

Seed Composition Response of Sesame (*Sesamum orientale*) to Applied Water Deficit at Certain Growth Stages.

Dina Z. Abdel-Kader and Amal A. H. Saleh

Botany Dept., Fac. of Sci., Suez Canal Univ., Ismailia, Egypt.

Sesame (*Sesamum orientale*) plants were subjected to drought stress at different growth stages. The control plants were irrigated at 4 days intervals, while the drought stress treatments were irrigated at 8 days intervals for three weeks during the vegetative stage (T1), flowering stage (T2) and seed growth stage (T3). Yield at maturity was measured in the four treatments. Plant height and number of fruits were measured on 20 randomly sampled plants for each treatment. Seeds were collected and prepared for determination of total carbohydrates, protein and lipid contents as well as fatty acid composition. Drought stress during the three stages induced significant decreases in plant height, pod numbers, weight of seeds, total proteins and oil content. Total carbohydrates were significantly increased while fatty acid composition was significantly affected under drought stress conditions. T1 and T3 were the most sensitive stages to drought stress while the T2 stage was the least sensitive. The data from this study indicated that water stress during any growth stage is detrimental to sesame plants and early and late season droughts are more damaging than midseason drought. The effect of drought stress on oil content and fatty acid composition has important implications for the quality of sesame oil.

Key words: Carbohydrates, drought stress, fatty acids, growth stages, irrigation, oil, protein, Sesame, yield components.

Droughty conditions are implicated in causing low seed yield, poor grade and decreased germination. To optimize irrigation management in terms of yield, it is necessary to establish which stages of growth are the most susceptible to water shortage. Moderate water deficits during early vegetative development do not cause significant yield loss and may even produce a slight yield advantage (Pallas *et al.*, 1979; Sivakumar and Sarma, 1986).

Stirling *et al.* (1989) reported that late-season drought is more detrimental to final pod yield in groundnut than early season drought. Drought stress decreased yield, seed weight, oil content and germination percentage of soybean plant (Dornbos *et al.*, 1989). They

also found that the linolenic and linoleic acid contents were reduced whereas the oleic acid content was increased by the drought stress.

Mozaffari *et al.* (1996) measured responses to water stress of eight sunflower hybrids. They found that water stress caused large reductions in seed yield, biomass and period of vegetative growth. Christiansen *et al.* (1998) examined the effect of drought stress at various growth stages on yield components in *Lupinus albus* and *Lupinus angustifolius*. They concluded that the yield component most sensitive to water stress was pods/plant, while seeds/pod was less affected. Granier and Tardieu (1999) found that reductions in final leaf area and cell number in a given zone of the leaf varied mainly with the timing of water deficit, with maximum effect for the earliest deficits.

The assimilation of nitrogen in leaves of higher plants requires both energy and carbon skeletons. Triose phosphate produced in the leaves as a result of photosynthetic carbon assimilation can be used for the synthesis of either carbohydrates or ketoacids (e.g. 2-Oxoglutarate) via the anapleurotic pathway. 2-Oxoglutarate produced in the cytosol is imported into the chloroplasts, where it may serve as the acceptor for NH_4^+ during amino acid synthesis. To meet the needs of growth and development for both carbohydrates and amino acids, carbon partitioning is coordinated by a sophisticated regulatory system. There are many points of reciprocal control between the pathways of carbon and nitrogen assimilation (Champigny and Foyer, 1992; Huber *et al.*, 1992, 1994-a, 1994-b, Foyer *et al.*, 1996). However effective regulation eliminates the competition observed in algae for available energy and carbon resources (Champigny and Foyer, 1992; Huppe and Turpin, 1994; Foyer *et al.*, 1996).

Protein, oil contents and fatty acid composition could be altered by environmental stress. Dornbos *et al.* (1989) imposed drought stress at seed fill stage of soybean plants. They concluded that the effect of drought and high temperature stress on phospholipid class and fatty acid composition has important implications for the quality of soybean seed oil and lecithin.

The present study was conducted to investigate which stages of sesame plant growth are the most susceptible to water deficit.

Materials and Methods

The experiment was conducted in the Botanical Garden of Faculty of Science, Suez Canal University, Ismailia, Egypt, 1999.

Pure seed of sesame (*Sesamum orientale* var. Giza 32) was obtained from the Egyptian Ministry of Agriculture.

The field was prepared as broad beds (80 cm wide) with furrows (30 cm wide) on either side of the bed. Sesame seeds were planted on 29 June 1999. The field was irrigated to field capacity after sowing.

The experiment was set up as a randomized complete block design with four treatments, each with three replications. The four treatments (Control, T1, T2 and T3) were the growth phases of the crop during which irrigation was varied. See Table (1) for the irrigation schedule.

Control: Continuous irrigation at 4 days interval from emergence to maturity, (Uniform irrigation).

T1: Uniform irrigation for 21 days after sowing (DAS), then irrigation at 8 days intervals for 3 weeks (vegetative phase) till the start of flowering. Uniform irrigation thereafter.

T2: Uniform irrigation for 41 DAS, then irrigation at 8 days intervals for 3 weeks (flowering phase) till the start of seed formation. Uniform irrigation thereafter.

T3: Uniform irrigation for 63 DAS, then irrigation at 8 days intervals for 3 weeks (seed growth phase). Uniform irrigation thereafter.

TABLE 1. Schedule of irrigation treatments applied during the experiment.

Date	DAS	Control	T1	T2	T3
29 June-20 July 1999	0-21	UI	UI	UI	UI
24 July	25	UI	-	UI	UI
28 July	29	UI	I	UI	UI
1 August	33	UI	-	UI	UI
5 August	37	UI	I	UI	UI
9 August	41	UI	-	UI	UI
13 August	45	UI	UI	-	UI
17 August	49	UI	UI	I	UI
21 August	54	UI	UI	-	UI
25 August	59	UI	UI	I	UI
29 August	63	UI	UI	-	UI
2 Sept.	67	UI	UI	UI	-
6 Sept.	71	UI	UI	UI	I
10 Sept.	75	UI	UI	UI	-
14 Sept.	79	UI	UI	UI	I
18 Sept.	83	UI	UI	UI	-
22 Sept.	87	UI	UI	UI	UI
26 Sept.	91	UI	UI	UI	UI

DAS: days after sowing. UI: uniform irrigation. I: irrigation

Final yield measurements (91 DAS): Yield at maturity was measured from the four treatments. Plant height and the number of fruits were measured on 20 randomly sampled plants for each treatment. Seeds were collected and prepared for measurements of total carbohydrates, protein and lipid contents, as well as, for fatty acid composition.

Total carbohydrates:

Total carbohydrates were extracted in test tubes containing dry seeds plus 5 ml of 2.5 N HCl. The tubes were placed in a boiling water bath for 3 hours. The samples were analyzed by the method of Hedge and Hofreiter (1962) using glucose as standard.

Total protein:

Total protein was extracted from the seeds with 1M NaOH. The extracts were centrifuged and an aliquot of the supernatant was analyzed for protein by the method of Lowry *et al.* (1951) using bovine serum albumin as standard.

Lipid content:

Lipid content was measured using the method of Folch *et al.* (1957) with some modifications. The seeds were extracted with chloroform: methanol (4:1). The extracts were bulked, filtered and reduced to dryness under vacuum at 35°C. The resultant material was reported as lipid content.

Investigation of fatty acid composition:

Fatty acid analysis of sesame lipid was carried out by saponification with 10% KOH in methanol (Karawya, 1988) followed by methylation according to Munshi, (1987). The resulting fatty acid methyl esters were injected into a gas liquid chromatograph Philips pu 4500, adapted with a computing integrator Philips pu 4815 and a 10% DEGS column. The following conditions were used: flow rate nitrogen 30 ml/min., hydrogen 30 ml/min., and air 300 ml/min., injection temperature 240°C, detector temperature 280°C, programmed 70°C-190°C/min.

Statistical analysis: Significant differences were identified with student's t-test (Underwood, 1997).

Results

Yield:

Table (2) summarizes the plant height, number of pods /plant, and weight of seeds/20 plant.

The data show that uniform irrigation led to significantly greater plant height, pod numbers and weight of seeds in comparison to the other treatments. The data showed that sesame was the most sensitive to drought during the T1 and T3 periods. The T2 period (flower initiation to beginning seed stage) was less sensitive to drought.

Total carbohydrates:

Total carbohydrate content of the seeds increased significantly by drought imposed at the T2 period while it decreased highly significant by drought at the T3 period (Table 3).

Total proteins:

Drought exposure at the T2 period had no significant effect on protein contents of sesame seeds while drought at the T1 and T3 periods induced a highly significant decrease in total proteins (Table 3).

Oil content:

The three drought treatments significantly decreased the oil content of sesame seeds however T2 drought had the smaller effect (Table 3).

Fatty acid composition:

The changes in fatty acid composition of seeds of sesame plants grown in the presence and absence of drought are listed in Table (4). It was found that drought during the T1 stage induced significant increases in palmitic and oleic acid and a highly significant increase in erucate acid. However, high significant decrease in linoleic acid and non-significant effect on linolenic acid percentage were observed. Drought during T2 stage exhibited highly significant increases in oleic and erucate acids content and highly significant decrease in linoleic acid content. The data also show that drought during T3 stage induced highly significant increase in eurcate acid and highly significant decrease in palmitic, oleic, linoleic and linolenic acids.

Discussion

Yield:

The data in Table (2) show that sesame plants are most sensitive during the T1 stage. This corresponds to the vegetative stage. These results are in agreement with those obtained by Stansell and Pallas (1985).

TABLE 2. The influence of water deficit at several growth stages on sesame plant height, and components of yield at final harvest.

Treatment	Height (cm)	No. of pods/plant	Wt. of seeds/20 plants, (g)
Control	100.2±4.4	29.6±0.9	108.1±8.7
T1	72.5±2.3*	16.4±0.5**	37.5±1.4**
T2	84.4±2.6	26.8±0.8	63.2±4.3*
T3	70.2±3.2*	15.2±0.7**	22.0±1.5**

-The data represent the mean ±SE of 20 plants.

*Significantly different from control (Student's unpaired t-test, $P<0.05$).

**Highly significantly different from control (Student's unpaired t-test, $P<0.01$).

In the present study water deficit during the vegetative stage caused the plants suffer severe vegetative damage, including some plant mortality. Water was insufficient to sustain vegetative growth and fruits development, resulting in severe yield and quality depression.

TABLE 3. The influence of water deficit at several growth stages of sesame plant on total carbohydrates, total protein and oil content of seeds.

Treatment	Total carbohydrates (mg/100g dry wt.)	Total protein (mg/100g dry wt.)	% of oil content
Control	4000±130	563±6	36.0±0.6
T1	4325±30	275±9**	27.2±0.6*
T2	5525±14*	575±9	32.4±0.5*
T3	2500±58**	601±3*	23.6±0.4*

-The data represent the mean ±SE of three replicates.

*Significantly different from control (Student's unpaired t-test, $P<0.05$).

**Highly significantly different from control (Student's unpaired t-test, $P<0.01$).

The present data show that the most damaging drought effect were observed in the T3 period, which corresponds to the beginning of the seed formation to the beginning the maturation stages. Sesame at 61 DAS is fully leafed and approaching the period of maximum evapotranspiration demand. As a result, initiation of drought at this

time resulted in a rapid depletion of soil water accompanied by severe wilting. These data are in accordance with those obtained by Stansell and Pallas (1985).

Moreover, Nageswara *et al.* (1985) and Pallas *et al.* (1979) found that water deficit during the pod-filling phase are more damaging to yield than drought during the pod addition phase. The decrease of peanut productivity under water stress could be ascribed to the decrease in auxin, gibberellins, cytokinins and increases in growth inhibitors (El-Meleigy *et al.*, 1999).

Carbohydrate and protein contents:

The present data show that drought periods during early and late stages of sesame plant growth caused the greatest changes in carbohydrate and protein contents of sesame seeds. These changes could result from prolonged periods of drought that decrease the availability of water for transport-associated processes and lead to changes in the concentrations of many metabolites. This may be followed by disturbances in amino acid and carbohydrate metabolism. Yancey *et al.* (1982) and Girousse *et al.* (1996) found an increase in the synthesis of compatible solutes such as certain amino acids (*e.g.* Proline), sugars and sugar-alcohols, and glycine betaine. Drought induces complex changes in carbon and nitrogen metabolism resulting from water deficits and from modifications in the availability of nutrients (Talouizite and Champigny, 1988; Larsson *et al.*, 1991; Beyrouthy *et al.*, 1994). Water deficit favored starch breakdown in C₃ species (Fox and Geiger, 1985) but caused accumulation of starch in maize (Foyer *et al.*, 1998). There was also a substantial increase in free glucose in droughted maize leaves, which may indicate that considerable starch turnover is favored under conditions of water deficit.

Carranca *et al.* (1999) found that legumes derived more than 60% of their N from the atmosphere under favorable soil conditions, but the proportion was reduced when the availability of soil moisture was constrained.

Acclimatization to drought requires responses that allow essential reactions of primary metabolism to continue and enable the plant to tolerate water deficits. In the complex interplay of natural conditions, water deficit is unlikely to occur alone, since water deficit affects the acquisition of essential nutrients such as nitrogen and phosphorus (Talouizite and Champigny, 1988; Larsson *et al.*, 1989 &

1991; Beyrouthy *et al.*, 1994). Indeed, drought-induced nitrogen deficiency was found to limit recovery of photosynthesis in prairie grasses once water was restored (Heckathorn *et al.*, 1997).

Carbohydrate-mediated repression of transcription of photosynthetic genes, such as those coding for the large and small subunits of Rubisco, is a possible mechanism of control when carbohydrate accumulates in leaves (Krapp *et al.*, 1993; Sheen, 1994; Graham, 1996; Koch, 1996). This may occur in water-stressed maize leaves following several days of water stress and provides at least a partial explanation for the reduction in photosynthetic capacity. Depletion of essential nutrients, particularly NO_3^- can also cause changes in gene expression and enzyme activity in stress situations (Scheible *et al.*, 1997-a, 1997-b). They are nevertheless indicative of the overall shift in carbon and nitrogen metabolism that occurs as a result of drought.

Oil content and fatty acid composition :

The present study shows that drought stress significantly decreased the oil content of sesame seeds and had a highly significant effect on fatty acid composition of the seeds. These results are in agreement with those obtained by Dornbos *et al.* (1989), who found that fatty acid composition of soybean plants was altered by drought and high temperature. They found that greater proportion of 16:0 and 18:0, and lower proportion of 18:2 and 18:3, were present in seeds exposed to high temperature and severe drought. More linolenic acid and less palmitic acid were present in the phosphatidylinositol fraction.

TABLE 4. The influence of water deficit at several growth stages of sesame plant on fatty acids composition of seeds.

Treatment	% of Fatty Acids				
	Palmitic C16:0	Oleic C18:1	Linoleic C18:2	Linolenic C18:3	Erucic C24
Control	17.1 \pm 0.0	10.1 \pm 0.4	40.4 \pm 0.7	5.6 \pm 0.1	12.4 \pm 0.2
T1	20.7 \pm 0.1*	14.1 \pm 0.3*	25.1 \pm 0.6**	5.8 \pm 0.3	27.4 \pm 0.9**
T2	17.1 \pm 0.1	17.7 \pm 0.5**	26.6 \pm 0.9**	5.9 \pm 0.5	22.8 \pm 0.7**
T3	12.7 \pm 0.1**	5.5 \pm 0.2**	23.0 \pm 0.9**	2.2 \pm 0.1*	52.1 \pm 0.7**

- The data represent the mean \pm SE of three replicates.

*Significantly different from control (Student's unpaired t-test, $P < 0.05$).

**Highly significantly different from control (Student's unpaired t-test, $P < 0.01$).

The effect of drought on oil content and fatty acid composition of sesame seeds may be caused by a reduced metabolic activity due to a decline in pod water content. This possibility was previously proposed by Musingo *et al.* (1989).

Conclusion

This study indicates that water stress during any growth stage is detrimental to sesame plants and early and late season droughts are more damaging than midseason drought. The effect of drought stress on oil content and fatty acid composition has important implication for the quality of sesame oil. Drought increased the saturated fatty acids and decreased the unsaturated fatty acids and this decreases the quality of sesame oil.

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تأثير مكونات بذرة السمسم (*Sesamum orientale*) بنقص الماء عند مراحل نمو معينة.

دينا زين العابدين و أمال أحمد حسن

قسم النبات - كلية العلوم - جامعة قناة السويس بالإسماعيلية - مصر .

عرضت نباتات السمسم للجفاف عند فترات نمو معينة متمثلة في المعاملات الآتية : T3; T2; T1 . ويمثل الكنترول T3. ويري الدائم للنباتات كل أربعة أيام حتى الحصاد ، أما معاملات الجفاف فكانت رى للنباتات كل ثمانية أيام لمدة ثلاثة أسابيع خلال فترة النمو الخضري (T1) وخلال فترة الإزهار (T2) وخلال فترة نمو البذرة (T3) .

تم تقدير المحصول عند الحصاد لكل المعاملات الى جانب قياس أطوال النباتات وعدد الثمار . كما تم قياس الكربوهيدرات الكلية ، البروتين ، الدهون وقدر محتوى الأحماض الدهنية في البذور .

أظهرت الدراسة أن تعرض نبات السمسم للجفاف أثناء المعاملات الأولى والثالثة قد تسبب في نقص معنوي في أطوال النباتات ، عدد الثمار . وأن وزن البذور قد تأثر بالمعاملات الثلاثة ، وقد زادت نسبة الكربوهيدرات الكلية زيادة معنوية أثناء المعاملة الثانية ولكنها نقصت نقصاً معنوياً أثناء المعاملة الثالثة. أما البروتين الكلى فقد نقص معنوياً أثناء المعاملة الأولى بينما زاد معنوياً أثناء المرحلة الثالثة. وبالنسبة للمحتوى الزيتى فقد نقص معنوياً أثناء المعاملات الثلاثة وكانت المعاملة الثالثة هي الأكثر تأثيراً .

وقد وجد أن أكثر فترات النمو تأثراً بالجفاف هي T1 و T2 و T3 . وقد أوضحت هذه الدراسة أن تعرض نبات السمسم للجفاف في أى مرحلة من مراحل النمو يعرضه لنقص معنوي في المحصول الى جانب أن الجفاف يؤثر على نوعية زيت السمسم الناتج حيث يزيد الأحماض الدهنية المشبعة ويقلل الأحماض الدهنية الغير مشبعة مما يقلل من نوعية الزيت .