Running Title: *Inheritance pattern of drought stress tolerance in sesame*

Inheritance pattern of drought stress tolerance and yield related traits in sesame (*Sesamum indicum* L.)

Ammara Fazal[1]*, Dr. Farooq Ahmad Khan[1], Dr. Humera Razzaq[1] and Dr. Bushra Sadia[2].

[1]Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, [2]Centre of Agricultural Biochemistry and Biotechnology (CABB), University of Agriculture, Faisalabad.

*Corresponding author's email: pbgian123@gmail.com

Abstract

The most significant abiotic stress is drought. Plant growth and developmental processes are most negatively impacted. Although it is an oilseed crop that can withstand drought, sesame is vulnerable to it during the growth and seedling phases. Future climate change is predicted to bring about severe droughts. The current study was designed to select sesame accessions in both normal and drought-stress situations with this situation in mind. In a controlled laboratory environment, we simulated drought conditions using polyethylene glycol (PEG-6000) and observed the effects on sixty sesame plants. (*Sesamum indicum* L.)3 accessions responded to drought stress during the growth and seedling stages. Normal and drought stress treatments, also known as T0= zero (control), T1= -4 and T2= -6 bar, were developed by dissolving zero, 188 gram, and 238gram of polyethylene glycol (PEG-6000) in one 1000 millilitres of distilled water, respectively. A completely randomised design with 3 replications was used for this trial. To assess how sesame accessions responded to normal and PEG-simulated drought stress treatments, wt indexes of fresh and dry roots and shoots were calculated. Principal component analysis was used to choose the accessions that could be affected by drought and those that could handle it. The accessions (TH-6), (TS-5), (95001), (96006), (TS-3), (90005), (97007), (95010), (95013) and (93003) were drought tolerant and had maximum mean values for most of the traits. Accessions (97005), (96019-2), (97001), (96014), (93004) and (96019) were drought sensitive. Former accessions may be exploited to create sesame varieties that can withstand drought stress. This research may be useful for identifying drought-tolerant sesame cultivars to be employed in future breeding programmes as well as for comparing drought indices in a controlled experimental test.

Keywords: Drought, *Sesamum indicum*, Stress index, Tolerance, weight, Biplot, yield contributing traits, oilseed

INTRODUCTION

Sesame (*Sesamum indicum* L.), a traditional oil seed crop belonging to the family Pedaliaceae (Ashri, 1998) and Tubeflorae order (Nayar, 1976) is a crop that does particularly well in the world's tropical and subtropical regions (Gandhi, 2009). It is an annual plant that stands tall and is herbaceous. The genus Sesamum has 37 species, however only the species Indicum is commonly used for agricultural purposes (Kobayashi, 1981). It has opposing leaves and blooms that are fashioned like bells. The length of its fruit is around 2.5 centimetres, and it has an oblong capsule that contains 64-128 medium to tiny seeds. After being sown, it takes between 80 and 100 days for the plants and pods to reach maturity. (Hu and Jones., 2004). Sesame In addition to their use as raw food, seeds are also used in candy, candies, and bakery goods. Additionally, oil extracted from the seeds is used in industry for producing soap, perfume, and carbon sheets, in addition to herbal oil. (Khan *et al*., 2001; Yol *et al*., 2010). A meteorological word, drought is generally understood to refer to a period of five years during which there has been insufficient precipitation. (Jaleel *et al*., 2009).Drought may have different influences on plant growth and yield, membrane reliability, pigment content, osmotic modification , water associations, and photosynthetic action in different plants. (Benjamin and Nielsen , 2006 and Praba *et al* ., 2009).

Although sesame has an excellent drought tolerance compared to many other crops, it is especially vulnerable to the effects of drought during the germination and seedling phases. (Benjamin and Nielsen, 2006). When crops are cultivated in dry and semi-arid settings, seed germination and seedling emergence phases are essential for the plant establishment process. The crop's potential ultimate output is established at those phases of plant development. (Praba *et al*., 2009). It was also discovered that water stress influenced the germination rate and seedling development rate when using NaCl or PEG, and it was reported that the germination rate and percentage of sesame decreased as the degree of drought grew. (Chemonics, 2002; Naturland, 2002). In the same way, that drought stress harms the development of other crops, and it also harms the growth of sesame. All of these factors are taken into consideration when determining a plant's height and weight (Hussein *et al*., 2015). Germination and seedling growth slowed down as a outcome of the drought stress. It reduces the total number of seedlings and puts an end to plant development. (Almansouri *et al.,* 2001 and Farooq *et al.*, 2009, 2015). Sesame plants suffer damage from extended periods of dryness, which leads to a decreased crop output. (Mensah *et al.,* 2006). Because of the ever-increasing need for sesame, it is essential to make efforts toward creating sesame cultivars that can provide high yields in drought. In this situation, developing such kinds capable of producing a larger yield despite the restricted supply of water is required.

MATERIALS AND METHODS

Experimental details and treatments

This experimentation was conducted at the Sunflower Research Laboratory of the Department of Plant Breeding and Genetics at the University of Agriculture in Faisalabad. Sixty Sesame Seeds accessions (Table-1) were obtained from the Oilseeds Research Institute, the Ayub Agricultural Research Institute located in Faisalabad, Pakistan, and Plant

Gene Resources of Canada (PGRC). The capacity of these accessions to tolerate the effects of drought was evaluated when they were in the seedling growth stages. The experiment was carried out using a design that was triplicated, totally randomised, and factorially organised with its treatments.

Table 1. List of Sesame germplasm in experiments

Oilseeds Research Institute, AARI, FSD, Pakistan										**PGRC Canada**	
Sr. no	Accessions	Sr no	Accessions	Sr no	Accessions	Sr no	Accessions	Sr no	Accessions	Sr no	Accessions
1	40021	11	40004	21	95009	31	95010	41	70002	51	UCR-101
2	L-101	12	P-1-3	22	90005	32	90004	42	87008	52	B-14
3	No-15/12	13	G-2	23	70004	33	95013	43	P-11	53	BS-5-6-6
4	20011	14	70005	24	P-5	34	No.Sc	44	No-6112	54	Kayankulm
5	87001	15	95001	25	L-9	35	93003	45	96019-2	55	Tc-25
6	No-57	16	L-66-1	26	TS-3	36	93004	46	L-W	56	UT-43
7	G-5008	17	96006	27	L-66-2	37	No-6002	47	97001	57	95009
8	TH-6	18	40038-1	28	97007	38	No-56	48	95013-1-1	58	Bs-15
9	TS-5	19	TS-3-1	29	Black Till	39	97005	49	96014	59	Tc-27
10	40038	20	L-41	30	P-5-2	40	87001-1	50	P-6	60	P-7

*** AARI (**Ayub Agricultural Research Institute), **PGRC (**Plant Gene Resources of Canada)

The experiment was set up in three separate petri dishes using a completely randomised design with different factorial configurations. On top of filter paper that had been soaked in distilled water , 5 seeds of all accession were planted, and each replication was given a different treatment. In order to generate one control and two degrees of drought stress, the bar levels T0 = 0 (control), T1 = -4, and T2 = -6 were dissolved in water. 0, 188 and 235 g L^{-1} of PEG-6000 in distilled water at 25°C respectively (Bahrami *et al*., 2012). Numbers of germinated seeds were counted daily till 15 days of sowing. Each treatment was tested with five seedlings from each accession, and data was gathered for each of the five replicates. on several seedling parameters. Fresh root wt., after fifteen days after seeding, measurements were taken of the root lengths, fresh shoot lengths, dry shoot lengths, fresh shoot wt., and dry root wt.. After drying the roots and shoots in an oven (Fisher Scientific, Model 655 F) at 65 °C for 72 hours (Tanveer ul Haq *et al.*, 2014), the dry wt.s of the roots and shoots were recorded and then reweighed in grammes using an electronic balance (Setra BL-410S).

Above recorded data were subjected to compute stress indices.

FRWi= (Fresh root wt. of stressed plant / Fresh root wt. of optimise plant) × 100.

FSWi= (Fresh shoot wt. of stressed plant / Fresh shoot wt. of optimise plant) × 100.

DRWi= (Dry root wt. of stressed plant / Dry root wt. of optimise plant) × 100

DSWi= (Dry shoot wt. of stressed plant / Dry shoot wt. of optimise plant) × 100

FRLi= (Fresh root length of stressed plant / Fresh root length of optimise plant) × 100

FSLi= (Fresh shoot length of stressed plant / Fresh shoot length of optimise plant) × 100

Statistical analysis

Analysis of variance, as described by Steel *et al* . (1997) and principal component analysis (PCA), as described by Ghaffari *et al*., (2012) were used to the data that had been recorded.

RESULTS

Analysis of variance for seedling indices is presented in Table 2. All indices of seedling qualities were examined, and the only exceptions were the fresh and dry root wt. indices. The accessions were found to vary significantly from one another. All of the characteristics were substantially different across treatments except for the fresh root wt. index. The accessions by treatment interaction did not turn out to be significant for any of the indicators. T2 stress levels, as measured by all indices, were much lower than those of T1, which was the reference point. This indicates that genetic variations among the accessions were subjected to different treatments, which may be used in the subsequent breeding program.Fresh root

Length index:

It ranged from 0.80 cm to 5 cm, 0.75 cm to 6.56 cm and 0.90 cm to 7.50 cm for T0, T1 and T2 respectively. Stress indices for fresh root length were observed 2.90% to 90.44% for T1 and 2.30% to 21.64% for T2.

Fresh shoot length index:

Ranges of fresh shoot length were observed from 1.10 cm to 8 cm for T0, 1.50 cm to 10 cm for T1 and 2.80 cm to 11.50 cm for T2. Stress indices for this trait ranged for T1 was 2.48% to 12.90% and for T2; 5.30% to 21.64%.

Fresh root weight index

It ranged from 0.0020 g to 0.0138 g, 0.002 g to 0.0094 g and 0.0012 g to 0.0220 g for T0, T1 and T2 respectively. Stress indices for fresh root wt. were observed 4.73% to 176.67% for T1 and 2.68% to 125.67% for T2.

Fresh shoot weight index

Ranges of fresh shoot wt. were observed from 0.0034 g to 0.0337 g for T0, 0.0065 g to 0.0288 g for T1 and 0.0038 g to 0.0593 g for T2. Stress indices for this trait ranged for T1 was 5.09% to 107.1% and for T2; 0.07% to 99.67%.

Dry root weight index

It ranged from 0.0003 g to 0.0013 g, 0.0002 g to 0.0004 g and 0.00012 g to 0.0002 g for T0, T1 and T2 respectively. Stress indices for fresh root wt. were observed 5.07% to 106% for T1 and 2.08% to 13.92% for T2.

Dry shoot weight index

Ranges of fresh shoot wt. were observed from 0.00031 g to 0.0024 g for T0, 0.0002 g to 0.0009 g for T1 and 0.0001 g to 0.0010 g for T2. Stress indices for this trait ranged for T1 was 5% to 106% and for T2; 0.06% to 94.37%.

Table 2. Stress indices in sesame accessions under normal and drought stress treatments: mean square values from analysis of variance

SOV	DF	FRLi	FSLi	FRWi	FSWi	DRWi	DSWi
Accessions (A)	59	3791.432*	1045.8632*	1312902*	298916	457.2	885102*
Treatments (T)	1	2719861*	5991.47*	104045	9.615*	22961.2 *	3367661*
A × T	59	1455.1	434.99	299258	2719861	455	198241
Error	238	1733.4	421.37	642775	3567837	0.0035	31.44

*=Significant at 0.05 probability level

SOV=Sources of variation, DF= Degrees of Freedom, FRLi= Fresh root length index, FSLi= Fresh shoot length index , FRWi= Fresh root wt. index, FSWi=Fresh shoot wt. index, DRWi=Dry root wt. index, DSWi=Dry shoot wt. index

Principle component analysis

When there are a large number of accessions to be picked and a large number of characteristics to be considered, principal component analysis can help simplify the process of selecting genotypes. Within the context of this study, every stress index exhibited a high total variance and more than one eigen value. a series of biplots illustrating the principal components i.e. PCA 1 for T1 and PCA 2 for T2 are presented in Fig. 1 and Fig. 2, respectively. In PCA1, accessions 27(95001), 49(TS-3), 50(90005), 53(95010), 60(97007), 28(TS-5), 18(TH-6), 17(96006), 54(95013), 26(40021), 48(93003) and 46(L-101) fall in Quadrate I and accessions 1(No-12/12), 3(20011), 6(87001), 34(No-57), 2(G-5008), 8(40038), 24(40004), 27(95001), 7(P-1-3), 5(G-2), 55(70005), 35(L-41), 38(P-5) and 58(L-9) were present in Quadrate II. Therefore, these accessions were considered drought tolerant . Accessions 12(P-11), 36(96014), 59(UCR-101), 21(93004), 16(No-Sc), 52(L-W), 14(97005), 33(TS-3-1), 22(P-5-2), 20(97001), 29(P-6), 56(87001-1), 4(96019) and 9(96019-2) in Quadrate IV were taken as drought sensitive. From the PCA 2, the accessions 45(95013-

1-1), 50(90005), 52(95009), 28(TS-5), 44(70004), 49(TS-3), 27(95001), 28(TS-5), 29(P-6), 48(93003), 5(G-2), 16(No-Sc), 18(TH-6), 54(95013), 53(95010), 46(L-101) and 60(97007) in Quadrate I and accessions 17(96006), 35(B-14), 6(87001), 11(BS-5-6-6), 40(Tc-25), 22(P-5-2), 43(UT-43), 41(Kayankulm), 38(P-5) and 31(70002) in Quadrate II were considered drought tolerant. Accessions 1(No-12/12), 3(20011), 4(96019), 19(90004), 2(G-5008), 21(93003), 42(Black till), 36(96014), 20(97001), 9(96019-2), 14(97005), 10(No-56), 55(70005) and 24(40004) in Quadrate IV were considered drought sensitive . Based on the results of Principle Component Analysis accessions 18(TH-6), 28(TS-5), 27(95001), 17(96006), 49(TS-3), 50(90005), 60(97007), 53(95010), 54(95013) and 48(93003) were selected as drought tolerant and 14(97005), 9(96019-2), 20(97001), 36(96014), 21(93004) and 4(96019) as drought sensitive.

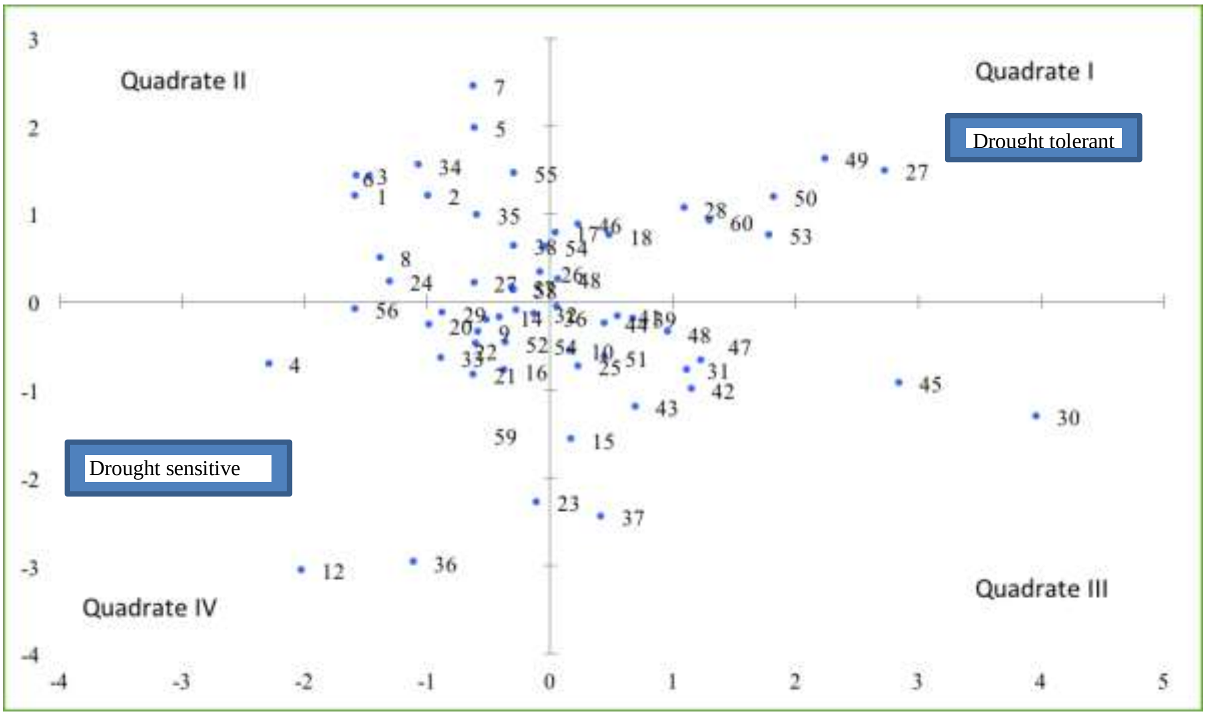

Fig. 1 Principal component Analysis 1 for stress indices of T1 in sesame accessions

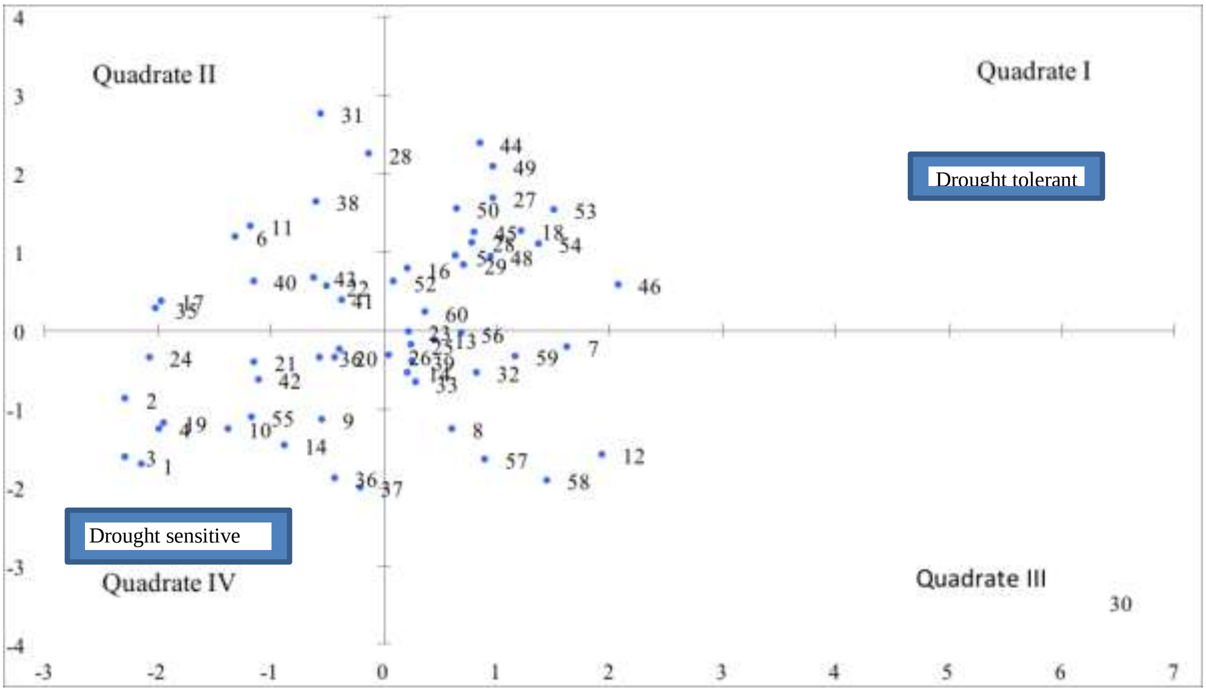

Fig.2 Principal component Analysis 2 for stress indices of T2 in sesame accessions

DISCUSSION

The experiment revealed that the drought stress levels harmed all indicators, including fresh root length indices, fresh root wt. indices, fresh root wt. indices, fresh shoot wt. indices, dry root wt. indices, and dry shoot wt. indices. These indices all decreased as the levels of drought stress increased. When the drought stress was increased, there was a drop in the wet seedling wt. index and a decrease in the dry seedling wt. index. Additionally, there was a reduction in the root length by Nath *et a l* (2001); Ahmad *et al*. (2009), Baghery *et al*. (2022), Hassanzadeh *et al.* (2010), Mehropouyan *et al*. (2011), Gabriel (1981), Yan and Kang (2011), Zlatev and Lidon (2012), Hussein *et al*., 2015, Tripathy *et al*., 2019 and Hussain *et al*. (2013). The plant growth and biomass are severely stunted by the strain caused by the drought. Those accessions are drought resistant which have the ability to maintain their growth rate as well as their fresh and dry wt.s under the stress of drought, and vice versa.. (Ahmad *et al*. 2022; Sharma *et al*. 2003; Boureima *et al*., 2012; Fazeli *et al.*, 2006; Kaya and Atakisi 2016; Gvozdenovic *et al.,* 2005; Hladni *et al*., 2006; Krishnaiah *et al*., 2010; Andarkhor *et al*., 2012 and Kar *et al*., 2001)

However, there is a need to utilise a strategy to increase the effectiveness of selection , which is a key difficulty in plant breeding programmes. This may be accomplished via a number of different approaches. It is regarded appropriate to use the mean values of these stress indicators for the assortment of accessions under drought stress circumstances. (Darvishzdaeh *et al*., 2010), but there is a need to use a method to develop the effectiveness of selection. When there are a large number of genotypes to choose from and a wide variety of attributes are being considered, principal component analysis is a useful tool (Gabriel ,1981). Accessions (TH-6), (TS-5), (95001), (96006), (TS-3), (90005), (97007), (95010), (95013) and (93003) were drought tolerant and had maximum mean values for most of the traits. Accessions (97005), (96019-2), (97001), (96014), (93004) and (96019) were drought sensitive. These accessions may be used to develop drought stress tolerant sesame types. There are many reports that agree with the present results that drought stress rigorously reduces the plant's growth and biomass. However, the accessions having the genetic potential to maintain higher development under stress conditions are drought tolerant.

Conclusions

It was indicated that the accessions were genetically variable due to the presence of genetic diversity between all of the accession for all indicators. (TH-6), (TS-5), (95001), (96006), (TS-3), (90005), (97007), (95010), (95013) and (93003) were drought tolerant and had maximum mean values for most of the traits . Accessions (97005), (96019-2), (97001), (96014), (93004) and (96019) were drought sensitive. These accessions might be used in the breeding of sesame varieties that are resistant to the effects of drought stress

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First Author – Ammara Fazal, Ph.D. Scholar,University of Agriculture, Faisalabad, Pakistan , pbgian123@gmail.com

Second Author – Dr. Farooq Ahmad Khan, Chairman, Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan

Third Author – Dr Humera Razzaq. PhD, Lecturer, Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan

Fourth Author– Dr Bushra Sadia, Associate Professor. Centre of Agricultural Biochemistry and Biotechnology (CABB), University of Agriculture, Faisalabad. Pakistan

Correspondence Author – Ammara Fazal, pbgian123@gmail.com , +923356622950