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Effect of Soil Type on Growth Vigour, Water Relations, Mineral Uptake and Contents of Fatty Acids and Protein of Yielded Seeds of *Linum usitatissimum*

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Abstract: Flax (*Linum usitatissimum*) were studied at three types of soil collected from new sites in Egypt (Western Mediterranean Coastal region, East of El-Kantara in Sinai, and Tushki is the new valley). The plants were subjected to a pot experiment cropped with *Linum*. Physical and chemical analyses were carried out for each soil. The effect of different soil types on growth criteria, total leaf conductivity, transpiration, photosynthetic pigments, ionic contents and the rate of transport of elements, percentage of fiber as well as protein pattern and fatty acid contents of the yielded seeds were studied. The growth of flax plants at various soils led to a significant decrease in all growth criteria and total pigments, however, total leaf conductivity and transpiration rates were increased at El-Kantara and Tushki sites and decreased at Northern Coastal site comparing with control. Comparing, the three soils, it was apparent that, at Northern Coastal site, there was a high seed yield than the other two sites. At El-Kantara, there was high percentage of fiber than the others. The amount of oil recorded a highly significant increase at Northern Coastal and Tushki site, while a significant decrease at El-Kantara site. The rate of transport of elements (J) for K, Na and Ca showed a significant decrease except for J_{Na} and J_{Ca} at Northern Coastal and for J_{Na} at El-Kantara site. Variable changes were obtained in the composition of polypeptides with a wide range of molecular weights and for the constituent saturated and unsaturated fatty acids.

Key words: *Linum usitatissimum*, water relations, mineral uptake, protein pattern, fatty acids

Introduction

Plant growth depends on the supply of inorganic nutrients. Typically, plants are exposed to nutrient levels that vary widely in both time and space. To cope with such fluctuating environments, plants have developed high levels of plasticity both at the individual level and within species and ecotypes. Nevertheless, extreme nutrient conditions will cause deficiency or toxicity to a varying extent for different plant species.

Sub-optimal rates of supply of mineral nutrients can reduce leaf growth rate, which may lead to reduction in plant photosynthetic capacity and yield (Snir & Neumann, 1997). Different growth responses may be appropriate for different types of target environments. For example, when plants are grown in arid and semi-arid regions, under low-input conditions, where combinations of soil salinity and terminal drought are expected, early onset of salt induced reduction in leaf growth rates and yields may represent an adaptive response (Tanji, 1990). Thus, early growth inhibition, by limiting transpirational losses and prolonging the availability of limited soil water reserves, could prolong the survival during dry months for long enough to allow the attainment of some reproductive yield (Neumann, 1995b).

For high-input crops produced under intermittent irrigation with moderately saline water, survival is not usually in question and yield is the main concern. In this case, early onset of inhibition of growth rates by the build up of soil salinity could directly limit the plant size and yield. Moreover, on going inhibition of leaf growth also decreases the volume of new leaf tissues into which excess salt ions can be safely accumulated. Continuous salt accumulation combined with limited production of new leaf volume could then lead to earlier build up of excess levels of salt. This might further accelerate the onset of leaf senescence and necrosis (Munns, 1993).

Salinity can rapidly inhibit the root growth and hence capacity for uptake of water and essential mineral nutrients from the soil (Neumann, 1995a). The influence of salt stress is aggravated by the simultaneous action of other xerothermic factors, particularly high temperature. The strategies and mechanism of plant adaptation to the synergistic action of

salinity and high temperature, cross-protection of salt stress by heat shock (HSP_s) has been demonstrated in few studies (Kuznetsov *et al.*, 1993).

Drought is a multidimensional stress affecting plants at various levels of their organization. The effect and plant response is most complex because it reflects the integration of stress effects and responses at all underlying levels of organization over space and time (Blum *et al.*, 1990). Drought caused a more pronounced inhibition in growth and photosynthetic rates in sensitive wheat (Loggini *et al.*, 1999).

The aim of present work was to throw light on the effect of different soil types on growth, water relations, rate of transport of elements, yield of fiber and seeds, protein pattern, oil contents as well as the composition of fatty acids, of *Linum usitatissimum*.

Materials and Methods

The present investigation was carried out during winter season (1998) using the local variety of flax (*Linum usitatissimum* L. var. Giza 7). Three sites were selected, the Northern Coastal area (i.e. 249 km north-west of Cairo); El-Kantara East (i.e. 180 km northeast of Cairo) and Tushki (i.e. 1150 km south of Cairo). The soils were collected from these sites. An experiment was carried out in pots (25 cm in diameter) containing equal amounts of soil. Pots were divided into four groups, each representing one type of soil besides the control set (garden soil, sand-clay; 2:1).

A similar lot of flax seeds were surface sterilized with 0.001 M HgCl₂ solution for three minutes and then washed thoroughly with distilled water. The sterilized seeds were divided into four sets. Each set was sown in each respective soil type. Irrigation was carried out according to the usual practice. Plants were exposed to normal day length with natural illumination in a green house of Faculty of Science of Mansoura University on October. The day/night temperature was about 25/11 ± 2°C.

Throughout the growth of plants, sampling was carried out at vegetative (i.e. 30-day old) and flowering (i.e. 70-day old) stages. At the time of sampling, 20 plants were collected at random from each of the selected soil and separated into

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shoots and roots for measurement of growth parameters, pigments as well as the ionic contents. At the termination of experiment stem diameter was determined using micrometer as well as percentage of fiber, protein fractionation and fatty acid contents were determined in the yielded seeds.

Analytical procedures

Plant analysis

Definitions and calculations :

Seed index = weight of 100 seeds in g.

Root/Shoot ratio (R/Sh) = W_R/W_S

$$\text{Relative growth rate (RGR)} = \frac{\text{Log}_e W_2 - \text{Log}_e W_1}{t_2 - t_1} \text{ gg}^{-1} \text{ days}^{-1}$$

Where W_R , W_S and W are the dry weights of roots, shoots and whole plant respectively. W_2 and W_1 are the plant dry weights at time t_2 and t_1 respectively. These were calculated according to the formula of Hunt (1982).

Estimation of pigments: Pigments (chlorophyll a, chlorophyll b and carotenoid) were determined by the spectrophotometric method as recommended by Metzner *et al.* (1965).

Retting and fiber extraction: Retting was carried out according to the procedure of Sharma (1988).

Measurements of total leaf conductance and transpiration rate and leaf temperature:

Total leaf conductance and transpiration rate of the second fully expanded leaf of *Linum* plants were measured using Li-1600 M steady state Porometer. The atmospheric pressure (PRES SET) and aperture area of the apparatus were adjusted to 101.3 Kpa and 1 cm² respectively (Ibrahim, 1999).

Estimation of cations: Cations were determined according to the method of Chapman and Pratt (1978). Flame-emission spectrophotometry was used for determining potassium and sodium, while calcium was measured by atomic absorption spectrophotometry.

The rate of transport of elements from root to shoot was estimated as:

$$J_{j,r} = (M_{s,2} - M_{s,1}) / (W_{r,2} - W_{r,1}) \times \text{RGR}_r$$

where :

$J_{j,r}$ is the transport of ion j from root to shoot.

$(M_{s,2}-M_{s,1})$ is the change in ion content of the shoot from time 1 to time 2.

$(W_{r,2}-W_{r,1})$ is the change in dry weight of the root from time 1 to time 2.

RGR is the relative growth rate of the root on a dry weight basis over this period.

These were calculated according to Ternaat and Munns (1986).

Data were first subjected to analysis of variance (ANOVA). If ANOVA showed significant ($p < 0.05$) effect, the least significant difference (LSD) was used to compare treatments (Snedecore and Cochran, 1980).

Protein banding pattern : Electrophoretic protein profiles of *Linum* seeds were analyzed by SDS-PAGE technique (Laemmli, 1970). Data were analyzed and identified by gel documentation system (GDS), with comparing polypeptide maps, molecular protein markers, percentages of band

intensity, molecular weight and mobility rate of each polypeptide in relation to standard markers using gel pro-analyzer version 3 MEDIA CYBERNE TICE Imaging Experts Software.

Determination of oil content : The methods followed for extraction and determination of the oil content were those described by Meara (1955).

Fractionation, identification and quantification of fatty acids:

For methylation of fatty acids for gas-liquid chromatographic analysis, the method used was essentially that adopted by Sink *et al.* (1964). One μL of fatty acid methyl ester was injected into a 6 feet x 1/8 inch internal diameter column packed with 20% diethylene glycol succinate (DEGS) on chromosorb 60-80 mesh. Fatty acid methyl ester were identified by peaks comparison with those of authentic standards and quantitated by integration of peak areas (Ferrante *et al.*, 1983).

Soil analyses: Soil samples collected from each site air dried, thoroughly mixed and passed through 2 mm sieve to remove debris. Physical and chemical analyses were carried out (Jakson, 1965) (Table 1).

Table 1: Chemical and physical data for experimental soil.

	Control	Northern coast	El-Kantara	Tushki
Chemical analysis				
Organic carbon %	0.80	0.48	0.30	0.72
CaCO ₃ %	3.40	40.65	1.15	8.9
HCO ₃ ⁻ %	0.06	1.20	0.051	0.33
CO ₃ ⁻ %	-	-	-	-
Cl ⁻ %	0.018	0.025	0.017	0.015
SO ₄ ⁻² (mg/100gm D. wt.)	0.08	0.067	0.046	0.064
NO ₃ ⁻ "	14.5	7.20	1.46	2.86
NH ₄ ⁺ "	4.21	0.077	0.72	0.077
iP x 10 ⁻³ "	0.55	1.47	5.5	0.60
Exchangeable cations				
K x 10 ⁻⁴ mM g ⁻¹ D. wt.	19.2	15.6	4.6	9.2
Na x 10 ⁻⁴ mM g ⁻¹ D. wt.	7.8	196.0	152.0	55.6
Ca x 10 ⁻⁴ mM g ⁻¹ D. wt.	336.5	429.0	151.0	349.1
Salinity (ppm)	121.6	177.9	108.2	116.5
pH	7.40	7.54	7.36	7.69
Physical analysis				
E.C. mmohs cm ⁻¹	0.19	0.28	0.17	0.18
W.H.C. %	61.0	33.4	18.5	28.7
Mean moisture content	27.3	1.61	0.26	1.25

Results and Discussion

Changes in growth parameters, total leaf conductivity and transpiration:

The available results for the growth of *Linum* plants on different Egyptian soils are shown in Table 2. In control plants, the root growth seemed to react comparatively to saline soil at the Northern Coastal site, there was a highly significant decrease in root elongation to about 50%, fresh and dry weight. In this connection Neumann, (1995a) stated that, salinity can rapidly inhibit the root growth and hence capacity for water uptake. However, a significant decrease at El-Kantara site and a highly significant increase in root length at Tushki site. For shoot, there was a significant decrease in both fresh, dry weights and plant height. This may be due to the decrease in water content and dry matter accumulation at three sites comparing with the control (Neumann, 1997).

Concerning R/S ratio, there was a highly significant decrease in plants grown at Northern Coastal site, highly significant increase at El-Kantara and a non-significant change at Tushki site. The increase in this ratio results from relatively greater

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Table 2: Effect of soil type on *growth criteria* of *Linum usitatissimum* (root and shoot length; root and shoot fresh and dry weights, R/S ratio; relative growth rate (RGR); stem diameter); total pigments; transpiration (T); total conductivity (C); leaf temperature (H) at relative humidity 34.6%; percentage of fiber, seed index and oil contents of seeds. * Non significant are indicated by LSD (P>0.05)

	Control	Northern Coast	El Kantra	Tushki	LSD at 5%	LSD at 1%
Root length (cm)	16.3	8.2	15.9	18.2	0.26	0.47
Shoot length (cm)	38.4	3.54	29.4	28.3	0.0004	0.0008
Root F. wt. (gm)	270.0	112.0	32.0	53.0	3.24	5.94
Root D. Wt (gm)	142.0	38.0	21.0	23.0	3.86	7.08
Shoot F. wt. (gm)	90.0	267.0	186.0	288.0	3.23	5.94
Shoot D. wt. (gm)	272.0	68.0	21.0	43.0	2.57	4.72
R/S	0.52	0.56*	1.0	0.53*	0.12	0.22
RGR g g ⁻¹ day ⁻¹ × 10 ⁻²	4.1	3.0	3.2	3.3	0.44	0.81
Stem diameter (mm)	3.08	2.33	2.51	2.22	0.026	0.047
Total Pigments (μg g ⁻¹ F.WT.)	2.990	0.180	0.165	0.380	0.03	0.06
T	8.0	7.4	10.9	9.5	0.26	0.48
C	320.0	276.0	359.0	341.0	7.39	13.56
H	29.0	29.7	35.4	30.0	0.45	0.85
% of fiber	33.84	20.10	21.60	14.20	0.24	0.45
Seeds index (gm)	16.36	10.49	10.33	10.29	0.015	0.027
mg oil/ 100 seeds	161.6	297.0	153.0	190.0	5.644	9.130

Table 3: Effect of soil type on ionic contents of *Linum usitatissimum* throughout vegetative (30-days old) and flowering (70-days old). Calculated as mmole g⁻¹ D. wt.

Soil type	K		Na		K/Na		Ca	
	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot
Vegetative								
Control	0.04	0.25	0.33	0.31	0.12	0.81	0.05	0.21
Northern Coast	0.015	0.088	0.16	0.15	0.093	0.59	0.036	0.058
El-Kantra	0.055	0.258*	0.66	0.24	0.083	1.08	0.077	0.095
Tushki	0.044*	0.163	0.91	0.85	0.05	0.191	0.059*	0.122
LSD at 0.05	0.012	0.012	0.0004	0.022	0.013	0.012	0.012	0.014
LSD at 0.01	0.021	0.021	0.0008	0.041	0.025	0.021	0.021	0.025
Flowering								
Control	0.23	0.11	0.48	0.21	0.48	0.54	0.087	0.12
Northern Coast	0.05	0.12	0.74	0.19	0.07	0.63	0.25	0.13
El-Kantra	0.04	0.21	0.35	0.20	0.11	1.05	0.08*	0.15
Tushki	0.07	0.31	0.52	0.39	0.13	0.33	0.06	0.18
LSD at 0.05	0.0004	0.0004	0.0004	0.0004	0.0004	0.0004	0.012	0.0076
LSD at 0.01	0.0008	0.0008	0.0008	0.0008	0.0008	0.0008	0.021	0.0139

* Non significant as indicated by LSD (P < 0.05).

Table 4 : Effect of soil type on the rate of transport of K(J_K), Na (J_{Na}) and Ca (J_{Ca}) in *Linum usitatissimum*.

	Transport of K J _K	Transport of Na J _{Na}	Transport of Ca J _{Ca}
Control	0.106	0.070	0.020
Northern Coast	0.010	0.150	0.050
El-Kantara	0.094	0.049	0.007
Tushki	0.030	0.102	0.019*
LSD at 5%	0.012	0.0044	0.0036
LSD at 1%	0.023	0.0082	0.0065

* Non significant as indicated by LSD (P < 0.05).

Table 5: Correlation coefficients between edaphic factors and dry weight of roots and shoots; percentage of fiber, seed index and oil contents of the yielded seeds of *Linum usitatissimum*.

	D. wt. root	D. wt. shoot	% of fiber	Seed index	Oil content
Organic carbon	0.652	0.570*	0.3220*	0.65	-0.14*
CaCO ₃	-0.243*	0.2090*	-0.287*	-0.342*	0.997
HCO ₃ ⁻	-0.535*	-0.1280*	-0.437*	-0.615	0.888
Cl ⁻	0.011*	0.4160*	0.084*	-0.083*	0.851
SO ₄ ⁻²	0.744	0.825	0.502*	0.709	0.168
NO ₃ ⁻	0.955	0.987	0.853	0.920	0.029*
NH ₄ ⁺	0.968	0.771	0.950	0.986	-0.483*
iP	-0.469*	-0.588	-0.115*	-0.424*	-0.313*
K ⁺	0.801	0.968	0.639	0.737	0.346*
Na ⁺	0.668	-0.370*	-0.446*	-0.717	0.631
Ca ⁺²	0.216*	0.541*	-0.056*	0.130*	0.752
Salinity	-0.068*	0.377*	-0.100*	-0.171*	0.969
pH	-0.395*	-0.252*	-0.687	-0.414*	0.392*
E.C.	0.069*	0.376*	0.099*	0.170*	0.964
W.H.C	0.968	0.939	0.814	0.945	-0.087*
Moisture	0.993	0.843	0.912	0.999	-0.368*

* Non significant as indicated by LSD (P < 0.05).

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Table 6: Comparative analysis of relative concentration, molecular weights and mobility rates of different types of protein bands of *Linum usitatissimum* seeds grown at different Egyptian soils.

Band No.	M. Wt. KDa	Mobility Rate R.M.	Band %			
			Control Lane (1)	Northern Coastal Lane (2)	El-Kantara Lane (3)	Tushki Lane (4)
1	157.16	0.12	-	-	0.835	-
2	150.33	0.14	-	1.79	-	-
	149.3	0.14	1.53	-	-	-
	148.29	0.14	-	-	1.14	-
	146.77	0.15	-	-	-	0.961
3	143.3	0.16	1.71	1.56	-	-
	140.4	0.16	-	-	-	0.669
	138.96	0.17	-	-	1.14	-
4	138.02	0.17	-	-	0.613	-
	137.08	0.17	-	-	-	0.875
5	126.28	0.20	2.03	-	-	-
	125.85	0.20	-	1.99	-	-
	125.42	0.2	-	-	2.07	-
	124.14	0.2	-	-	-	1.98
6	115.15	0.23	-	3.05	-	-
	114.76	0.23	3.28	-	-	-
	114.37	0.23	-	-	2.81	-
	113.59	0.23	-	-	-	1.47
7	109.03	0.24	-	-	-	0.476
8	97.068	0.28	-	-	-	3.37
	96.737	0.28	-	3.44	2.71	-
	96.407	0.29	3.72	-	-	-
9	86.42	0.32	4.56	4.71	7.94	4.91
10	78.804	0.35	-	-	-	1.0
	78.535	0.35	-	2.01	-	-
	78.0	0.36	2.29	-	-	-
	74.102	0.37	-	-	1.3	-
11	71.369	0.38	-	-	-	1.54
	66.2	0.41	-	-	2.1	-
12	60.822	0.42	-	3.21	-	2.22
	59.128	0.42	1.72	-	-	-
	53.815	0.43	-	-	1.21	-
13	37.476	0.52	-	-	-	20.0
	35.862	0.53	-	13.1	-	-
	35.379	0.54	-	-	19.8	-
	35.14	0.54	25.2	-	-	-
14	29.297	0.61	-	-	1.14	-
15	27.283	0.64	-	-	2.34	-
	26.95	0.64	-	1.89	-	-
	26.818	0.65	2.92	-	-	-
	26.361	0.65	-	-	-	3.51
16	24.852	0.68	6.07	-	-	-
	24.731	0.68	-	6.25	-	-
	24.309	0.69	-	-	9.98	-
	23.836	0.7	-	-	-	8.67
17	22.638	0.72	-	-	4.09	-
	22.362	0.73	5.28	4.32	-	-
18	20.847	0.75	-	-	-	2.6
	19.4	0.77	-	20.6	-	-
19	18.053	0.79	-	-	3.92	-
	17.416	0.79	19.9	-	-	-
	13.609	0.85	-	0.735	-	0.948
20	11.785	0.88	-	-	1.71	-
	11.31	0.89	0.548	-	-	-
	10.258	0.91	-	0.331	-	-
21	8.658	0.95	-	-	-	0.639
	8.057	0.96	-	-	1.72	-
	7.6535	0.97	0.961	-	-	-
	7.3831	0.98	-	-	-	-
Total number			15	16	19	17
Soil responsive proteins			-	13	17	16

decrease in shoot than in root growth under drought stress (Malik *et al.*, 1979). The increase in dry matter root/shoot ratio often implies the development of a larger ratio of root length density to leaf area, which translate into a better capacity for sustaining plant water status under a given

evapotranspirational demand (Malik *et al.*, 1979). Osmotic adjustment and turgor maintenance in growing region was also important in sustaining root growth at low water potential (Morgan, 1995). As compared to the control plants, there was a highly significant decrease in RGR of *Linum*,

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Table 7: Effect of different soil types on percentage of fatty acids in the oil of yielded seeds of *Linum usitatissimum*.

Fatty acid	Control	Northern Coastal	El-Kantara	Tushki
Hexanoic (6)	0.466	-	0.392	-
Heptanoic (7)	1.167	0.536	0.989	-
Nonanoic (9)	0.566	0.389	0.271	-
Decanoic (10)	0.567	0.269	0.133	-
Undecanoic (11)	5.776	1.503	3.269	-
Tridecanoic (13)	1.716	0.466	2.153	-
Pentadecanoic (15)	0.970	0.551	1.993	-
Palmitic (16)	12.640	7.178	9.919	16.480
Stearic (18:0)	28.468	9.968	23.678	13.780
Oleic (18:1)	12.492	7.179	9.872	13.427
Linoleic (18:2)	3.186	2.110	2.495	2.961
Linolenic (18:3)	3.858	3.250	2.128	8.76
Arachidonic (20:4)	4.874	6.324	4.245	11.402
Docosahexaenoic (22:6)	1.032	3.618	3.866	5.838
Unsaturated	25.442	22.481	22.606	42.388
Saturated	52.336	20.86	42.797	30.26
Unsat./Satur.	0.486	1.903	0.528	1.400

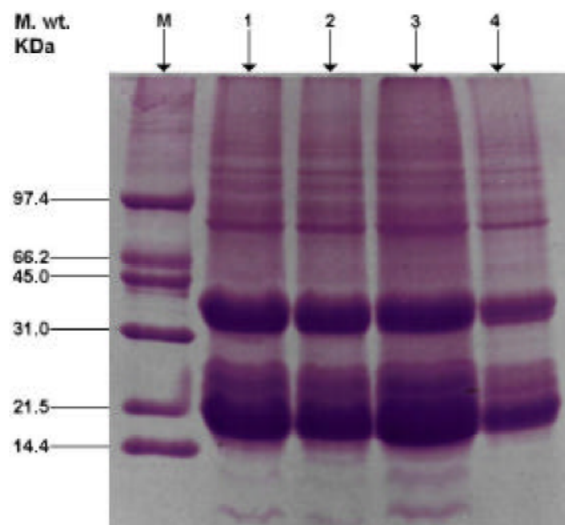


Plate 1: Electropherogram of soluble protein pattern by one-dimensional SDS-PAGE showing the changes of protein bands (marked by arrowheads), in response to different soil type. Each lane contains equal amounts of protein extracted from *Linum usitatissimum*. Protein bands in the gel were visualized by coomassie blue stain.

Lane M protein markers.
 Lane 1 Control seeds.
 Lane 2 Northern Coastal seeds.
 Lane 3 El-Kantara seeds. Lane 4 Tushki seeds.

grown at Northern Coast and El-Kantara and a significant decrease at Tushki site. The importance of the internal water balance in plant water relations is generally accepted because of the close relationship between this balance and turgidity, to the rate of physiological processes that control the quality and quantity of growth (Aldesuquy and Ibrahim, 2000). Measurements of electrical conductance of leaf surfaces have been used in the past to investigate the leaf wetness status (Burkhardt and Gerchau, 1994), the leaf water content (Kreeb, 1988) or the transport processes of ions through the cuticle (Tyree *et al.*, 1992). Bowling (1989) observed a close

relationship between leaf surface current and stomatal conductance. Thus, the data in Table 2 showed that leaf conductance and transpiration rate were reduced, whereas leaf temperature showed an increase at Northern Coastal site. This effect is mainly due to stomatal closure (Amon and Gupta, 1995). Reduced transpiration may therefore constrain the nutrient supply and plant growth in a very humid atmosphere, though active guttation (Pedersen, 1993) and coupled circulation in the phloem and xylem (Münch counter flow, Tanner & Beevers, 1990) may alleviate these restrictions. Meanwhile at El-Kantara and Tushki sites, there was an increase in leaf conductance and transpiration rate comparing with that of the control. Monteith (1995) found that increasing the water potential gradient between the guard cells and other epidermal cells or lowering leaf water potential, could directly decrease the turgor pressure of guard cells and other epidermal cells or affect the hormonal distribution.

Changes in pigment content: As can be seen from Table 2, a marked significant decrease in total pigments in *Linum* at three sites of Egyptian soils comparing with the control. This may be due to reduction in mass of leaves (Van Deen Boogaard *et al.*, 1996) or due to the raising of elements in leaves to the toxic levels (Munns, 1993) or due to closure of stomata (Sanchez-Rodriguez *et al.*, 1999) and the net result is the inhibition in the rate of photosynthesis under stress conditions either by salinity or by drought. In this connection, Valladares & Pearcy (1997) stated that, the interaction between high light which induce high leaf temperature (Flexas *et al.*, 1999) as shown in Table 2 and other environmental stresses such as drought (at El-Kantara & Tushki) and salinity (Northern Coast) lead to photoinhibitory processes.

Changes in ionic contents: It is clear from Table 3, that during the growth of *Linum* in soil collected from Northern Coastal site, concentration of K, Na, Ca and K/Na ratio decreased significantly at vegetative stage in both shoots and roots. These results are in good conformity with those of Flowers and Yeo (1995) and in K of root; Na of shoot at flowering stage, while there is a highly significant increase in K in shoot and Na; Ca ions in root and a non-significant change in Ca ion in the shoot at flowering stage. There was a significant decrease in root and shoot growth (length, Fresh and dry weights) as shown in Table 2. In this connection, Van Deen Boogaard *et al.* (1996), stated that, salt induced inhibition of leaf growth can also slow down the canopy closure during seedling establishment as shown in Table 2. This may decrease the seedling water use efficiency, since more soil water is lost via direct evaporation from the soil surface than from the leaves. Finally, salinity can rapidly inhibit the root growth and hence capacity for uptake of water and essential mineral nutrients from the soil (Neumann, 1995a). From another point of view, the rate of transport of K (J_K) (Table 4) was significantly decreased, while the transport of Na and Ca (J_{Na} and J_{Ca}) were significantly increased. Garica *et al.* (1997) stated that, other factors determining the expression of salt damage include nutrient acquisition and efficiency in the use of water. Furthermore, Marschner (1995) stated that, Na competition at transport sites for K entry into the symplast may result in K deficiency and cytoplasmic Na competes for K binding sites and hence inhibits the metabolic processes that crucially depend on K. Therefore, many authors pointed out that one of the key elements in salinity tolerance is the capacity to maintain a high cytosolic K/N ratio, as clear from shoot at flowering stage (Maathuis & Amtmann, 1999). However, the plants grown at El-Kantara and Tushki sites,

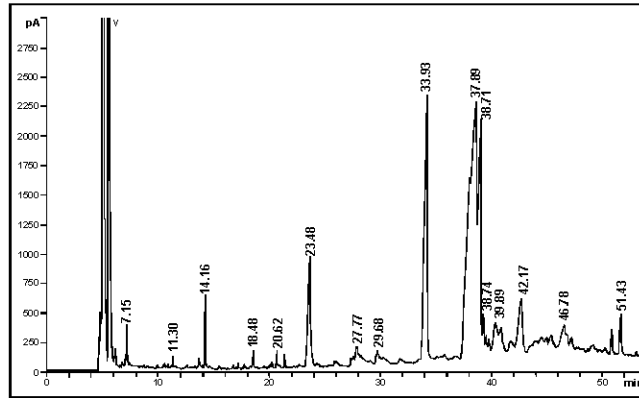


Fig. 1: GLC Chromatogram showing the fatty acids composition of *Linum usitatissimum* seed oil in control plants.

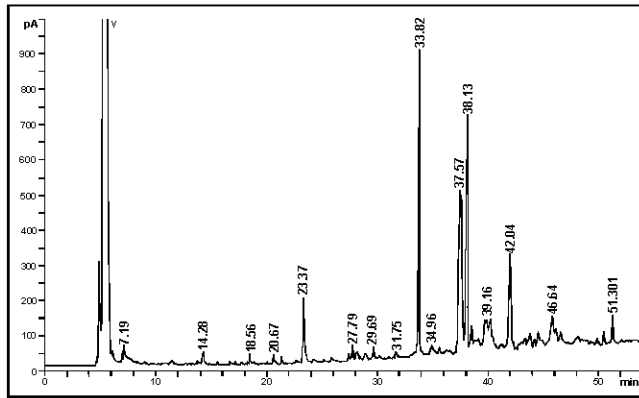


Fig. 2: GLC Chromatogram showing the fatty acids composition of *Linum usitatissimum* seed oil at Northern Coastal site.

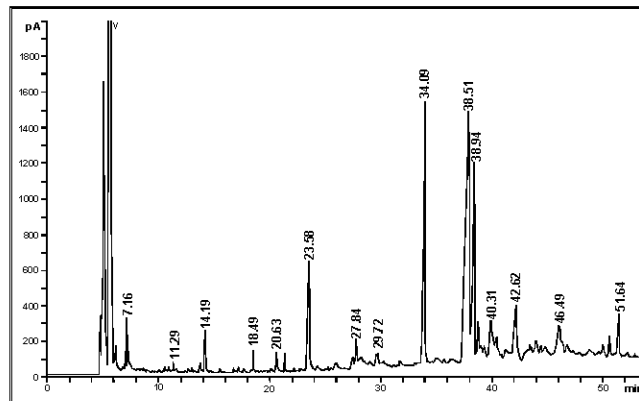


Fig. 3: GLC Chromatogram showing the fatty acids composition of *Linum usitatissimum* seed oil at El-Kantara site.

showed variable significant changes in K, Na & Ca ions in both shoots & roots at vegetative and flowering stages, and at El-Kantara there was a significant decrease and increase in K/Na ratio in root and shoot respectively, at two stages and a significant decrease in K/Na ratio in shoots & roots at two stages at Tushki site (Table 3). Consequently, the rate of K, Na & Ca ($J_j K$; $J_j Na$ & $J_j Ca$), transport was significantly decreased (Table 4), at El-Kantara site. Meanwhile, a

significant decrease and increase in K and Na transport respectively at Tushki was recorded, and a non-significant change was obtained in Ca transport. In this respect Munns, 1993 stated that, continuous salt accumulation combined with limited production of new leaf volume, that could lead to earlier build up of excess (toxic) levels of salt. This might further accelerate the onset of leaf senescence and necrosis

(Yadav *et al.*, 1996).

Changes in yield and fiber yield: It is clear from Table 2, that, the yield components represented by seed index, were significantly decreased at three sites. In connection to these results, there are many reports on the soil fertility levels and linseed yield (Aegehehu and Honermeier, 1997; Singh & Verma, 1997). The highest seed index was obtained for plants growing at North Coastal site, while the lowest one was obtained at Tushki site and medium one was obtained at El-Kantara site. Case *et al.* (1999) stated that Linseed yield varied in response to weather and soil type.

There was a highly significant decrease in percentage of fiber at all sites comparing with the control. However, in comparison between three sites, the highest fiber yield was obtained for plants at El-Kantara site, while the lowest one was obtained for plants at Tushki site, and the medium one was obtained at Northern Coastal site. These results are in conformity with the results of the stem diameter as shown in Table 2 and obtained by El-Haak *et al.* (1999).

Statistical analysis was carried out to evaluate the relationships between soil factors and dry weight of shoot and root; percentage of fiber, seed index and oil content seeds of *Linum* using multiple correlation analysis (Zar, 1984). The correlation coefficients (*r*) of the significant relationship ($P < 0.05$) only, are listed (Table 5).

There was a positive correlation between organic carbon and each of dry weight of root and seed index ($r = 0.65$), sulphate concentration and dry weight of root ($r = 0.74$), dry weight of shoot ($r = 0.83$) and seed index ($r = 0.71$); nitrate concentration and each of dry weight of root ($r = 0.96$) dry weight of shoot ($r = 0.99$) percentage of fiber ($r = 0.85$) seed index ($r = 0.92$); ammonium concentration and dry weight of root ($r = 0.97$), dry weight of shoot ($r = 0.77$) percentage of fiber ($r = 0.95$), seed index ($r = 0.99$); K concentration and dry weight of root ($r = 0.80$), dry weight of shoot ($r = 0.97$), percentage of fiber ($r = 0.64$), seed index ($r = 0.74$); water holding capacity and dry weight of root ($r = 0.97$), dry weight of shoot ($r = 0.94$), percentage of fiber ($r = 0.81$), seed index ($r = 0.95$); moisture content and dry weight of root ($r = 0.99$), dry weight of shoot ($r = 0.84$), percentage of fiber ($r = 0.91$), seed index ($r = 0.99$). Also a positive correlation was recorded between oil content and each of CaCO_3 ($r = 0.997$); HCO_3^- ($r = 0.888$); Cl^- ($r = 0.851$); salinity ($r = 0.969$) and E.C. ($r = 0.964$).

A negative correlation was found between concentration of inorganic phosphorus and dry weight of shoot ($r = -0.59$); Na concentration and seed index ($r = -0.72$); pH percentage of fiber ($r = -0.69$). These results are in accord with those of Casa *et al.* (1999) and Riffkin *et al.* (1999), who found that the linseed yields, vegetative growth varied greatly in response to fertility levels and soil types.

Soil responsive proteins: The changes in protein electrophoretic pattern in the yielded seeds of *Linum usitatissimum* growing at different sites in Egypt are shown in plate 1, analyzed and recorded in Table 6.

In the seeds of control *Linum* (Table 6), the separation of 15 protein bands was apparent. Their molecular weight ranged between 157.16 and 7.383 KDa. Growth of *Linum* in different sites of Egyptian soil, Northern Coastal region, El-Kantara and Tushki increased the protein bands in seeds to 16, 19 and 17. In addition, some proteins disappeared in seeds of *Linum* growing at three sites of Egyptian soil, while a new set of proteins was de novo synthesized (Table 6). These results indicate that, the growth of *Linum* in El-Kantra induced a high synthesizing rate of new protein, while the growth in the coastal region is least one in de novo synthesis rate. In this respect, Robinson *et al.* (1990) suggested that the disappearance of polypeptides during stress compensates the

increased synthesis of others. Moreover, in response to salt stress, despite reduction in protein levels (Singla and Grover, 1994). Cells preferentially synthesize few specific proteins, which are termed as stress proteins (Pareek *et al.*, 1995). One of the most important mechanisms involved in cell protection against stress (either drought and salt) is the induction of de novo synthesis of a specific set of proteins (Kermode, 1997). In present investigation, the growth of *Linum* at Northern Coastal region induced de novo synthesis of 13 proteins in the seeds. The growth at the El-Kantara site induced the de novo synthesis of 18 protein sets (M. wts; 157.16; 148.29; 138.96; 138.02; 125.42; 114.37; 96.73; 74.10; 66.2; 53.81; 35.37; 29.29; 27.28; 24.31; 22.63; 18.05; 11.78 and 8.05 KDa). Moreover, the seeds from the plants growing in new valley (Tushki) were characterized by the presence of a new 16 proteins (M wts; 146.77; 140.4; 137.08; 124.14; 113.59; 109.03; 97.06; 78.81; 71.36; 60.82; 37.47; 26.36; 23.83; 20.84; 13.61 and 8.65 KDa). In addition, the concentration of polypeptide band of molecular weight 86.42, was a common one in seeds of control *Linum* seeds grown in, coastal site, El-Kantara as well as the Tushki site. The variation of this polypeptide was found to depend mainly on the type of soil. El-Kantara site induced a highest concentration of this polypeptide if compared to other types of soil. In this respect, HSP 90 (a group of HSP_s) with molecular weights in the range of (80 to 90 KDa) accumulates in response to drought and salt stress (Krishna *et al.*, 1995). Such proteins are referred to as stress associated proteins (SAP_s).

The appearance of de novo synthesis protein of M wts; 26.95 at Northern Coastal, 26.36 and 20.85 at Tushki sites indicate that, these salt responsive proteins might have an osmoprotection function and protect the cellular structure of *Linum*, resulting in increasing the tolerance.

The new proteins (37.47 KDa) which were detected in seeds of *Linum* grown in Tushki and 35.86 KDa at Coastal and 35.38 KDa at El-Kantara are suggested to be dehydrin. The dehydrins may have molecular weights of 31.37 and 40 KDa (Han *et al.*, 1997). They have a protective role in survival under water loss due to their function as ion trap in dehydrating cells; sequestering ions as they become concentrated (Close, 1996).

The protein (23.83 KDa) which was detected in seeds of Tushki appeared to be osmotin-like (Yen *et al.*, 1994) and germin-like proteins (Michalowski and Bohnart, 1992). Accumulation of osmotin has been correlated to increase the salt tolerance in tobacco cells (La Rosa *et al.*, 1989). Osmotin appeared to provide osmotic adjustment to the cell by facilitating the accumulation of solutes and/or providing metabolic alterations in the cells which may be helpful in osmotic adjustment (Singh *et al.*, 1987). Furthermore, proteins with M. wt of 13.61 KDs were apparent in seeds of Northern Coastal and Tushki groups. Similar results were obtained by Mundy and Chua (1988), who isolated a gene induced in rice plants in response to water stress.

The protein of molecular weight 8.65 and 8.06 KDa, detected in Tushki and El-Kantara sites were appeared to be Ubiquitin. It is a small heat stress protein consisting of 76 amino acid; 8.5 KDs (Kruse and Kloppstech, 1992). Ubiquitin appeared to protect protein from degradation by protease by tagging the protein (Hershko, 1988). Therefore, constitutive synthesis of specific polypeptides of M wt 60.82 KDs, 27.3 KDs & 24.3 KDs at El-Kantara and 60.8 KDs; 37.5 KDs & 23.8 KDs at Tushki sites were appeared to be responsible for higher thermotolerance. These results are in accordance with those of Kuznetsov & Shevyakova (1997), who stated that, the phosphorylation of some polypeptides (23, 24, 27, 31, 32 & 47 KDa) in salt-tolerant cells at elevated temperatures is accompanied by enhanced thermotolerance.

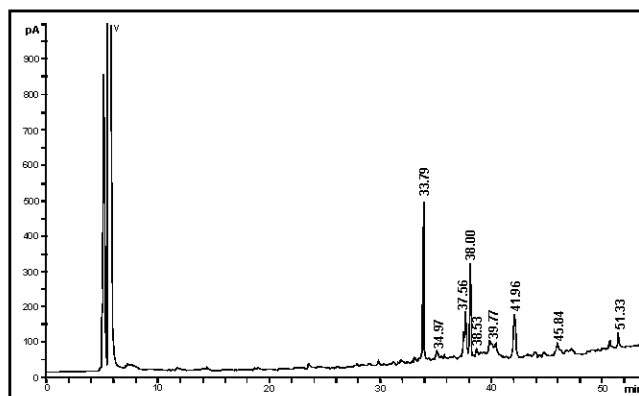


Fig. 4: GLC Chromatogram showing the fatty acids composition of *Linum usitatissimum* seed oil at Tushki site.

Oil content and fatty acid composition of yielded seeds: In *Linum*, the oil content was found to increase significantly at Northern Coastal and Tushki sites, however, a significant decrease was recorded at El-Kantara site as shown in Table 2. With respect to fatty acid composition of oil, the results in Table 7 and Figs. 1, 2, 3 and 4 show that, the predominant fatty acids in the oil of control *Linum usitatissimum* seeds were arranged as follows on the basis of their percentage : stearic (18:0), palmitic (16), oleic (18:1), undecanoic (11), arachidonic (20:4), linolenic (18:3), linoleic (18:2), and docosahexanoic (22:6). In this observation Wanasundara *et al.* (1999) stated that linolenic (18:3), linoleic (18:2) and oleic (18:1) acid were the predominant fatty acids of all the lipid fractions of flaxseed. In oil of seeds at Northern Coastal site, stearic (18:0) palmitic (16), oleic (18:1), undecanoic (11), linolenic (18:3) and linoleic (18:2) acids showed, in general, decreases below the control, whereas a rise in the level of arachidonic (20:4) and docosahexanoic (22:6) acids, so, there was a decrease in saturated and unsaturated fatty acids, but, a high value for unsaturated/saturated ratio was recorded. For El-Kantara site, in general there was a decrease in all fatty acids except docosahexanoic (22:6) acid. A decrease in saturated and unsaturated fatty acids were recorded. Unsaturated/saturated ratio showed a slight increase. Saturated fatty acids [hexanoic (6) heptanoic (7), nonanoic (9), decanoic (10) undecanoic (11), tridecanoic (13), and pentadecanoic (15)] revealed that, there was a gradual disappearance in the oil of flaxseed at Tushki site, however, there was an increase in unsaturated fatty acids [oleic (18:1), linolenic (18:2), arachidonic (20:4) and docosahexanoic (22:6)] and saturated fatty acid palmitic (16). Whereas a decrease was recorded in stearic (18:0) and linoleic (18:2) acid. So, a high increase in total unsaturated fatty acids and a decrease in total saturated fatty acids was recorded that lead to the calculated increase in the ratio of unsaturated/saturated. In this respect, Agegnehu and Honermeier (1997) stated that, the compositional changes in fatty acids of oil and the proportion of saturated and unsaturated varied according to soil fertility. Flaxseed has been used as an edible grain in different parts of the world. However, use of flaxseed oil has been limited due to its high content of polyunsaturated fatty acids (Wanasundara and Shahidi, 1998). Flaxseed oil containing low levels of linolenic acid are available for edible oil extraction. So, in comparing the composition of fatty acids in oil of flaxseed at three sites, it was apparent that, we can use the oil extracted from seeds at El-Kantara and at Northern Coastal site. These results are in accord with those obtained by Saeidi and Rowland, (1999), who found that flax is an edible - oil

crop that must have less than 5% linolenic acid in its seed oil. Meanwhile, the oil of seeds at Tushki can't be used as an edible oil. For the final conclusion, it seemed that, *Linum* can be cultivated at Northern Coastal site for high seed index and oil content at El-Kantara for high percentage of fiber and for a suitable edible oil with low percentage of linolenic acid, but at Tushki, *Linum* had a low seed index, fiber percentage, however it had a medium oil content, but it was not suitable as an edible oil with a high percentage of linolenic acid.

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