.21	37.43 ± .0461	3.26
Total unsaturated fatty acids	62.87	62.53	62.55	63.85	63.83	63.17	64.35	63.31 ± 0.269	1.12

Table 4. Percent proportion of unsaturated and saturated fatty acids and their ratio in the seed-oil samples of *Thespesia populnea*. TU / SA ratio in cotton is given for comparison.

Thespesia populnea						Cotton – TU / SA
Plant Code	SA	MU	PU	MU + PU	TU / SA	
K1	36.70	24.49	38.39	62.88	1.71	2.75–3.02 % *
K2	37.37	24.79	37.74	62.53	1.67	
N1	37.34	25.08	37.47	62.55	1.68	
B1	35.99	28.23	35.62	63.85	1.77	
B2	35.61	25.11	38.72	62.83	1.79	
B5	35.75	27.28	35.89	63.17	1.77	
HB	35.06	26.14	38.21	64.35	1.84	
Mean =	36.40	25.87	37.73	63.61	1.75	
SE =	0.322	0.532	0.461	0.269	0.024	
CV(%) =	2.34	5.44	3.26	0.711	3.56	

*, Sawan *et. al.* (2006); Acronyms – SA, Saturated; MU, Monounsaturated; PU, Polyunsaturated; TU, Total Unsaturated fatty acids

Over the ECe range of 1.56 to 43.03 dS.m-1 (Table 2), the variation in fatty acid composition of seed oil of *T. populnea* was too small to make any significant difference in the oil quality as is evident from the compositional similarity amongst the oil samples taken out from various trees (Table 5**).** It is then certain that *T. populnea* may be grown under saline conditions without any qualitative change in its seed oil.

Subbaram (1954) have carried out fatty acid composition of *T. populnea* seed oil from Madras (India) and reported only five fatty acids (Myristic (1%), palmitic (21.4%), Stearic (1.9%), Oleic (32.5%) and Linoleic acid (43.2%). Although composition of *Thespesia* seed oil as found in the present case is substantially different from that reported by Subbaram (1954), yet the major fatty acids are the same – linoleic (37.27%), oleic (17.73%) and Palmitic (31.96%) - the variation largely being in concentration of Palmitic and Oleic acids. Such a compositional variation may, besides possible methodological differences in extraction in the two cases, may also be probably attributable to the environmental differences in plant's habitat in Madras (South India), an area of potentially heavy rains, and Karachi (Pakistan), an area deficient in rains. The environmental influence on the fatty acid composition is known from the literature in at least micro-organisms (Olsen and Ingram, 1975; Wada and Murata, 1990). The share of linolenic and linoleic acids of whole quantity of fatty acids is reported to have negative correlations with temperature in rapeseed, sunflower, and soybean (Werteker, 2010). There is a general theory that production of polyunsaturated fatty acids (PUFAs) is enhanced under low temperature that facilitates to keep the microviscosity of biological membranes low for necessary metabolic activities.

From literature regarding oil content in seeds of some conventional and non-conventional plants, it clear that seed oil content in *T. populnea* seeds is some what low than that in *Lepidium sativum* (23.08%), much lower than that in *Eruca sativa* (33.46%) and *Thlaspi arvense* (29.8%) – the cruciferae plants (Dolya *et. al.,* 1976). The oil content in *Thespesia* seeds is almost half to that in *Linum usitatissimum* L. ssp. *Usitatissimum* (mean = 38.3 ± 1.74 %) on the basis of observation on 2934 accessions from 72 countries (Diederichsen and Raney, 2006). CANOLA oil yield is also reported to be much higher (32.7 – 37.8%) than *Thespesia* (Starner *et al.,* 1999). *T. populnea* seed oil yield is also much lower than that of coconut varieties (from 49.25 % - Chowgat dwarf green variety to 88.54 % in Adaman giant) (Akapan *et al.,* 2006). *Jatropha curcas* (Euphorbiaceae) seed oil yield is also much higher (67% on aqueous extraction of kernels of seeds) (Shah *et al.,* 2005). Family Malvaceae on the basis of analysis of 10 species is reported to contain seed oil around 14.0 % on dry weight basis (Levin, 1974). Sawan *et al.* (2006) have reported cotton seeds to contain 19.6% oil. The seed oil content of *T. populnea* seeds is, therefore, comparable to that in cotton seeds (Table 7).

Table 5. Similarity* among *Thespesia* seed-oil samples (extracted from seeds of various trees growing under wide range of soil salinity). Similarity calculated on the basis of fatty acid composition (a) and proportion of saturated and total unsaturated (mono- and polyunsaturated) fatty acids (b) in the oil.

K1	K1						
K2	a) 98.2	K2					
	b) 98.9						
N1	97.8	98.3	N1				
	98.6	99.6					
B1	95.6	95.9	96.3	B1			
	96.1	96.4	96.7				
B2	97.7	97.1	97.8	95.6	B2		
	98.3	98.1	98.2	96.7			
B5	96.0	96.3	96.7	97.7	96.1	B5	
	96.1	96.4	96.7	98.6	96.6		
HB	97.7	97.0	96.6	95.8	97.3	96.6	HB
	97.7	97.5	97.9	96.8	98.3	97.1	

*, calculated on the basis of Czekanowski's (1913) index of similarity as SI = 2 min (xi, Yi) / ∑ (xi + Yi) x 100, where Xi equals the concentration of a fatty acid belonging to one oil sample and Yi is this measure for the fatty acid in the other oil sample. Location & Dispersion parameters are**:**

a) N = 21, mean = 96.86, SE = 0.1912, Min. = 95.6, Max. = 98.3, Cvar (%) = 0.90

b) N = 21, mean = 97.40, SE = 0.2330, Min. = 96.1, Max. = 99.6, Cvar (%) = 1.05

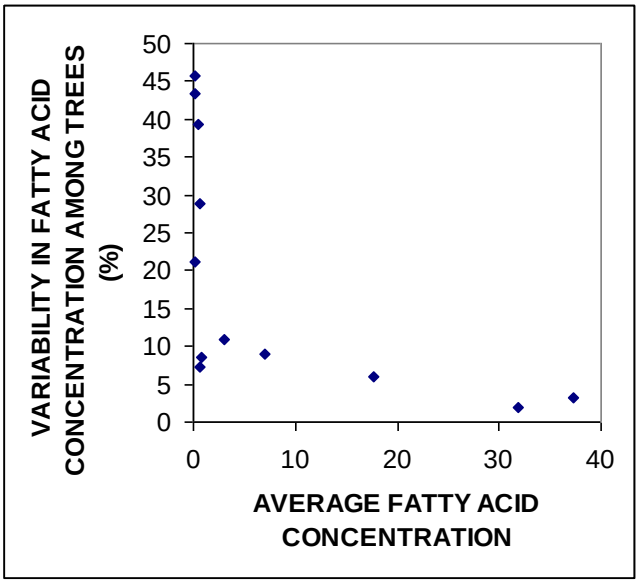

Fig.1. Relationship between % variability in concentration of 12 fatty acids among *Thespesia populnea* trees and average fatty acid concentration in their seed oil.

The amount of saturated fatty acids (SA) in *T. populnea* seed oil was substantially lower than that of total unsaturated fatty acids (TU). TU/SA ratio ranged from 1.67 to 1.84 (mean = 1.75 ± 0.024) and the variability of TU/SA ratio in terms of coefficient of variability was only 3.56 % (Table 4). The TU/SA ratio was found to be much lower than that for cotton seed oil (2.75 – 3.02; Azhar *et al.,* 1999). Cucci *et al.* (2010) has reported increase in oleic / linoleic acid ratio in sunflower with increase in salinity. No such increase in oleic / Linoleic acid ratio in *T. populnea* seeds was observed with the rise of salinity (Fig. 2). It remained almost constant - around 0.48.

CANOLA oil is considered to be the best oil for human consumption. The oil content in CANOLA seeds is good, c 40% and large share of 90% of the fatty acids is constituted by unsaturated fatty acids (Declercq and Daun, 1998). In comparison *T. populnea* oil content in seeds is exactly half to that in CANOLA. The share of total unsaturated fatty acids is 63% which indicates that it may be of potential human use. Saponification number in *Thespesia* seed oil is 204 being little higher indicates the presence of higher molecular weight fatty acids. The oils of low saponification number are thought to be better for human consumption. Further studies are needed to determine the usefulness of *T. populnea* seed oil as human diet or in the industry. There are some limitations with *Thespesia* oil as regard to its inclusion in human diet.

Gossypol ($C_{30}H_{30}O_8$) is a well-known principle from cotton seeds, a compound that has contraceptive effects in humans. It causes hypokalemia as a result of kidney malfunction, and causes symptoms of fatigue, muscle weakness and its most extreme effect, the paralysis. (+)-Gossypol has also been reported in bark and fruit of the *T. populnea* also (Datta *et. al*., 1972). The presence of gossypol may likely be expected in *T. populnea* seed oil. It has been reported that oral administration of (+)-gossypol in dose levels of 10, 30 and 100 mg/kg showed 33%, 63% and 79% anti-implantation activity in female albino rats respectively (Murthy *et. al*., 1981). The floral extract of *T. populnea* has been reported to exhibit anti-steroidogenic activity in mouse ovary (Kavimani, *et. al*., 1999). The flowers contain kaempferol, kaempferol-7-glucoside and gossypetin and fruit kernels are reported to contain-sitosterol, ceryl alcohol and a yellow pigment, thespesin (Srivastava *et al.,* 1963; Data *et. al.,* 1973). A mixture of two groups of long chain fatty acids extracted in petroleum ether and ethyl acetate and subsequent crude alcoholic extract from seeds of *T. populnea* showed anti-implantation activity in rats (Ghosh and Bhattacharya, 2004). Schmidt *et al.* (1994) have developed and patented a technique of gossypol removal from the cotton seed oil by using urea in a borate containing buffer. Rathore (2007) has asserted that cotton seed oil could be used for edible purpose after gossypol removal. Since technique of Schmidt *et al.* (1994) is also applicable on several malvaceous plants other than cotton as well, such a process should remove gossypol from *Thespesia* seed oil also.

Table 8. Comparative account of yield and composition of seed oil of some plants including *T. populnea*.

species	Yield (%) from seed mass	Fatty acid (%)						
		Oleic	Stearic	Linoleic	Palmatic	Lino-lenic	Lauric	Reference
Neem (India)	20 -32.6	-	-	-	-	-	-	Kaura *et. al* (1998)
Neem (Mexico)	15.4 – 24.5	45.55	15.23	16.67	17.21	1.33	-	Munoz – Valenzuela *et. al.* (2007)
Sesame - cultivars	45.37- 45.96	-	-	-	-	-	-	El-Nakhlawy & Shaheen (2009)
Moringa	34.80	73.22	5.50	-	6.45	-	-	Anwar and Rashid (2007)
Coco nut*	71.24	6.13 ± 0.58	3.15 ± 0.15	2.00 ± 0.22	10.33 ± 0.11	-	41.43 ± 1.18	Akpan *et al.* (2006)
Coco nut	-	6.0	3.0	-	5.0	-	45.0	Webb (1926)
CANOLA	32.7 – 35.8	-	-	-	-	-	-	Starner *et. al.* (1999)
CANOLA	40.0	Unsaturated fatty acids = 90%						Declercq & Daun (1998)
Pea nut		51.60	6.00	26.0	8.50	-	-	Webb (1926)
Soybean	-	20.0	2.0	64.0	11.0	3.0	-	
Maize	-	44.0	2.0	48.0	6.0	-	-	
Olive	-	51.60	6.00	26.0	8.50			
Nelumbo nucifera (lotus)	3.62	11.7	3.0	19.9	33.3	3.4	2.04	Hamed *et al.* (2004)
Hippophae rhamnoides	Seed: 10 Pulp: 4.5	-	2.6 1.5	29.6 12.5	11.1 34.5	-	-	Abiid *et al.* (2007)
Cotton	18.3 – 24.5P 18.5– 23.9H	-	-	-	-	-	-	Azhar *et al.* (1999)
Cotton	19.6	-	-	-	-	-	-	Sawan *et. al* (2006)
Cotton	-	31.6	-	45.0	23.4	-	-	Webb (1926)
Arthrocnemum indicum (16) **	22- 25%	-	3.17	63.02	26.93	-	0.23	Weber *et al.* (2007)
Alhagi maurorum (13) **		-	11.01	53.28	29.38	-	0.11	
Cressa cretica (08) **		-	8.26	62.7	25.75	-	0.0	
Halopyrum mucronatum (14) **		-	3.27	68.43	21.79	-	0.0	
Haloxylon stocksii (9)**		-	3.94	66.24	24.20	-	0.38	
Suaeda fruticosa (13)**		-	4.61	72.08	17.04	-	0.15	
Salvadora persica	43.3-45.5	-	-	-	-	-	40-45	Reddy *et al.*, (2008)
T. populnea	20.0	32.50	1.90	43.2	21.4	-	-	Subbaram (1954)
T .populnea (12) **	19.81	17.73	2.95	37.27	31.96	-	-	Present investigation

P, Parents; H, Hybrid plants. *, data are the means for dry samples of six coconut varieties viz. dwarf green, PB121, Tall green, Dwarf Orange, ordinary and Adaman Giant. According to Akapan *et al.* (2006), these coconut varieties contain 88.2% saturated fatty acids. **, Number of fatty acids detected.

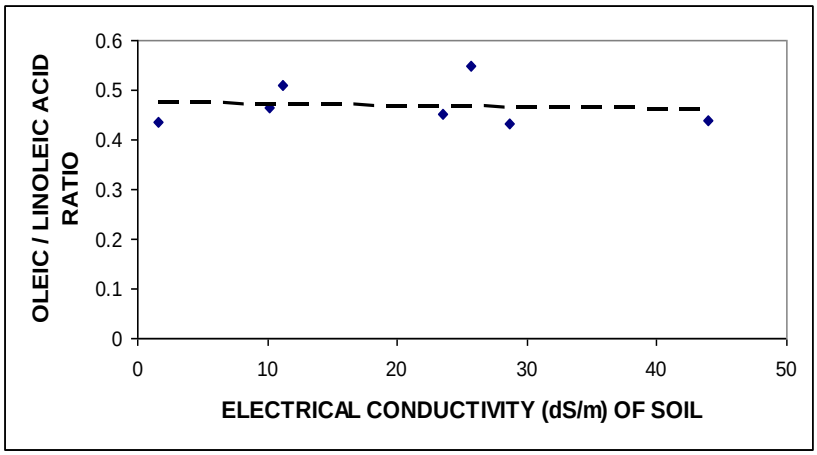

Fig. 2. Relationship between Salinity and ratio of oleic acid to linoleic acid concentration in seed oil of *T. populnea*.

REFERENCES

Abedin, S. (1979). Malvaceae. *Flora of West Pakistan*. 130: 1 - 107.

Abid, H., A. Hussain and S. Ali (2007). Physicochemical characteristics and fatty acid composition of Sea-Buckthorn (*Hippophae rhamnodes* L.) oil. *J. Chem. Soc. Pakistan*. 29 (3): 256-259.

Ahmad, F., S.M. Aqil Burney and S.A. Hussain (1991). Monthly average daily global and diffuse solar radiation and its correlation with hours of bright sunshine of Karachi, Pakistan. *Renew. Energy* 1: 115 – 118.

Ahmed, M., M. Naveed, M. A. Javed, T. A. Khan, and M. Y. Raie (1996). "Lipid studies of *Annona Squamosa", Pak. J. Sci. Ind. Res.* vol. 139, 9-12.

Akapan, E.J. O.E. Etim, Akapan, H.D. and I.F. Usoh (2006). Fatty acid profile and oil yield in six different varieties of fresh and dry samples of coconuts (*Cocos nucifera*). *Pak. J. Nutrition* 5 (2): 106 – 109.

Anwar, F. and U. Rashid (2007). Physico-chemical characteristics of *Moringa oleifera* seeds and seed oil from a wild provenance of Pakistan. *Pak. J. Bot.* 39(5): 1443 – 1453.

Aronson, J.A.(1989).Haloph - A Database of Salt Tolerant Plants of the World (Ed. E.E. Whitehead). Office of Arid Lands Studies. The University of Arizona. Tucson, Arizona. x + 75 pp.

Ashton, E.C. and D.J. Macintosh (2002). Preliminary assessment of the plant diversity and community ecology of the Samartan mangrove forest, Sarawak, Malaysia. Forest Ecology & Management 166: 111-129.

Azhar, F.M., S.U.K. Ajmal and A.I. Khan. 1999. Combining ability of *Gossypium hirsutum* L. parents for seed oil contents. *Pak. J. Biol. Sciences* 2(4): 1458 – 1461.

AOCS (2003). Official Methods and Recommended Practices of the AOCS. 5TH Edition,

Binder-Barhava, N. and B. Ramati. (1967). Tolerance of three olive varieties to soil salinity in Israel. *Exp. Agric*. 3: 295 – 305.

Conway, J., W. M. N. Ratnayake and R. G. Ackman. (1985). "Hydrazine Reduction in the Gas Liquid chromatographic Analysis of the Me-esters of cyclopropenoic fatty acid*". J. American Oil Chemist Society*, vol. 62, No. 9:1340-1343.

Cucci, G., T. Rotunno, A. de Caro, G. Lacolla, R. Di Cateria and E. Tarantino (2010). Effects of saline and sodic stress on yield and fatty acid profile in sunflower seeds. *Italian J. Agronomy (eISSN 2039-6805)*. DOI: 10.4081/ija.2007.13

Czekanowski, J.(1913). Zarys method statystycznych. Die Grunduge der Statischjen methoden) Warsaw.

Datta S. C, V.V.S. Murti, and T.R. Seshadri.1972 Isolation and study of (+) - Gossypol from T*hespesia populnea. Indian J Chem*. 10:263-6.

Datta S. C, V.V.S.Murti, N. N. Sharma, and T.R. Seshadri.1(973). Glycosidic components of *Thespesia populnea* flowers. *Indian J Chem*.11:506-7.

Declercq, D.P. and J.K. Daun (1998). Quality of 1997 Ontario CANOLA. Final Report, Grain Research Lab. Winnipeg, Manitoba, Canada: Canadian Grain commission.

Diederichsen, A. and J.P. Raney. (2006). Seed colour, seed weight and seed oil content in *Linum usitatissimum* accessions held by plant gene resources of Canada. Plant Breeding 125 (4): 372 – 377.

Dolya, V.S., K.E. Korshchuk, E.N. Shkurpii and N.A. Kaminskii. (1976). Oil of three representative of the family Cruciferae II. *Chemistry of Natural Compounds* 10(4); 447 – 449.

El-Nakhlawy, F.S. and M.A. Shaheen. (2009). Response of seed yield, yield components and oil content to the sesame cultivar and nitrogen fertilizer rate diversity. *Electronic J. Environmental, Agricultural and Food Chemistry*. 8(2): 287 – 293. (ISSN-1579-4377).

Fatima, R., R. B. Quadri, V. Ahmed and K. Fizza.(1992). " Poly unsaturated PUFA" Proceeding of first National Biochemistry Symposium, March 1992, pages 1-7. Editors: S. N. Hasnain, M. A. Haleem, H. S. A. Ather. B.C.C. & T. Press, University of Karachi.

Gosh, K. and T. Bhattacharya. (2004). Preliminary study of compounds from the extracts of seeds of *Thespesia populnea*. *Ind. J. Pharmacology* 36 (15): 288 – 291.

Hamed, S., H. Akhtar, A. Waheed and I. Khokar (2004). Fatty acid composition of lipid classes of *Nelumbo nucifera* seed oil. *J. Chem. Soc. Pakistan*. 26 (4): 382- 385.

Khan, D., M. Faheemuddin, S.S. Shaukat and M. M. Alam (2000). Seasonal variation in structure, composition, phytomass and net primary productivity in a *Lasiurus scindicusa* Henr and *Cenchrus setigerus* Vahl dominated dry sandy desert site of Karachi. *Pak. J. Bot.* 32: 171-210.

Kaura, S.K., S.K. Gupta and J.B. Chowdhury. (1998). Morphological and oil content variation in seeds of *Azadirachta indica* A. Juss. (Neem) from northern and western provenances of India. Plant Foods for Human Nutrition 52: 293 – 298.

Kavimani S, R. Ilango, S. Karpagam, K. Suryaprabha, and B. Jaykar.(1999). Antisteroidogenic activity of floral extract of *Thespesia populnea* Corr. in mouse ovary. Indian J Exp Biol 37:1241-2.

Khatri, L.M., M. K. A. Nasir, R. Saleem and F. Noor. (1995). "Characteristics and chemical composition of the fixed oil of *Achras zapota* seeds". Pak. J. Sci. Ind. Res. 38: 11-12.

Levin, D.A.(1974). The oil content of seeds: An ecological perspective. The Am Nat. 108 (96^{o}): 193 – 206.

Lewkowitsch, J. (1922). Chemical Technology and Analysis of Fats, and Waxes. Pp. 395 – 400.

Levin, D.A.(1974). The oil content of seeds: An ecological perspective. The American Naturalist 108 (960): 193 – 206.

Menon, A.K. (1910). J. Soc. Chem. Ind. 29: 1431. (Seen in Subbaram (1954).

Murphy, D.J. (1996). Engineering oil production in rapeseed and other oil crops. *Tibitech* 14: 205-213.

Murthy R.S.R., D.K. Basu, and V.V. S.Murti. 1981. Antifertility activity of (+) gossypol on female albino rats. Indian J Pharmacol 13:86.

Munoz-Valenzuela, S., A.A. Ibarra-Lopez, L.M. Rubio-Silva, H. Valdez-Davila and J. Borboa-Flores. (2007). Neem tree morphology and oil content. *Issues in New crops and Use*s. (eds. J. Janick A. Whipkey). ASHS Press, Alexandria, VA.

Olsen, G. J., and L.O. Ingram (1975). Effects of temperature and nutritional changes on the fatty acids of *Agmenellum quadruplicatum*. *J. Bacteriol*. 124: 373 – 379.

Qadir, S.A., S.Z. Qureshi and M. Ahmad (1966). A phytosociological survey of Karachi University Campus. *Vegetatio* 13: 339 – 362.

Rathore, K.S. (2007). Reducing gossypol in cottonseed may improve human nutrition. ISB News Report. July 2007.

Richards, L.A. (Ed.)(1954). *Diagnosis and improvement of saline and Alkali Soils*. Agric. Handbook # 60. USSLS / USDA, Washington.

Sawan, Z.M., S.A. Hafez, A.E. Basyony and Abou-El-Ela R. Rahman. (2006). Cottonseed, protein and oil properties as influenced by potassium fertilization and foliar application of Zinc and Phosphorus. *World J. Agric. Sciences* 2 (1); 66 – 74.

Reddy, M.P., M.T. Shah and J.S. Patolia (2008). *Salvadora persica*, a potential species for industrial oil production in semi-arid saline and alkali soils. *Industrial Crops and products* 28:273-278.

Schmidt, J.H., R. Wells and J.C. Bailey. (1994). Method for removal of gossypol from cotton seeds meal by the use of urea in a borate containing buffer. US patent # 5277909.

Shah, S., A. Sharma and M.N. Gupta. (2005). Extraction of oil from *Jatropha curcas* L. seed kernel by combination of ultrasonication and aqueous enzymatic oil extraction. *Bioresource Tech*. 96(1): 121 – 123.

Starner, D.E. A.A. Hamama and H.L. Bhardwaj. (1999). Canola oil yield and quality as affected by production practices in Virginia. Perspective in New Crops and New USES. (ed. By J. Janick) ASHS Press, Alexandria.

Subbaram, M.R. (1954). Analysis and characteristics of oil from seeds of *Thespesia populnea*. *Proc. Indian Acad. Sciences* (section A) No. 1 Jan. 1954. Vol. 39, Pages 301 -304.

Wada, H. and n. Murata (1990). Temperature-induced changes in the fatty acid composition of *Cyanobacterium synechocystis* PCC6803. *Plant Physiol.* 92: 1062-1069.

Webb, E.T.(1926). Oils and Fats in Soap Manufacture. Soup Gazette and Perfumer. Oct. 1926. XXXVIII, 302.

Weber, D.J., R. Ansari, B. Gul and M. A. Khan (2007). Potential of halophytes as source of edible oil. *J. Arid Environs*. 68: 315 – 321.

Werteker, M., A. Lorenz, H. Johannes, E. Berghofer and C.S. Findlay (2010). Environmental and varietal influences on the fatty acid composition of rapeseed, soybeans and sunflowers. *J. Agron. & Crop Sci*. 196: 20-27. doi: 10.1439-037X.2009.00393.x

Zubenok, L.I. (1977). Annual potential evapo-transpiration Map sheet no. 18 In: Atlas of World Water Balance (M.I. Budyko, Ed.) UNESCO, Press, Paris.

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