

Effect Of Water Stress During Seed Formation On Seed Quality Of Irrigated Soybean

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Abstract:

These experiments were carried out at Agricultural experiments and Research Station, Faculty of Agric., Cairo University, Giza, Egypt during 2018 and 2019 summer seasons to study the effect of water stress during seed formation on seed quality of soybean. Four treatments were used: 1-non-stress, mild stress, moderate stress and high stress. The present study indicated that exposing irrigated soybean crop to water stress during seed formation up to harvest maturity reduced the normal seeds of the total yield by 20% and increased the defective seeds, green seeds and seeds with cracked seed coat percentage. This was in parallel with reduction in germination percentage, normal seedling, seedling length, seedling dry weight and seedling vigor index. Also, when water stress started at R5 stage (seed formation) or earlier at R3 stage (pod formation) caused significant reduction in oil and protein content, but carbohydrate and fiber contents were increased. Also saturated fatty acids (palmitic & stearic) of the extracted oil did not change while, oleic (18:1) increased by 4.76 percent point, while linoleic (18:2) decreased by 4.51 percent points and linolenic (18:3) by 2.11 percent points when the parental plants exposed to water stress at R3 stage up to harvest maturity compared to non-stressed plants. These results indicated that to obtain high quality seeds from irrigated soybean in dry region such as Egypt, it is important to avoid exposing soybean crop to water stress before physiological maturity because the nutritional value of seed as well as its physiological traits with regard to viability and vigor parameters well be decrease.

Keywords: soybean, water stress, seed viability, seed constituents, fatty acids.

Introduction

Growing soybean with high quality seed is one of the factors enhance its productivity through high germination percentage, high emergence ability of good and healthy seedling tolerant to adverse condition in the field to achieve recommended plant density with good distribution on the soil. For that it is very important to know the reasons of the deterioration of soybean seeds which affect its quality.

Egypt is a very dry country because the precipitation rate is about 100-150 mm / year falls on the north coast in winter season (Dec.- Feb.), while soybean grown in summer season (June- Sept.), thus its cultivation in Egypt depends on irrigation only. However, Egypt faces many challenges with regard to fresh water resources due to the fixed share from Nile water, high rate of Egyptian population increases and the conflicts around the water supply from the Blue Nile. Furthermore, the expected climate changes may affect the amount and distribution

Of rainfalls on Nile sources which represent the main water supply for Egypt. This means that Egypt may face a great shortage in irrigation water in the near and far future. The insufficient irrigation water may expose the field crops in Egypt, especially the summer ones, to water stress which may be affect their seed yield and its quality, thus the objectives of this investigation are to study the effect of water stress on physical seed traits, seed constituents, seed viability and seed vigor parameters of soybean.

There are a lot of research work indicated that climate change and water stress caused a marked reduction in soybean yield (Desclaux et al, 2000; Frederick et al, 2001; Thornton et al, 2014; Leng and Hall, 2019; Wijewardenaet al, 2019a). Some studies indicated that the abiotic stresses could induce transmittable effects for the F1 generation of parental plants (Figueroa et al 2010, Tielborger and Petru, 2010 and Cendan et al 2013). In context, Wijewardanaet al. (2019 b) stated that the environment of maternal plants has strong effect on germination and vigor of the produced seeds. They found that water stress during the seed filling period has pronounced effect on seed quality and this effect has transgenerated to the progeny and affect the seedling vigor. Exposing soybean to water stress after flowering decreased seed germination percentage and germination rate of exposing parental plants, also seed size and weight strongly affected (Wijewardanaet al. 2019a). This means the maternal environment has marked effect on germination velocity, viability and vigor of produced seeds.

Dornbos et al (1989) stated that the germination and vigor of the produced seeds from plants exposed to water stress during seed filling period of soybean was slightly decreased and this was attributed with reduction in seed weight. Dornbos and Mullen (1991) added that this effect was increased when the stress accompanied with high air temperature (35 C°). Egli and Zhenwen (1991) stated that the reduction in carbon assimilation due to drought during seed filling period of soybean resulted smaller and lighter seed accompanied with poor seed quality. Thus, Du et al (2020) stated that exposing soybean plants at early seed formation stage (15 day after flowering) caused no reduction in seed weight. Thereafter, it decreased by 41.5% when the plants exposed to water stress during the period of 30-45 days after flowering.

Vieira et al (1991) found that few misshapes and shrivelled seeds were produced due to water stress during R5and R6 growth stages of soybean, however hard and small seeds were increased. In context, seed germination and vigor were slightly affected.

High and rapid percentage of seed germination and seedling emergence are needed to obtain the recommended plant population density and high yield (Ghassemi-Golezanied et al,2010). Water stress during seed filling period affected seed quality of soybean, the plants supplied enough water produced large, heavy and uniform seeds (Ghassemi-Golezanied et al, 2012).

Under the condition of controlled greenhouse, Dornbos and Mullen (1992) found that seed oil content decreased while protein content increased when soybean plants exposed to drought during seed filling stage. However, Shamsi(2015) stated that holding up irrigation at flowering stage of soybean decreased seed oil content without significant increase in protein content. However, Naoe et al. (2021), in Brazil, stated that seed oil and protein content were not significantly affected by drought stress (25% of ET) starting at R3 stage.

Ghassemi-Golezani et al. (2012) found that the highest EC of the seed leachate, lowest viability, lowest germination%, lowest germination rate and lowest seedling dry weight were resulted when Soybean plants were

subjected to drought stress during seed filling period. They added that the EC significantly and negatively correlated with germination attributes and seedling dry weight. Unless, there is a positive and significant correlation between seed weight and each of germination rate and percentage as well as seedling dry weight. On other hand, it negatively correlated with EC of the seed leachates. Also, they found the EC significantly and negatively correlated with the former mentioned traits

In Egypt, Morsy et al. (2018) irrigated soybean at irrigation interval 15, 20 and 25 days. They reported that increasing irrigation interval decreased seed weight, germination% and seed oil content and increased seed protein content and EC of the seed. They added that seed germination was significantly and positively correlated with oil and protein while, it negatively correlated with seed electrical conductivity.

Soybean contains more unsaturated fatty (80-85%), acids with high level of linoleic (18:2), followed by oleic (18:1) and linolenic (18:3), while it contains less saturated fatty acids (stearic-18:0) and palmitic-16:0). The fatty acid composition of oil affects its stability and health functions. The fatty acid fractions of the oil of oil seed crops could be modified genetically and may be change due to growing under certain environment. (Kandil et al. 1990, Kandil et al. 2017 a, Kandil et al 2017b) on soybean). Lee et al (2008) grown nine genotypes of soybean with and without irrigation and found that irrigation increased oleic (18: 1) and linolenic (18:3), while decreased linoleic (18:2) contents. In context, Gao et al. (2009) stated that the precipitation rate during soybean growing season affects its oil fatty acid profile, oleic and linoleic are most affected than linolenic and saturated fatty acids, i.e., palmitic and stearic. Similar results were found by Bellaloui et al (2011) they grown four soybeans' genotypes under water stress (-90 to -100 KPa) and under non - stress (-15 to 20 KPa) in the greenhouse. They found little variation in saturated fatty acids (16:0 and 18:0), while the monounsaturated oleic (18:1) was increased under stress conditions, while the polyunsaturated ones (18:2) and (18:3) were decreased. However, Ozkan and Kulak (2013), in sesame, found that water stress caused reduction in oleic (18:2) content but the linoleic did not change. Meanwhile, Osman and Taha (1975), in sunflower, Joshan et al (2019)in safflower, and Wijewardanaet al. (2019) in soybean, reported that water stress increased oleic (18:1) content in the oil of these crops. In context, Boydak et al. (2002), in Turkey, reported that irrigation soybean every 12 days increased linoleic and decreased oleic fatty acids contents.

Materials and methods

Growingconditions

Two field experiments were carried out during 2018 and 2019 summer seasons at Agricultural experiments and Research Station, Faculty of Agriculture, Cairo University, at Giza, Egypt (30.00N, 31.3E and elevation of 24m). To obtain seeds supplied from plants exposed to water stress during the seed formation, soybean Giza111 cv. was sown in four strips (each strip contains 30 rows, 60 cm apart, 5 meter long). Strips were separated by canals 150cm width to avoid water leakage between strips.

Soybean seeds were drilled manually at rate of 20 gm. / row 5 m long, (70 kg /ha.)in a pre-irrigated soil and when soil contains enough moisture to seed germination. The soil of the experimental site was clay loam in texture, has 42.3% and 16.3% moisture at field capacity and wilting point, respectively, and contains 9.5 ppm available N, 3.0 ppm available P and 580 ppm available K, with pH of 7.7 and EC 1.3 ds/m (1:2.25) as an average of both seasons. Calcium super phosphate ($15.5\% P_2O_5$) was added before ridging at 30 kg P_2O_5 /faddan (one faddan = 4200 m² =

0.42 ha). Just before 1st and 2nd irrigation, weed control was done by hand hoeing twice, two and four weeks after sowing. Nitrogen fertilizer (60 kg N / faddan) as ammonium nitrate (33% N) was added in two equal doses at 15 and 30 days after sowing. The sowing date was on May 30 in 2018 and June 23 in 2019.

Meteorological data

Meteorological parameters in the 2018 and 2019 growing seasons of soybean were obtained from Agrometeorological Station at Giza, Egypt from May to October in 2018 and 2019 seasons (Figures 1-2).



Figure 1. Maximum, minimum, average and day time temperature during the growing season.



Figure 2. Relative humidity percentage (RH) and possible sunshine duration (PSSD) in hours during the growing season.

Treatments

1- Non -stress (control): soybean of the first strip; irrigated at 15-day interval until harvest maturity on 30/9 and 31/10 in 2018 and 2019, respectively.

2- Mild Stress: soybean of the second strip; irrigated at 15- day interval until R7 growth stage, i.e., physiological maturity (appearance one mature colored pod on the main stem), plants reached this stage at 11/9and 9/10 in 2018 and 2019 seasons, respectively.

3-Moderate Stress: soybean of the third strip; irrigated at 15- day interval up to R5 growth stage (seed formation in a pod at one of the four uppermost nodes on the main stem), plants reached this stage at 25/8 and 24/9 in 2018 and 2019 seasons, respectively.

4- High Stress: soybean of the fourth strip; irrigated at 15- day interval up to R3 growth stage (appearance a pod 4-5 mm on one of the four uppermost nodes on the main stem plants) reached this stage at 14/8 and 7/9 in 2018 and 2019 seasons respectively.

Data Recorded

Soil moisture content

For treatment 1 (control), soil moisture was measured at a depth of 25 cm at mid irrigation intervals and the end of irrigation intervals. For treatments (2, 3, 4), readings continued to be taken at the same time as the first treatment even after the irrigation has been hold up using Delta-T Devices - Model HH2 - Moisture Meter.





Figure 3. Soil moisture content of non -stress treatment at mid and end of irrigation intervals during the growing season.



Figure 4. Soil moisture content of mild stress treatment at mid and end of irrigation intervals during the growing season.



Figure 5. Soil moisture content of moderate stress treatment at mid and end of irrigation intervals during the growing season.



Figure 6. Soil moisture content of high stress treatment at mid and end of irrigation intervals during the growing season.

Seed Measurements

Harvesting was done on dates as shown in 30/9, 27/9, 24/9 and 20/9 in 2018 and on 30/10, 29/10, 27/10 and 23/10 in 2019 for the non-stress, mild stress, moderate stress and high stress treatments, respectively. When 95% of the plant pods reached to color of maturity (yellow-brown), plants of each strip were uprooted, tied in bundles, left to dry in shaded open air field lab for 4-5 days. Thereafter, pods were hand separated, moved to the lab for seed separation by hand, taking care to avoid loss any seed. The seeds left to dry at room temperature with ventilation, when the seed moisture content reached to 8-10 %, they stored in textile bags until to carry out the following measurements, six months after harvesting date of each treatment.

Physical seed characteristics

Four seed samples (500 gram each) were randomly taken from the yield of each strip, the following traits were recorded:

1. Defective seeds percentage (DS %), by weight: (damaged, shriveled, infected, atrophic and misshaped seeds).

2. Green seeds percentage (GS %), by weight: the seeds of green color (dark or light) were separated and weight, then calculated as percentage.

 $\label{eq:seedspectrum} 3. Normal seeds percentage (NS\%), by weight: the rest of 500 gm. seeds sample was calculated as percentage.$

4. Seeds with cracked seed coat percentage (SCSC%): four replications of 100 normal seeds for each were mixed with red powder in color, next day the seeds were visually fixed, those with red cracks on seed coat were counted and calculated as percentage.

5.500- seed weigh-g (SW): as an average weight of four samples of 500 normal seeds taken randomly from the yield of each treatment.

The rest of the normal seeds were used for the following tests

Seed viability and vigor parameters

1- Germination test was performed using the normal seeds only, four replications of 25 soybean seeds from each treatment were soaked in a 5% hydrogen peroxide for 3 minutes, then rinsed several times with distilled water before replaced and sown in sterilized sand in petri dishes 12.5cm diameter then placed in an incubator at $25\pm2^{\circ}$ C. Total number of germinated seeds were counted daily until the 7th day the recorded data at the 7th day were used to calculate the following traits:

1. Seed germination percentage (SG %), was calculated as described in the Rules for Testing Seeds (Association of Official Seed Analyst (A.O.A.C.) 1986).

2. Speed germination index (SGI): It was calculated as described in the Association of Official Seed Analysis (A.O.A.C., 1983) by following formula:

SGI= (No. of germinated seed/days of first count) + (......) + (No. of germinated seed/days of final count). 3-Percentage of normal seedling (NSL %): the seedling considered normal when it has all of the essential structures present for normal growth.

4. Percentage of abnormal seedling (ASL %): the seedling considered abnormal when it missing one or more of its essential seedling structures; this maybe the root, the shoot or the terminal bud.

5. Seedling length (SLL): average length (cm) of 10 seedlings.

- 6. Seedling dry weight (SLDW): average weight (g) of 10seedlings.
- 7. Seed vigor index I (SVI-I) = Germination (%) x Seedling length (Root +Shoot)

8. Seed vigor index II (SVI-II) = Germination (%) x Seedling dry weight (Root +Shoot) According to Abdul Baki and Anderson (1973).

9. Seed electrical conductivity (EC) by using electrical conductivity cell model 115, samples of 100 seed of each treatment in four replicates, 25 seed each, according to the procedures outlined by (ISTA, 1993). The seeds were weighed and placed in Erlenmeyer flasks (250ml) containing 200 ml of deionized water and cover by aluminum foil. The flasks were then placed in an incubator chamber at 25° C for 24 hours. The conductivity of seed steep water was measured immediately after the removal of samples from the incubator with a pipette-type conductivity cell attached to a bulk conductivity meter. The seed conductivity values were expressed as uS / cm/g results put in the formula:

Conductivity (uS cm-1)

-----=uS/cm/g

Seed sample weight (g)

The seed viability and vigor tests were carried out at Laboratory of seed technology Research Department, Agriculture Research center (ARC) at Giza, Egypt.

Seed constituents

Four seed samples were randomly taken from the stored normal seeds of each treatment for determination the following seed constituents:

1. Oil content (%) Crude oil content (%) was determined using Soxtherm apparatus (A.O.A.C., 2000).

2. Protein content (%): nitrogen content (%) was determined using kjeldatherm and vapadest50s apparatus (A.O. A.C., 1995), and then N content was multiplied by 6.25.

- 3. Total Carbohydrate (%): was determined according to Dubois et al. (1956)
- 4. Ash (%): was determined according to A.O.A.C.) (1990).
- 5. Fiber (%): was determined according to A.O.A.C. (2005) Gaithersburg, Maryland, U.S.A.

Fatty acid composition in the soybean oil

One sample from the normal seeds of each treatment of 2018 season were used to extract the oil and determination fatty acids profile according to standard official methods (ISO). The preparation of methyl esters of fatty acids (FAME) was carried out according to the International Standard ISO 5509:2000 - boron tri fluoride (BF3) method (EN ISO 5509:2000). The fatty acid methyl esters were analyzed by using a Shimadzu GC-2010 Plus gas chromatography system, equipped with auto sampler, oven, flame ionization detector, and Lab solution software (version 2.32.00).

Statistical analysis

Data were statically analyzed using (MSTAT-C v. 3.1., 1988). Least significant difference (LSD) was applied to compare mean values.

1. Results of the physical seed traits, seed viability, seed vigor and seed constituents' parameters were statistically analyzed as completely randomized design.

2. Relationship between seed viability, seed vigor parameters and physical seed characteristics as well as chemical analysis traits were examined through simple correlation coefficient.

3. Simple linear equation and coefficient of determination between soil moisture content as independent factors and each of studied characters as dependent factors were computed.

Results

Physical seed traits:

Table 1shows the effect of water stress during seed formation of soybean on its physical seed traits in 2018 and 2019 seasons. Results revealed that the tabulated traits were significantly affected by the applied treatments. The greatest normal seed percentage (NS%) was observed for the plants grown without stress (82.30% as an average of the two seasons), and significantly decreased from the seed of plants exposed to water stress with start of R7 (mild stress), R5 (moderate stress) and R3 (high stress) in 2018. This was also true for moderate and high stress in 2019 (Table 1).

With regard to defective seed percentage (DS %), results in Table 1 revealed the lowest value was observed for the seed of non- stress treatment in both seasons (10.42%, average of the two seasons). It gradually and significantly increased with increase the period of water stress, i.e., when water stress started with beginning of R7, R5 and R3 (mild, moderate and high stress treatment, respectively) in both seasons.

Concerning green seeds percentage (GS%), the lowest value (1.22) was observed in the seed of moderate stress treatment in 2018 and mild stress (0.67) in 2019, while the greatest value was shown in the seed of plants exposed to high stress treatment in 2018(6.85) and moderate stress in 2019(5.52).

With regard to seed of cracked seed coat (SCS%), Table 1 show that the lowest value (9.25%, average of the two seasons) was observed in the seeds of plants grown under the condition of moderate stress, however the greatest value (15.75 and 13.00) was found in the seed produced without stress in 2018 and those grown under high stress in 2019, respectively.

Table 1 show that the 500- seed weight (SW%) was significantly decreased when the plants grown under high stress treatment in 2018 and under moderate and high stress in 2019. However, the greatest 500-seed weight was obtained from the plants grown without stress in both seasons (70.55g, average of the two seasons). It significantly reduced by 2.5% and 6.3% for the moderate and high stress treatments, respectively (as an average of the two seasons).

		20	018		2019			
Physical seed Traits	Non stress	Mild stress	Moderate stress	High stress	Non stress	Mild stress	Moderate stress	High stress
Normal seeds (NS %).	85.26 a	79.55 b	70.44 c	70.17 c	91.33 A	87.91a	81.83 b	81.65 b
Defective seeds (DS %).	13.41 d	16.94 c	28.08 a	24.55 b	7.43 C	11.18 b	12.66 a	13.87 a
Green seeds (GS %).	1.39 c	3.48 b	1.22 c	6.85 a	1.19 C	0.67 d	5.52 a	4.48 b
Seeds of cracked seed coat (CSC%).	15.75 a	8.25 c	8.50 c	11.50 b	12.75 A	12.50 a	10.00 b	13.00 a
Seed weight- 500 (g) (SW).	68.50 a	67.68 a	68.30 a	63.90 b	72.60 A	70.58 ab	69.23 b	68.25 b

Table 1. Effect of water stress during seed formation of soybean on its physical seed traits in 2018 and 2019 seasons.

Means followed by the same letter (s) are not significantly different at 5% probability.

Seed viability and vigor parameters

Results in Table 2revealed that seed germination percentage (SG %) was significantly affected by the applied treatments in both seasons. The high stress treatment showed the lowest value (72.50%) as an average of the two seasons with significant difference from moderate and high stress treatments. On other side, the highest SG % (84.38%, average of the two seasons) was recorded for the non - stress treatment followed by the mild stress one (87.50%) without significant difference between them.

Regarding speed germination index (SGI), normal seedling percentage (NSL %) and abnormal seedling percentage (ASL %), Table 2 shows that significantly affect by the applied treatments in 2018 only. The greatest value of SGI (11.73) and NSL (97.77%) as an average of both seasons was obtained from non-stress treatment with significant difference from the high stress treatment in 2018 season only, however, high stress treatment showed the least value of SGI (10.10) and NSL (94.09%) as an average of the two seasons. The abnormal seedling percentage (ASL %) showed an opposite trend compared to the NSL % in both seasons (Table 2).

With regard to seedling length (SLL) and seedling dry weight (SLDW), they significantly affected by water stress treatments in both seasons. Results in Table 2 show that as an average of the two seasons, the mild stress treatment showed longest seedlings (10.21 cm), however, the heaviest seedling dry weight (2.03g) was obtained from seedling of non- stress treatment. However, according to the present results of each season, the difference in SLL was in significant in 2018 between non -stress and mild stress treatments, while in 2019 season the differences between the three stressed treatments did not reach the significance level. For that the highest value of SVI-I was observed for the mild stress treatment in both seasons (with average of 8.95) with significant difference from moderate and high stress in 2018 and from the other stress levels in 2019 (Table 2).

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On the other side, the highest SLDW (2.18 g) of non -stress was significantly surpassed the other three stress treatments in 2018. In 2019 the non -stress treatments and mild stress treatments showed nearly the same SLDW (1.88 and 1.89 g) which they significantly different from that of moderate and high stress treatments (Table 2). For that, the highest value of SVI-II (1.90) in 2018 was shown at non - stress treatment which showed high SG% (87.5%) and highest SLDW (2.18 g) (Table 2). In 2019 the highest value of SVI-II (1.57) was for mild stress treatment because it showed the highest SG% (91.25%) and highest SLDW (1.89) (Table 2).

With regard to EC, this test is based on the premise that when the seed deterioration starts and progress, the cell membrane become water permeable allowing the cell contents to escape into the soaking water, thus the EC will increase. Results in Table 2 cleared that this trait was markedly affected by water stress treatment in both seasons. The least value (26.42 and 38.23 uS / cm/g in 2018 and 2019, respectively of EC was recorded from the seed leachates of non -stress treatment with gradual and significant increase from moderate and high stress treatment in both seasons (Table 2).Data also show that the EC values of 2019 is higher than 2018 values, this may be due to the high relative humidity during seed filling period and maturity (Sep.-Oct.) compared that of same stage of 2018 season (Aug.-Sep.).

		2018				2019				
Germination Traits:	Non	Mild	Moderate	High	Non	Mild	Moderate	High		
	stress	stress	stress	stress	stress	stress	stress	stress		
Seed germination (SG %).	87.50 a	91.25 a	77.50 b	70.00 c	81.25 a	83.75 a	78.75 ab	75.00 b		
Normal seedling (NSL %).	98.44 a	97.05 a	94.13 b	91.93 b	97.09 a	97.03 a	97.02 a	96.25 a		
Abnormal seeding (%).	1.56 b	2.95 b	5.88 a	8.07 a	2.91 a	2.97 a	2.99 a	3.75 a		
Speedgermination index (SGI).	13.03 a	15.65 a	12.78 ab	9.42 b	10.43 a	11.46 a	12.0 a	10.79 a		
Seedling length (cm).	9.23 ab	10.44 a	7.99b	8.06 b	8.70 b	9.98 a	9.00 ab	9.18 ab		
Seedling dry weight (SLDW) (g/10 seedling).	2.18 a	1.78 b	1.72 bc	1.53 c	1.88 a	1.89 a	1.63 b	1.60 b		
Seedling vigor index one(SVI-I)	8.08 a	9.54 a	6.21 b	5.67 b	7.06 b	8.36 a	7.08 b	6.88 b		
Seedling vigor index two (SVI-II).	1.9.0 a	1.62 b	1.33 c	1.07 d	1.52 a	1.57 a	1.27 b	1.20 b		
Seed electrical conductivity (EC) uS cm/g.	26.42 c	25.13 c	28.76 b	38.38 a	38.23 c	39.52 c	46.43 b	57.59 a		

Table 2. Effect of water stress during seed formation of soybean on seed viability and seedling vigor attribut	tes
traits in 2018 and 2019 seasons.	

Means followed by the same letter (s) are not significantly different at 5% probability.

Seed constituents

Table 3 show that the effect of water stress during seed formation of soybean on its seed constituents was significant in 2018 and 2019. Seed oil content (%) decreased when the soybean plants exposed to water stress at R3 growth stage (high stress) and thereafter in both seasons. This reduction estimated by 3.15percent points in 2018 and 3.29percent points 2019, compared to non - stress treatment. However, water stress with start of R7 stage (physiological maturity) did not cause significant reduction in seed oil content in both seasons (Table 3).

With regard to protein content, it significantly reduced in both seasons, this reduction was pronounced under the condition of high stress treatment (hold up irrigation at R3 stage which estimated by 2.52percent points in 2018, while it was 4.72 percent points in 2018(Table 3).

Results in Table 3 show that the highest values of total carbohydrate (19.50 and 21.12) were observed in seeds of plants exposed to water stress which start at R3growth stage (high stress treatment and thereafter) in both seasons. Results indicated that as seed oil and protein contents were decreased with each increase of stressed period, carbohydrate was increased in both seasons.

Despite of the significant effect of applied treatments on ash and fiber contents in both seasons, their range was very narrow for both traits. Ash content increased from 5.75% (moderate stress) to 6.10% (mild stress) in 2018, while it increased from 5.72% to 5.88% on the order in 2019. With regard fiber content, it increased from 16.46% (non stress) to 17.70% (high stress) in 2018 and from 15.32% to 16.42% in 2019 on the same order (Table 3).

	2018				2019			
Seed constituents:	Non stress	Mild stress	Moderate stress	High stress	Non stress	Mild stress	Moderate stress	High stress
Oil content (%).	18.25 a	17.56 a	16.23 b	15.10 b	20.59 a	20.12 a	18.42 b	17.70 b
Protein content (%).	40.60 ab	41.65 a	39.80 b	38.08 c	37.55 a	35.83 b	34.33 c	32.83 d
Carbohydrate (%).	17.98 ab	17.04 b	19.19 a	19.50 a	18.11 a	18.42 b	18.80 ab	21.12 a
Ash (%).	5.78 b	6.10 a	5.75 b	6.00 ab	5.86 a	5.88 a	5.72 b	5.83 a
Fiber (%).	16.64 bc	16.48 c	17.34 ab	17.70 a	15.32 b	15.72ab	16.42 a	16.29 ab

Table 3. Effect of water stress during seed formation of soybean on its chemical constituents in 2018 and 201	9
seasons.	

Means followed by the same letter (s) are not significantly different at 5% probability.

Fatty AcidComposition

Table 4 shows the effect of water stress during seed formation on the main fatty acids of soybean oil. Results indicate that the unsaturated fatty acids (16:0) and (18:0) were not greatly changed. On other side, the monounsaturated fatty acid (18:1) tend to increase with each increase of stress period, while the poly unsaturated fatty acids (18:2) and (18:3) tend to decrease.

The highest value (26.74%) of oleic (18:1) was observed at high stress treatment when the irrigation holds up with start of R3growth stage compared to 21.98% at the non -stress treatment. On other hand, linoleic acid (18:2) took an opposite trend, it decreased from 52.96% for the no - stress treatment to 48.45% for the high stress treatment. The Linolenic acid (18:3) show similar trend of linoleic but smaller figures (6.57% to 4.46%) on the same order (Table 4).

		Fatty acids (%)									
Treatment	Palmitic (16:0)	Stearic (18:0)	Oleic (18:1)	Linoleic (18:2)	Linolenic (18:3)						
Non stress	12.79	4.51	21.98	52.96	6.57						
Mild stress	12.76	4.59	22.56	50.09	6.94						
Moderate stress	12.96	4.68	25.43	48.60	5.42						
High stress	13.18	4.67	26.74	48.45	4.46						

Table 4. Main fatty acids (%) in the oil as affected by water stress during seed formation.

Correlation study

To know which seed component is more attributed to seed viability and vigor, simple correlation coefficient between seed constituents and these parameters was computed and illustrated in tables5and 6 for 2018 and 2019, respectively.

Results in Table 5 indicated that seed oil and protein contents are positively and highly significantly associated with all tabulated viability and vigor parameters, except seedling length with both components and seedling dry weight with protein content. On the other hand, abnormal seedling percentage showed negatively and highly significant relationship in 2018 season (Table 5). Also, in 2019, SG%, SLDW and SVI-II were positively and significantly correlated with oil and protein content (Table 6).

With regard to the relationship between carbohydrate content, the viability and vigor parameters Table 5 show negative and significant or highly significant relationship with each of germination %(in both seasons), normal seedling %in 2019, SVI-I in 2018 and (SVI-II)this was true for SG% only in 2019 (Table 5&6).

Table 5 and 6 cleared that seed ash content show no significant association with all parameters of seed viability and vigor in both seasons.

With regard to the relationship between seed fiber content and these parameters, there are negative and significant or highly significant associations with all parameters, except seedling length and seedling dry weight in 2018 only (Table 5).

Parameters	Oil content (%).	Protein content (%).	Total Carbohydrate (%).	Ash (%).	Fiber (%).
Germination(SG) (%).	0.780**	0.838**	-0.602*	0.11	-0.742**
Normal seedling (NSL) (%).	0.697**	0.707**	-0.443	-0.057	-0.699**
Abnormal seedling (ASL) (%)	697**	-0.707**	0.443	0.057	0.699**
Seedling length (SLL) (cm).	0.381	0.476	-0.488	0.454	-0.458
Seedling dry weight (SLDW) (g/10 seedling).	0.689**	0.476	-0.293	-0.109	-0.428
Seedling vigor index one (SVI-I).	0.567*	0.664	-0.573*	0.350	-0.614*
Seedling vigor index two (SVI-II).	0.805**	0.675**	-0.456	-0.027	-0.616*

 Table 5. Simple correlation coefficient (r) between Seed constituents parameters, Germination characters and

 Seedling establishment Parameter in 2018 season.

Table6. Simple correlation coefficient (r) between Seed constituents' parameters, Germination characters andSeedling establishment Parameter in 2019 season.

Deremeters	Oil content	Protein	Total Carbohydrate	Ash	
Parameters	(%).	Content (%).	(%).	(%).	Fiber (%).
Germination(SG) (%).	0.652**	0.618*	-0.655*	0.241	-0.184
Normal seedling (NSL) (%).	0.306	0.401	-0.642**	0.064	-0.015
Abnormal seedling (ASL) (%)	-0.306	-0.401	0.642**	-0.064	0.015
Seedling length (SLL) (cm).	0.005	-0.037	-0.180	0.418	0.030
Seedling dry weight (SLDW) (g/10 seedling).	0.734**	0.645**	-0.369	0.189	-0.439
Seedling vigor index one (SVI-I).	0.308	0.258	443	0.443	-0.056
Seedling vigor index two (SVI-II).	0.840**	0.754**	-0.568*	0.251	-0.404

Relationship between soil water content and seed quality traits

Soil moisture content was measured at mid and end of irrigation intervals, then the linear regression equation and coefficient of determination (R^2) with seed quality traits were computed.

Physical seed traits

According to the slope values tabulated in Table 7, it could be concluded that the change in soil moisture at the mid of irrigation interval do not cause valuable change in physical seed traits. However, the values of slope at the end of irrigation (Table 8) were greater than that at mid irrigation interval. The values indicated that each increase in soil moisture content at end of irrigation interval could increase normal seed % and 500 seed weight, but decrease the defective and green seed percentage (Table 7 and 8). According to R² values, the

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variation in soil moisture at mid irrigation interval was responsible about small portion of variation in normal seeds, defective seeds percentages and 500-seed weight in both seasons (Table 7). However, it was responsible for 81.4% of variation in green seeds in 2019, 44.8% and 56.1% of variation in seeds of cracked seed coat percentage in the two successive seasons.

With regard to the soil moisture content at the end of irrigation interval, it was responsible for about 43.5% and 60.7% of variation in normal seed percentage in the two successive seasons, 82.6% variation of defective seed percentage in 2018 and 87.8% of variation in green seed percentage in 2019.

Physical cood Traits:		2018		2019			
Physical seed mails.	intercept	slope	R ²	intercept	slope	R ²	
Normal seeds (NS %).	68.78	0.746	0.147	72.14	0.181	0.001	
Defective seeds (DS %).	23.18	-0.525	0.212	6.92	0.192	0.446	
Green seeds (GS %).	7.57	-0.203	0.043	49.14	-1.977	0.814	
Seeds of cracked seed coat (SCS %).	3.45	0.380	0.448	27.08	-0.692	0.51	
Seed weight- 500 (SW) (g).	63.80	0.281	0.133	62.52	0.238	0.245	

 Table 7. Linear regression equation and coefficient of determination of physical seed traits in relation to soil

 moisture content at mid of irrigation interval at maternal plants in 2018 and 2019 seasons.

Table 8. Linear regression equation and coefficient of determination of physical seed traits in relation to soil moisture content at the end of irrigation interval at maternal plants in 2018 and 2019 seasons.

Physical cood Traits		2018		2019			
Physical seed fraits.	intercept	slope	R ²	intercept	slope	R ²	
Normal seeds (NS %).	18.37	3.901	0.935*	46.50	2.384	0.607	
Defective seeds (DF %).	70.67	-3.359	0.826	27.88	-1.010	0.316	
Green seeds (GS %).	13.82	-0.712	0.246	26.75	-1.447	0.878	
Seedsofcrackedseedcoat (SCS %).	-10.23	1.429	0.557	6.48	0.340	0.145	
Seed weight -500 (SW)(g).	57.40	0.652	0.304	57.14	0.792	0.428	

Seed constituents

Data in Table 9 show that the slope values due to soil moisture content at the mid of irrigation interval in relation to seed constituents was smaller than that the moisture content at the end of irrigation intervals (Table10). From these results it could be concluded that each decrease in soil moisture content at the end of irrigation interval could cause a decrease in the oil and protein percentage as well as seed fiber content.

According to R^2 values in Tables9 and 10 it could be concluded that the variation in soil moisture content at the mid of irrigation intervals was responsible about small portion of variation in seed constituents. However, the variation in soil moisture content at the end of irrigation intervals was responsible about 79.3% and 72.6% of variation in oil content, 31.0% and 49.7% of variation in protein content, 35.8% and 42.3% of variation in carbohydrate content, 55.8% and 56.6% of variation in fiber content in the two successive seasons.

D Sood constituents:		2018		2019			
D-Seed constituents.	intercept	Slope	R ²	intercept	slope	R ²	
Oil content (%).	7.913	0.382	0.105	17.37	0.081	0.021	
Protein content (%).	28.385	0.502	0.158	30.62	0.199	0.057	
Total Carbohydrate (%).	19.266	-0.036	0.005	16.29	0.125	0.049	
Ash (%).	8.243	-0.101	0.494	5.55	0.012	0.181	
Fiber (%).	19.394	-0.101	0.0438	18.18	-0.099	0.223	

 Table 9. Linear regression equation and coefficient of determination of chemical seed constituents' relation to soil moisture content at mid of irrigation interval at maternal plants in 2018 and 2019 seasons.

Table 10. Linear regression equation and coefficient of determination of chemical seed constituents' relation to soil moisture content at end of irrigation interval at mother plants in 2018 and 2019 seasons.

Sood constituents:		2018		2019			
Seed constituents.	intercept	Slope	R ²	intercept	slope	R ²	
Oil content (%).	6.597	0.685	0.793	6.871	0.751	0.726	
Protein content (%).	33.185	0.461	0.310	20.034	0.918	0.497	
Total Carbohydrate (%).	23.951	-0.372	0.358	28.514	-0.572	0.423	
Ash (%).	6.272	-0.025	0.069	5.282	0.033	0.533	
Fiber (%).	20.556	-0.232	0.556	20.013	-0.248	0.566	

Seed viability and seed vigor

Results in Tables 11 and 12 indicate that the slope values of soil moisture content of parental plants at mid irrigation interval in relation to seed viability and seed vigor is smaller than that of moisture content at the end of irrigation interval. This means the increase in soil moisture content at mid of irrigation interval cause smaller change in seed viability and seed vigor parameters. For example in 2018 if soil moisture content increased by 1% at mid of irrigation interval germination could increase by 2.13 % compared with 3.72% for the increase of soil moisture content at the end irrigation interval of mother plants. On the same order, normal seedling increased by 0.74% compared with 1.43 %.

According to R^2 values, the variation in soil moisture at mid of irrigation intervals induced less variation in seed viability and seed vigor parameters compared to the variation at the end of irrigation intervals (Table 11 and 12). For example, the variation in soil moisture content at the end of irrigation intervals participate by 49.5% and 84.5% of variation in germination percentage in 2018 and 2019 seasons, respectively. However, these values were 6.90% and 9.60% on the same order due to variation in soil moisture content at the mid of irrigation intervals. Also, in the two successive seasons, the variation for normal seedling due to the variation in soil content at the mid of irrigation intervals. Also, in the two successive seasons, the variation for normal seedling due to the variation in soil moisture content at the mid of irrigation intervals. Similar findings were observed for seedling dry weight (92.3% and 88.3%) compared with (11.7% and 1.1%), as well as seedling vigor index one (SVI-I) (37.4% and 74.8%) compared with (0.20 % and 32.6%). Seedling vigor-II(SVI-II) (87.30% and 91.95%) compared to (9.40 and 0.1%) and electrical conductivity (35.60 % and 61.5%) compared to (1.50% and 37.20%) in 2018 and 2019,

respectively(Table 14 and 15). From these results it could be concluded that for growing soybean under irrigation conditions it is important to regulate the applied irrigations to avoid exposing plants to water stress during seed formation which may produce low quality seeds.

Germination Traits :	2018			2019			
	intercept	slope	R ²	intercept	slope	R ²	
Seed germination (SG) (%).	32.14	2.129	0.069	90.40	-0.473	0.096	
Normal seedling (NSL) (%).	78.25	0.738	0.091	98.01	-0.512	0.099	
Seed germination index (SGI).	- 9.68	0.965	0.204	17.372	-0.273	0.920	
Abnormal seeding (ASL) (%).	21.75	-0.738	0.092	1.99	0.051	0.099	
Seedling length (SLL) (cm).	11.43	-0.108	0.012	12.33	-0.137	0.381	
Seedling dry weight (g/10 seedling).	00.01	0.077	0.117	1.59	0.007	0.011	
(SVI- I)	554.95	7.782	0.002	1096.90	-15.980	0.326	
Seedling vigor index two SVI-II. Seed electrical conductivity	- 6.5556	0.922	0.094	14.589	-0.0283	0.001	
(EC) uS / cm/g.	35.35	0.450	0.015	101.07	-3.075	0.372	

able 11. Linear regression equation and coefficient of determination of germination attributes traits relation
to soil moisture content at mid of irrigation interval at maternal plants in 2018 and 2019 seasons.

Table 12. Linear regression equation and coefficient of determination of germination attributes in relation tosoil moisture content at the end of irrigation interval at maternal plants in 2018 and 2019 seasons

	2018			2019		
Germination Traits:	intercept	slope	R ²	intercept	slope	R ²
Seed germination (SG) (%).	26.26	3.72	0.495	43.36	2.211	0.845*
Normal seedling (NSL) (%).	74.12	1.43	0.798	94.41	0.148	0.330
Seed germination index (SGI).	4.44	0.55	0.158	11.916	-0.045	0.010
Abnormal seeding (ASL) (%).	25.88	-1.43	0.798	5.59	-0.148	0.330
Seedling length (SLL) (cm).	4.01	0.33	0.273	5.80	0.208	0.352
Seedling dryweight (g/10 seedling).	-0.30	0.142	0.923*	0.20	0.094	0.883*
Seedling vigor index one (SVI- I)	- 128.88	58.17	0.374	109.48	38.064	0.7482
Seedling vigor index two SVI- II.	- 12.392	1.833	0.873*	- 4.664	1.134	0.9195*
Seed electrical conductivity (EC) uS / cm/g.	58.80	-1.960	0.356	120.34	- 4.553	0.615

Discussion

Our results indicated that the physical seed traits significantly affected when the parental plants exposed to water stress during seed formation. For example, as an average of two seasons, defective seeds percentage accounted 14.63, 20.37% and 19.21% of the seed yield of the plants exposed to water stress at R7 (physiological maturity - mild stress), R5 (start of seed formation- medium stress) and R3 (start of pod formation- high stress), respectively compared to 10.42% for the plants grown without stress (Table1).

In context, green seeds percentage also increased due water stress, its percentage become 1.42%, 2.08%, 3.36% and 5.66% for the non-stress, mild stress and high stress as an average of both seasons respectively (Table 1). Thus, the normal seed percentage decreased due to exposing its mother plant to water stress during seed formation. The normal seed percentage of plants exposed to high stress treatments (hold up irrigation at R3) was 75.91%, increased up to 76.41%, 83.73% and 88.31% for the plants exposed to moderate, mild and non-stress treatments as an average of the two seasons, respectively (Table 1). This means exposing soybean plants to water stress with start of pod formation and thereafter reduce the portion of seed yield which consider normal seeds and suitable to use by farmers by nearly 25%.

The weight of normal 500-seeds decreased from 70.55gm. under the condition of non-stress to 66.08 gm. for the high stress treatment as an average of the two seasons. These results may be due to the water stress during seed formation and maturation could reduce the gas exchange and carbon fixation during photosynthesis process leading to reduction in metabolites and its translocation from source to sink. This increases immature seeds, miss shape seeds, shriveled and less weight and size seeds in seed yield of plants exposed to water stress. Some researchers stated that adverse environmental conditions during seed formation causes enforced seed maturation leading to significant decrease in seed quality (França-Netoet al., 2005; Padua et al., 2009; Ghassemi-Golezani et al., 2012; Maleki et al. 2013). Du et al.(2020) stated that drought stress during seed filling period affect soybean seed weight because drought regulate the production of photosynthesis assimilates. Drought during the early seed development stage inhibit sucrose transformation from leaves to seeds which result it reduction in seed weight.

With regard to seed constituents, our results indicated seed and oil contents decreased when plants exposed to water stress during seed formation and maturation (Table 3). As an average of the two seasons seed oil content decreased from 19.42% for the seeds of unstressed plants to 16.40% for seeds produced from high stressed plants (hold up irrigation at R3-bgning of pod formation and thereafter). On the same order, protein content decreased from 39.08% to 35.46% as an average of the two seasons, while carbohydrate content increased from 18.05% to 20.31% on the same order. These results are in agreement with results of Maleki et al. (2013) who stated that the lowest seed oil and protein contents were observed in the seeds of the plants exposed to water stress during seed filling period.

In context, despite of the significant effect of water stress treatments on ash and fiber contents (Table3)the range of their values was relatively narrow. This means, the major seed constituents i.e., protein and oil and to lesser extend carbohydrate are more affected when soybean plants exposed to water stress with start of pod formation up to maturity. These results are in line with those obtained by (Wijewardanaet al, 2019 a) who stated that the environment of maternal plants has marked effect on seed weight, oil, protein and sucrose content. Suter and Widmer (2013) suggested that the changes in seed quality due to the changes in the environment of parental plant may be due to some epigenic changes such as histone modification, DNA

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methylation and changes in RNAs function.

Regarding seed viability and vigor tests our results indicated that the highest value of germination percentage, seed germination index, seedling length and seedling dry weight were obtained from the plants exposed to water stress with start of physiological maturity without great difference from those of non-stressed plants. In context, these traits were markedly reduced when soybean plants exposed to water stress with beginning of seed formation and thereafter. This reduction was more pronounced where the water stress starts at the beginning of pod formation. As an average of both seasons the values become 72.50% for germination percentage, 10.10 seed germination index, 8.62cm for seedling length and 1.56. gm. for seedling weight. This could be explained by the high value of EC of seed leachate of moderate and high stress treatments which estimated by 37.60 and 48.17 uS / cm/g, respectively, compared with 32.32 and 32.32 for the non -stress and mild stress treatments may be caused some changes in their seeds allowed to loss some of its stored components. Amandeep et al (2013) reported that adverse conditions cause loss in cell membrane permeability which cause an increase in leaching of seed constituents and lead to decrease seed viability.

Our results indicated that the seeds of high germination percentage and other viability and vigor parameters are those contains high oil and protein contents Tables (2 and 3). The simple correlation coefficients (Tables 5 and 6) show a strong association between seed oil, protein content and germination percentage. These results go in line with those obtained by Wijewardana et al. (2019 a) who explained that higher content of oil and protein contents are able to produce more energy required for maximum and faster germination.

According to the simple regression equation and R²values (Tables7 and 8) between soil moisture content at the mid and end of irrigation intervals and physical seed traits, seed constituents, seed viability and vigor traits, it could be conducted that the soil moisture content at mid of irrigation interval was enough for soybean consumption and did not cause valuable variation in these traits. However, the variation in the soil moisture at the end of irrigation interval caused valuable variation in some traits such as normal seeds percentage, defective seeds percentage and green seeds percentage (87.8% in 2019). However small variation in seed constituents was observed due to variation in soil moisture contents, except oil percentages, its R² values indicated that 79.3% of variation in oil content in 2018 and 72.6% in 2019could be explained by the variation of the soil moisture content at end of irrigation intervals (Table8). This means there are other environmental factors, in addition to soil moisture content has shares in the variation of the most of physical seed traits and seed constituents. Some research works indicated that water stress associated with heat stress during seed formation negatively affected seed weight, seed oil and protein content (Dornbos and Mullen 1992; Abdellatifet al 2012; Bellaloui et al., 2015).

In context, according to R² values of the seed viability and vigor parameters (Table12) it could be concluded that the variation in these parameters was more pronounced due to the variation in soil moisture content at the end of irrigation intervals of the parental plants than the seed constituents. This means the variation in soil moisture content during seed formation of soybean was responsible about the most variation in its seed germination and seedling vigor parameters, for that to obtained high quality seed it is important to avoid water stress during seed formation of soybean grow n under irrigation conditions.

The present results showed that the water stress during seed formation increased the mono unsaturated fatty acid (oleic) when soybean plants exposed to water stress during seed formation, this was more obvious for the

moderate stress (start of R5stage) and high stress treatments (start of R3 stage). This may become favor results because the edible oil of high oleic content is good for oil stability, length of shelf life and health, but this may become not good for oil yield due to the reduction in seed yield caused by water stress. The increase in oleic content under the condition of water stress may be lead lower water content of plant tissues, reduction in respiration rate which cause an increase in canopy temperature. Wilcox and Cavins(1992) and Bachlava and Cardinal (2009) stated that the change of air temperature especially at seed- fill stage affect the functions of enzyme which control the biosynthesis of oil and its fatty acids in soybean. Burton (1991) explained this conclusion as the temperature may affect oleat and linoleat desaturase activities

According to the present results and results of many aforementioned investigators water stress during seed formation and maturation could increase the proportion of oleic (18:1) fatty acid in the oil which may become favored from health point of view. High oleic vegetable oil has more stability against oxidation which increase the period of shelf life and high health benefits through increasing the HDL/LDL ratio which decrease the risks of heart diseases (Waters, 2004) for that the seed companies worked hard to introduce high oleic low linoleic (HOLL) oil seed varieties to satisfy the increasing demand from these oils (Napolitano et al, 2018).

Conclusion

Our results indicated that the drought stress before physiological maturity of irrigated soybean grown in a dry and hot summer season environment looks like Egypt, has strong effect on the percentage of normal seeds without cracked seed coat and its seed weight. This cause reduction in germination percentage, seedling length and dry weight, then the seedling vigor index. Also, the nutritional value of the produced seeds was influenced due to the reduction in oil and protein content, however, the oil quality become somewhat better due to the increase in the mono- unsaturated fatty acid (oleic) and the decrease in poly- unsaturated fatty acids (linoleic + linolenic) which may be enhance oil stability.

The global faces a great change with regard to global warming and fresh water resources, these changes are become sever in hot and dry regions. Based on our results, it is important to introduce new soybean verities tolerant to heat and water stress during reproductive and seed formation stage and has the potential to transfer this tolerance to their progeny. This is important not only to increase seed production but also to enhance seed quality.

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