EVALUATION OF SESAME GENOTYPES FOR TOLERANCE TO WATERLOGGING

by

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THESIS

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DECLARATION

I, hereby declare that this thesis entitled **"Evaluation of sesame genotypes for tolerance to waterlogging"** is a bonafide record of research work done by me during the course of research and the thesis has not previously formed the basis for the award of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

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Athul .V

Dedicated to

My

Nation and Nature

CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

LIST OF ABBREVIATIONS AND SYMBOLS USED

%	Per cent
@	At the rate of
µg	Microgram
A_{460}	Absorbance at 460nm
A_{480}	Absorbance at 480nm
A_{510}	Absorbance at 510nm
A_{520}	Absorbance at 520nm
A_{645}	Absorbance at 645nm
A_{663}	Absorbance at 663nm
ATP	Adenosine tri phosphate
CD	Critical difference
Cm	Centimeter
CRD	Completely Randomized Design
et al.	and other Co workers
Fig.	Figure
GA	Genetic advance
GCV	Genotypic coefficient of variation
h^2	Heritability
h	Hours
i.e.	That is
KAU	Kerala Agricultural University
Kg	Kilogram
m	Meter
mha	Million hectare
nm	Nano meter
°C	Degree Celsius
PCV	Phenotypic coefficient of variation
r	Correlation coefficient

R^2	Residual effect
ROS	Reactive oxygen species
rpm	Revolution per minute
s	Seconds
SOD	Super oxide dismutase
TSP	Total soluble protein
VE	Environmental variance
VG	Genotypic variance
viz.	Namely
VP	Phenotypic variance

Introduction

1. INTRODUCTION

Sesame (*Sesamum indicum* L), belonging to the family Pedaliaceae is one of the important oil seed crop of India and known as "Queen of oil seed crops" by virtue of its excellent quality. It is a diploid species with 2n=26 and an annual, occasionally perennial crop which needs a growing period of 70 to 150 days. Most of the sesame seeds are used for oil extraction and the rest are used for edible and religious purposes. The chemical composition of sesame seed shows that it is an important source of oil (44-58%), protein (18-25%), carbohydrate (~13.5%) and ash (~5%) (Borchani *et al.*, 2010). Due to the presence of natural antioxidants such as sesamolin, sesamin and sesamol, oil is stable and has high keeping quality and resistance to rancidity. Sesame oil is also used in paints, soaps, cosmetics, perfumes and insecticides. Sesame seeds which are sources of phyto-nutrients such as omega-6 fatty acids, flavonoids, phenolic anti-oxidants, vitamins and dietary fiber are also used in traditional medicines for their nutritive, preventive, curative, potent anti-cancerous and health promoting properties.

India ranks first in area and production and contributes to one third of the world production. Nearly 30% of the sesame acreage in the world is in India (Bedigian and Harlan, 1986). The total area under cultivation of sesame is 1.94 million hectares with a production of 0.755 million tonnes and productivity of 332 kg ha^{-1} (Jadhav and Mohrir, 2013). The crop thrives best on moderately fertile, well drained soils with a pH ranging from 5.5 to 8.0 and is highly tolerant to drought. It grows in most of the well drained soils and various agro climatic regions and is adapted to different crop rotations. In Kerala, sesame is cultivated in summer rice fallows and rabi uplands. Onattukara is the major sesame growing tract where the soil is sandy loam with poor drainage and high water table.

Waterlogging caused by flooding, excessive rains and poor drainage is a serious abiotic stress determining crop productivity worldwide and it affects 10% of the global land area (Setter and Waters, 2003). The yield penalty resulting from

waterlogging may vary between 15% and 80% depending on the soil type and duration of stress. The situation may become worse due to climate change which may increase the frequency and severity of the abiotic stress. In waterlogged soils, compounds like carbon dioxide, ethylene, manganese and iron may accumulate in concentrations which are potentially toxic to plants. However, oxygen deficiency is the most important cause of flooding injury (Kozlowski, 1984).

Sesame is very sensitive to excess moisture and crop losses due to waterlogging are considerably high. It is mainly cultivated by small and marginal farmers under rainfed condition. Developing resistant varieties is the most ideal and economic approach to manage the stress as agronomic measures and engineering structures are costly and have their own limitations.

In this context the present study entitled "Evaluation of sesame genotypes for tolerance to waterlogging" was carried out with the following objectives.

- To identify sesame genotypes which are tolerant to excess soil moisture conditions.
- To elucidate the mechanism for tolerance.
- To find out the selection index for identification of tolerant genotypes.

Review of Literature

2. REVIEW OF LITERATURE

2.1 SESAME AND IMPORTANCE OF WATERLOGGING TOLERANCE

Sesame *(Sesamum indicum* L.), otherwise known as sesamum or benniseed, member of the family Pedaliaceae, is one of the most ancient oilseeds crop known to mankind. It is known under several names in different countries *viz:* simsim, benniseed, til, gingelly and a jonjoli (Khidir, 1997). Sesame seed contains 38-54% oil and 18-25% protein. Because of its high oil quality and a wide use in raw foods, confectioneries and bakery industries, the demand of sesame seed is increasing significantly in the global market (Ashri, 1989). Sesame oil is a good source of vegetable oil since it has antioxidants such as sesamin, sesamol and sesamolin and ideal fatty acid composition. The antioxidants make the oil very stable and it has therefore a long shelf life (Suja, 2004).

Sesame is a poor man's crop normally cultivated under rainfed condition and is very sensitive to excess moisture (Khidir, 1997). Bennet (1995) already reported that good drainage is important for sesame as it is very susceptible to short duration of waterlogging. Climate change is causing increased periodic flood throughout the world resulting in severe crop reduction (Olesen *et al.*, 2011). Development of flood tolerant cultivar is the way to overcome this problem (Hussain *et al.*, 2014). Setter and Waters (2003) defined waterlogging tolerance as the maintenance of high yields under water logged compared to non-waterlogged conditions. Some of the relevant works and their findings in connection with the present study has been reviewed and presented in this chapter.

Tolerance to waterlogging varies with species. A study conducted by Al Ani *et al.* (1985) revealed that seeds with carbohydrate reserves such as rice and wheat were generally more tolerant to hypoxia (low oxygen) or even anoxia

(absence of oxygen) than seeds with fatty acid reserves such as sunflower, cotton and sesame.

Among different oil seed crops, sesame was the most susceptible one to water logged and acidic soil condition (Ashri, 1997). Poor soil aeration associated with excessive moisture was affecting plant growth in a negative way (Boru *et al.*, 2011). De Simone *et al.* (2002) reported that in most plant species, flooding induced hypoxic and anoxic conditions in soil reducing the capability of roots to supply nutrients and water for plant growth and development and this led to the reduction of crop yield.

The degree of stress on sesame in waterlogging soils depends on the crop stage, duration of flooding, soil type, growth conditions and genotypes. When two sesame varieties, BARI Til 2 and BARI Til 3 were studied, varietal difference was seen with respect to the effect of waterlogging and its duration. Yield was rapidly decreased as duration increased. The varieties recorded a maximum yield loss of 51.67% and 58.24% respectively for a continuous period of 36 hours of waterlogging at two crop (vegetative and flowering) stages (Sarkar *et al.*, 2016).

Seedling test conducted in controlled conditions is the common method to screen for waterlogging tolerance (Zhou *et al.*, 2010). Hussain *et al.* (2014) screened sixty accessions of cotton (*Gossypium hirsutum* L.) at three different growth stages viz. seedling, flowering and boll formation stages and was found that seedling stage was the most sensitive growth stage and selection at the seedling stage can enhance tolerance to flooding stress.

2.2. EFFECTS OF WATERLOGGING ON THE IMPORTANT BIOMETRIC CHARACTERS.

According to Parelle *et al.* (2010), genetic variability for waterlogging tolerance can be assessed indirectly as survival percentage, crop damage indices

and negative influence on growth and yield. Waterlogging reduced plant height, number of primary branches plant^{-1}, number of capsules plant^{-1}, 1000 seed weight and seed yield significantly in sesame (Shajahan, 2016).

2.2.1 Survival percentage (%)

Martin *et al.* (2006) and Hussain *et al.* (2014) reported that plant responses to flooding stress in terms of survival percentage is a vital factor to assess the degree of flooding tolerance. Variability in this trait is directly related with genetic variability for flooding tolerance (Parelle *et al.*, 2010).

In rapeseed, waterlogging at the seedling stage showed a large diversity in survival ability and it was significantly correlated with the yield index (Zhou , 2014).

2.2.2 Plant height

Plant height and leaf area in castor bean *(Ricinus communis)* were reduced by waterlogging and maximum plant growth reduction was occurred when the plants were imposed to waterlogging for 9 days (Pollane, 1995).

Zhou *et al.* (1997) observed that plant height was decreased significantly by waterlogging at the seedling and stem elongation stages in sesame. In a pot culture experiment to study the effect of waterlogging on morpho physiological character of sesame , maximum plant height was found under non waterlogging condition (Hossain and Salahuddin ,2001).

2.2.3 Number of Primary Branches Plant^{-1}

Increase in duration of waterlogging caused a decrease in branch number in sesame (Ghoi *et al.*, 1996). Waterlogging at seedling and stem

elongation stages significantly reduced the number of branches plant^{-1} (Zhou *et al.*, 1997 and Hassan *et al.*, 2001).

2.2.4 Number of Capsules Plant^{-1}

Waterlogging has reported to have significant effect on capsule number, capsule weight and time taken for capsule formation. Zhou *et al* (1997) and Hossain and Salahuddin (2001) found that number of capsules plant^{-1} was significantly reduced in sesame by waterlogging.

Duration of Waterlogging significantly reduced the weight of capsules (Beltrao *et al.*, 1997). Rincon *et al.* (1997) observed that the water stress reduced duration of capsule formation and capsule number plant^{-1}.

2.2.5 Thousand seed weight

According to Choi *et al.* (1990) and Beltrao *et al.* (1997), longer period of waterlogging decreased 1000 seed weight in sesame. In contradiction to this, Hassan *et al.* (2001) reported that thousand seed weight was not affected due to waterlogging.

2.2.6 Root morphology

Dong *et al.* (1983) reported that longer duration of waterlogging affected root morphological factors like root dry weight, number of roots etc and resulted in complete decay of the root in wheat. Normal root development was replaced by the adventitious roots and it eventually led to increase in number of roots compared to control (Mano and Omori, 2007).

Satomi *et al.* (2015) studied differences in root development among ninety two soybean lines during initial growth stages in response to flooding and reported that root dry weight, root length and root surface area are important indices of flood tolerance. Flooding reduced the root and plant dry weights but not

shoot dry weight and these mean values varied widely in different lines in both flooded and controlled conditions. Total root length and root surface area were severely decreased by flooding.

2.2.7. Seed Yield

Excess water stress depending on duration and crop stage can affect crop growth and yield. In Soybean, flooding during vegetative stage caused reduction in yield and it was mainly due to decreased stem dry weight (Choi *et al.*, 1990). Persistent waterlogging for four days and eight days during vegetative, flowering and capsule development stages significantly reduced the yield by 8-11% and 13-33% respectively (Sorte *et al.*, 1997). In rapeseed decrease in yield started from three days of waterlogging and it is mainly due to lower number of seeds per plant (Gutierrez *et al.*, 1997).

Yadav and Srivastava (1997) reported that waterlogging during reproductive phase caused maximum reduction in yield in sesame. The seed yield plant^{-1} was reduced by 48 h of flooding (Hassan *et al.*, 2001). Mensah *et al.* (2009) reported that continuous flooding and severe drought adversely affected the crop resulting in low yield.

2.3 ANATOMICAL STUDY

Most of the plants are very susceptible to waterlogging condition. Oxygen diffusion in water is 10^4 times less than that in air. So roots surrounded by water have very less oxygen uptake and Adenosine triphosphate (ATP) production is greatly decreased with the oxygen deprivation, resulting in the lack of energy in waterlogged plants. This will lead to death of plants (Colmer and Voesenek, 2009).

Formation of aerenchymatous tissue made up of large intercellular spaces and adventitious roots are important traits for waterlogging tolerance.

Aerenchymatous tissue provides less resistance in internal pathway for the exchange of gases between aerobic shoot to the anaerobic root. The formation of aerenchyma is one of the most critical factors for waterlogging tolerant plants (Jackson and Armstrong, 1999).

According to Marashi and Mojadham (2014), aerenchyma tissue in wheat (*Triticum aestivum*) was developed after increasing period of waterlogging. Maximum and minimum aerenchyma tissue formation was observed under two weeks and one week of waterlogging, respectively. Waterlogging duration for three weeks caused decay and change of aerenchyma tissues. In barley, faster formation of aerenchyma and adventitious roots are key factors for waterlogging tolerance in barley (Zhang *et al.*, 2015).

Adventitious roots are developed from the submerged part of the stem in flooded plants and grow horizontally. Study of Garthwaite *et al.* (2003) revealed that adventitious root number increased in an anoxia treatment compared with that in aerated conditions in barley and wheat. So formation of aerenchymatous tissue and adventitious roots has significant role in flood tolerance.

2.4 BIOCHEMICAL STUDY

2.4.1 Chlorophyll

In all green plants present on the surface of earth, chlorophylls form the most important pigment system since these are playing a key role in the conversion of solar energy into chemical energy. The relative chlorophyll content has a positive relationship with photosynthetic rate. Chlorophyll includes chlorophyll a, b, c and d and chlorophyll a plays a leading role in photosynthesis while chlorophyll b is secondary in function.

According to Valladares and Niinemets (2008) green pigment composition analysis in leaves is very important in plant eco-physiological studies. It gives information about physiological changes of plants under water stress conditions.

Significant reduction in yield under waterlogged condition was caused due to increased leaf senescence and consequent reduction in the reproductive period in cowpea *(Vigna unguiculata)* (Umaharan *et al* ., 1997).

Wang *et al.* (1999) and Xu *et al.* (2012) reported that waterlogging treatment reduced photosynthesis and chlorophyll content markedly resulting in decrease in yield in sesame.

2.4.2 Proline

Proline, also known as L-proline, is a non-essential amino acids having multifunctional role as important cytoplasmic penetrate and dehydrating agent which can improve the water holding capability of plant tissue and protect enzymes and membrane *in vivo* (Ma ,1994) .

Plants accumulate proline in cell under water logged condition (Xing and Cai , 1998) and has a key role on the defense mechanisms under various abiotic stresses (Nanjo *et al.*, 2003). It is related also to the non-enzymatic detoxification of free radicals (superoxide, peroxide or hydroxyl) that are generated excessively under stress (Radyukina *et al.*, 2008).

Xu *et al.* (2012) reported increased proline content under flooding stress in all the three sesame genotypes of his study, proving the importance of proline under stress condition. Proline acts as an important osmolyte for osmotic adjustment and contributes to the stabilization of cell structures, protection of membranes and proteins against reactive oxygen species (ROS) (Steffens *et al.*, 2012).

2.4.3 Super oxide dismutase (SOD)

Excessive formation of reactive oxygen species (ROS) is an integral part of many stress situations, including hypoxia. Super oxide dismutase (SOD) is an antioxidant present under abiotic stress condition and its activity is increased in

cells under waterlogging and is vital in the protection of plants against oxidative stress.

Short anoxic stress increased the potential for superoxide production and it has a crucial role in the survival of the plant during flooding stress (Van Toai and Bolles ,1991). Yan *et al.* (1996) reported that prolonged flooding led to an increase in the activities of SOD in maize. Xu *et al.* (2012) reported that SOD activities in sesame leaves increased due to flooding and SOD activity in the flood tolerant sesame genotype WTG is increasing more than that of the susceptible genotype WSG.

2.4.4 Phenol

Phenols are important secondary metabolites in plants, possessing one or more aromatic rings with one or more hydroxyl groups. Plant phenolics include phenolic acid, flavonoids, tannins and less common lignins.

According to Rice- Evans *et al.* (1996) phenolics are strong antioxidants that help plant to survive stress condition and have proved to be more potent than ascorbic acid, tocopherol and carotenoid (Mittler, 2002). Short term waterlogging stress caused enhancement in polyphenol content in waterlogging sensitive plant *Chlorophytum borivillianum* (Nikam, 2007).

2.4.5 Total Soluble Protein (TSP)

There are only a few reports on changes in concentration of total soluble protein in response to anoxic or hypoxic conditions.

Plants subjected to flooding showed a decrease in total soluble proteins in roots and leaves. This may be due to use of protein as a substrate in the anaerobic respiration (Zubay, 1993). Significant reduction in total soluble protein content after flooding was reported in castor bean (Gadallah ,1995) and yellow lapacho (Alves *et al.*, 2012).

2.5 SEED YIELD AND COMPONENT CHARACTERS

The available literature on seed yield and component characters in sesame is reviewed under the following headings.

1. Variability
2. Heritability and genetic advance
3. Correlation analysis
4. Path analysis
5. Divergent studies
6. Selection index

2.5.1 Variability

Variability present in the gene pool is very important since it is a necessary requirement for selection of superior types. Variability can be partitioned in to heritable and non-heritable components measured as genotypic and phenotypic coefficients of variation (GCV and PCV) and heritability (Singh and Narayanan, 2013). PCV and GCV values ranging from 0 to 10%, 10 to 20% and >20% are ranked as low, medium and high respectively (Shivasubramanian and Menon , 1973).

Vasline *et al.* (2000) and Saravanan and Nadarajan (2003) reported high phenotypic and genotypic coefficient of variation for number of capsules plant^{-1}.

In sesame, phenotypic variances were generally higher than respective genotypic variances revealing the role of environmental factors. Highest values of PCV and GCV were recorded for number of branches plant^{-1}, number of capsules plant^{-1} and seed yield plant^{-1} and PCV was low for morphological traits. Good genetic variation (high GCV) was observed for stem height to the first capsule, number of branches plant^{-1}, number capsules plant^{-1} and seed yield plant^{-1} which shows that they were utilizable specific traits required for the development of cultivars (Shabana *et al.*, 2015).

Lal *et al.* (2016) studied variability of seeds pod^{-1}, number of pods plant^{-1}, number of pods main stem^{-1}, number of leaf nodes and days to maturity in sesame and reported highly significant differences among all the characters under study except leaf nodes per plant indicating considerable amount of genetic variation present in the material.

2.5.2 Heritability and Genetic advance

Estimates of heritability and genetic advance will be helpful in planning future breeding programmes. Heritability can be classified as broad sense heritability and narrow sense heritability. The broad sense heritability, the proportion of genotypic variance to the phenotypic variance is an important parameter for planning future breeding programmes. Johnson *et al* .(1955) stated that heritable variation with broad sense heritability estimates would give reliable indication of the expected improvement through selection.

Panse and Sukhatme (1985) stated that if the heritability was mainly due to additive and non-additive gene effects, the expected genetic gain would be low and if there are additive gene effects only, a high genetic gain may be expected through selection. Heritability values greater than 80% are very high, values from 60 to79% are moderately high, values from 40 to 59% are medium and values less than 40% are low (Singh ,2001).

In sesame high heritability and genetic advance would be valuable in selection programmes. (Mishra *et al.*, 2008). 1000 seed weight and capsule length showed very high values of heritability and low to moderate genetic advance (Reddy *et al.*, 2001; Sudhakar *et al.*, 2007). High heritability coupled with high expected genetic advance for plant height, number of capsules plant^{-1}, number of seeds capsule^{-1}, number of branches plant^{-1} and seed yield plant^{-1} was reported in sesame by Narayanan and Murugan (2013).

2.5.3 Correlation analysis

Correlation analysis is a useful technique, which provides information about the degree of relationship between plant characters and is also a good index to predict the yield response in relation to the change of a particular character (Muhammad *et al.*, 2007). According to Gnanasekaran *et al.* (2008) correlation studies provide reliable information on nature, extent and direction of selection.

Pathak and Dixit (1992) reported that the number of seeds pod^{-1} was positively and significantly correlated with length of fruiting nodes, length of pods, number of pods and days to maturity in sesame.

Interrelationship studies between characters across the one hundred accessions of sesame revealed positive and significant correlation between single plant seed yield (g) and characters such as number of seeds pod^{-1}, number of pods plant^{-1}, number of pods main stem^{-1} and days to maturity .This suggests that any increase in such traits will lead to improved single plant seed yield (Tomar *et al.*, 1999).

Parameshwarappa *et al.* (2009) also reported that seed yield plant^{-1} was significantly correlated with number of capsule plant^{-1} and plant height. However, negative and significant correlation was observed between 1000 seed weight with number of seeds pod^{-1} which indicates that increase in number of seed may produce low 1000 seed weight. Therefore a balance between these two important attributes should be maintained while breeding for high yield in sesame.

A significant and positive correlations recorded between growth characters *viz.* plant height, number of primary branches, number of leaves, crop dry weight, number of capsules, seeds per capsule, 1000 seed weight, harvest index and seed yield in sesame which suggested an interdependency between these characters as important yield determinants.(Muhamman *et al.*, 2010 and Haruna *et al.*, 2012). It was also established that yield and its components are polygenically inherited and

strongly affected by the environment as well as yet to be identified factors (Ibrahim and Khidir, 2012).

Correlation studies by Siva Prasad *et al.* (2013) revealed that knowledge of the nature of association between seed yield and its components can determine the appropriate characters to use in indirect selection for seed yield improvement in sesame.

Shabana *et al.* (2015) reported that the genotypic correlation coefficients were slightly higher than the phenotypic correlation coefficients in sesame. This indicated the masking effect of the environment was limited and did not mask the expression of the genotypes.

2.5.4 Path analysis

Path coefficient analysis has been widely used in breeding programmes to determine the nature of relationship between grain yield and its contributing components and to identify those components with significant effect on grain yield for potential use as a selection criteria (Pathak and Dixit ,1992).

According to Azeez and Morakinyo (2011) path analysis is a technique to determine the direct influence of one variable on another and is also used to separate the correlation coefficient into direct effect (path coefficient) and indirect effects (effects exerted through other independent variables).

In sesame seeds per capsule made the greatest direct percent contribution to seed yield. This could be attributed to the fact that most of the assimilates produced were translocated to the sink (capsule) which bears the seeds (Haruna *et al.*, 2012). Studies of Gelalcha and Hanchinal (2013) noted that the concept of path analysis also measures the relative importance of causal factors which provide information for effective selection during crop improvement programme.

In an experiment carried out to study the character association and path analysis in sesame, Lal *et al.* (2016) derived information about important agronomic traits determining seed yield per plant. These were number of pods per plant, number of pods on main stem, days to maturity and breadth of pods which had a positive direct effect with seed yield. They also found that these characters should be increased to achieve potential yield of sesame in high moisture and acidic soil conditions.

2.5.5 Genetic divergent studies

Genetic diversity plays an important role because hybrids between lines of diverse origin, generally display a greater heterosis than those between closely related parents. D^2 Statistics is one of the potent tools for measuring genetic divergence (Singh and Narayanan, 2013).

The quantitative assessment of genetic divergence was made by adopting Mahalanobis D^2 statistics for yield and its contributing characters. Rao (1952) suggested the application of this technique for the assessment of genetic diversity in plant breeding.

Alarmelu and Ramanathan (1998) reported that there was a wide range of variation in the cluster mean values for most of the characters under study in sesame. Therefore a crossing programme should be initiated between the genotypes belonging to different clusters. The greater the distance between two clusters, the wider the genetic diversity among the parents. The number of seeds per capsule contributed highest towards the divergence followed by number of capsules per plant. The remaining characters showed negligible contribution. But Velusami *et al.* (2008) reported that seed yield and 1000 seed weight contribute more towards the genetic divergence in sesame.

Tripathi *et al.* (2013) studied the germplasm of hundred black seeded sesame accessions and found that inter cluster distance was larger than intra

cluster distance suggesting wider diversity among the germplasm of different groups. The germplasm lines belonging to the distant clusters could be used in hybridization programme for obtaining a wider range of variability.

2.5.6 Selection index

Construction of a selection index is the best way of exploiting genetic correlation of dependant variable, say yield with several traits having high heritability. The mathematical genetic theory, in the form of selection index, developed by Smith (1936) is the basis for simultaneous selection of several traits. Hazel (1943) developed a simultaneous selection based on the approach of path analysis. The selection index technique can theoretically determine the genotypic worth of individuals or families in an objective manner (Subandi and Empig, 1973). A selection index most often aims at giving appropriate weight to the components maximizing gains from selection (Falconer, 1983).

Lee and Chang (1986) worked out the conventional selection index in sesame considering 14 traits including yield plant^{-1} using 82 cultivars, they stated that the highest genetic advance was for index that included all traits. As it is expensive and time consuming, he suggested only three characters to be included in the index (days to maturity, length of stem with capsules and capsules plant^{-1}) for future breeding programmes.

Materials and Methods

3. MATERIALS AND METHODS

The present study entitled "Evaluation of sesame genotypes for tolerance to waterlogging" was carried out in Onattukara Regional Agricultural Research Station, Kayamkulam during the year 2014-2016. Details of materials used and the methods followed for the study are presented below.

3.1 EXPERIMENTAL SITE

The project involving pot culture and field experiment was conducted at the instructional farm, Onattukara Regional Agricultural Research Station, Kayamkulam which is located at latitude 9^0 10'20''N longitude $76^0$30'00''E and an altitude of 3 m above mean sea level.

3.2 EXPERIMENTAL DESIGN

Experiment was conducted as pot culture and field experiment.

3.2.1 Pot culture experiment

Pot culture experiment was conducted during 2014-15 summer season in a Completely Randomized Design with thirty genotypes in three replications. Potting mixture used was sandy loam soil mixed with coir pith compost. Excess plants were thinned out and three plants per pot were maintained. Waterlogging was imposed twenty days after sowing in the pots. Duration of waterlogging was 24 hours, 48 hours and 72 hours and water level was maintained 2 cm above soil surface by replenishing frequently. After the treatment period, water was drained out from the pots and plants were allowed to grow to maturity. Out of thirty genotypes, ten were selected on the basis of survival percentage for conducting field experiment.

Plate 1.Waterlogging in pot culture experiment

Plate 2. Waterlogging in field experiment

3.2.2 Field experiment

The experiment was conducted during 2014-15 rabi season in a Randomized Block Design with ten genotypes in three replications. Experimental field was prepared to a fine tilth by tilling followed by leveling. Seeds were dibbled at a spacing of 30 cm between rows and 20 cm between plants. Excess plants were thinned out and a single plant was retained. Twenty days after sowing, waterlogging was imposed in the field for 72 hours. Control plot was maintained in separate row where the ten genotypes were maintained under normal condition. All the crop production practices as per the package of practices recommendation of Kerala Agricultural University (2011) were followed to raise a successful crop. Observations of five randomly selected plants of ten genotypes from each plot were recorded

3.3 EXPERIMENTAL MATERIALS

The seed material for the study consisted of thirty genotypes of sesame collected from the germplasam maintained at the Department of Plant Breeding and Genetics, Onattukara Regional Agricultural Research Station. The list of genotypes used in the present study is given in Table 1.

3.4 OBSERVATIONS RECORDED

Observations were recorded on individual plants of each replication in pot culture experiment. The characters studied were seedling survival percentage, plant height, days to 50% flowering, days to maturity, number of primary branches plant^{-1}, length of capsule, seeds capsule^{-1}, root length, root dry weight, number of roots plant^{-1}, 1000 seed weight and seed yield plant^{-1}.In the field experiment, five competitive plants were selected at random in each replication. The observations were recorded on survival percentage, plant height, days to 50% flowering, days to maturity, number of primary branches plant^{-1}, number of capsules plant^{-1}, length of capsule, number of seeds capsule^{-1},

Plate 3. General view of field

Table 1. List of Sesame genotypes used for evaluation

Sl No.	Name	Source	Sl No.	Name	Source
1	*Thilarani	ORARS, Kayamkulam	16	PT 2	ORARS, Kayamkulam
2	TC-289	ORARS, Kayamkulam	17	GT 4	ORARS, Kayamkulam
3	*Ayali	ORARS, Kayamkulam	18	*SV 2	ORARS, Kayamkulam
4	**Sesanum malabricum*	ORARS, Kayamkulam	19	JLT 408	ORARS, Kayamkulam
5	*Thilak	ORARS, Kayamkulam	20	*Rama	ORARS, Kayamkulam
6	AT 282	ORARS, Kayamkulam	21	JTS 8	ORARS, Kayamkulam
7	*TKG 308	ORARS, Kayamkulam	22	Subhra	ORARS, Kayamkulam
8	*GT 10	ORARS, Kayamkulam	23	TKG 21	ORARS, Kayamkulam
9	RT 369	ORARS, Kayamkulam	24	Surya	ORARS, Kayamkulam
10	JLS 606-7-2	ORARS, Kayamkulam	25	RT 362	ORARS, Kayamkulam
11	MT 10-8-1	ORARS, Kayamkulam	26	CUHY 57	ORARS, Kayamkulam
12	OSC 208	ORARS, Kayamkulam	27	OSC 560	ORARS, Kayamkulam
13	*TKG 22	ORARS, Kayamkulam	28	Thilathara	ORARS, Kayamkulam
14	JLS 9707-2	ORARS, Kayamkulam	29	*OSC 207	ORARS, Kayamkulam
15	DS 5	ORARS, Kayamkulam	30	DS 10	ORARS, Kayamkulam

*Genotypes selected for field experiment

root length, root dry weight, number of roots plant^{-1}, 1000 seed weight , seed yield plant^{-1}, seed yield plot^{-1} and oil content. Techniques adopted to record the observations are given below.

3.4.1 Seedling survival %

Number of plants of all the genotypes in each plot was recorded before and after flooding treatments. Seedling survival percent was calculated as,

$$\text{Seedling survival \%} = \frac{\text{Number of Seedlings Survived}}{\text{Total number of Seedlings}} \times 100$$

3.4.2 Days to 50% Flowering

Number of days taken for flowering in 50 per cent of the plants in each entry was recorded.

3.4.3 Plant height (cm)

Height of selected plants was measured from the soil surface to the top of the longest leaf by a graduated scale and recorded.

3.4.4 Days to Maturity

Days taken to mature 75 percent of capsules in 75 percent of plants within each plot were recorded as days to maturity.

3.4.5 Number of Primary Branches Plant^{-1}

Number of branches emerging from main stem of selected plants was counted at harvest and average was expressed as number of primary branches per plant.

3.4.6 Number of Capsules Plant^{-1}

Total number of seed bearing capsules on each selected plant including those on main stem and primary branches was counted and recorded.

3.4.7 Length of Capsules (cm)

Five matured capsules taken at random from the selected plants were measured and their mean length was recorded in centimeters.

3.4.8 Number of Seeds Capsule^{-1}

Seeds in five capsules of selected plant were counted and recorded after harvest.

3.4.9 Seed yield plant^{-1} (g)

Selected plants were uprooted, capsules were collected, uniformly dried, seeds were extracted and seed weight per plant was recorded in grams.

3.4.10 Seed yield plot^{-1} (g)

Plants from each plot were uprooted, dried uniformly, extracted seeds and seed weight was recorded in grams.

3.4.11 1000 seed weight (g)

One thousand randomly selected seeds of each genotype were weighed and recorded in grams.

3.4.12 Number of Roots Plant^{-1}

Whole roots including tap, lateral and adventitious roots of selected plants were counted and recorded.

3.4.13 Root length(cm)

Length of primary root was measured and recorded in centimeters

3.4.14 Root dry weight (g)

Roots of five randomly selected plants were collected, oven dried, weight was taken in an electronic balance and recorded in gram.

3.4.15 Number of Plants Plot^{-1}

Number of plants after treatments were counted and recorded.

3.4.16 Oil content (%)

30g seed of the ten genotypes was taken and oil content was estimated on the basis of pulsed nuclear magnetic resonance (NMR) signal of hydrogen in the liquid fraction. Mean value was recorded as the oil percentage.

3.4.17 Pest and disease incidence

Incidence of pests and disease were observed and recorded.

3.5 ANATOMICAL STUDY

Roots of the selected plants under study were collected from the experimental plot and were used for anatomical observations.

Mature adventitious roots were collected and washed thoroughly in tap water for anatomical studies. Very thin cross sections were taken using fresh stainless steel blade and kept in a watch glass for staining with safranin (2%). The cross sections were kept in the diluted stain for two minutes followed by washing in distilled water. These sections were observed under the Leica (10x35 X) microscope. Photographs of cross sections in the desirable fields were captured and analysed.

3.6 BIOCHEMICAL STUDY

3.6.1 Estimation of chlorophyll (DMSO method) (mg g^{-1})

Chlorophyll content of leaf samples were estimated as per the procedure described by Arnon (1949). A weighed quantity of leaf sample (0.5g) was taken from third fully expanded leaf and it was cut into small bits. These bits were put into test tubes and incubated overnight at room temperature with 10 ml DMSO: 80% acetone mixture (1:1 v/v). The coloured solution was transferred into a measuring cylinder and made up to 25 ml with the DMSO-acetone mixture. The absorbance was measured at 663 nm and 645 nm.

The chlorophyll content was calculated as mg/ using following formulae.

$$Chla = (12.7 \times A_{663} - 2.69 \times A_{645}) \times \frac{V}{1000} \times \frac{1}{freshweight}$$

$$Chlb = (22.9 \times A_{645} - 4.68 \times A_{663}) \times \frac{V}{1000} \times \frac{1}{freshweight}$$

A - Absorbance at specific wavelengths

V -Final volume of chlorophyll extract in 80% acetone

W- Fresh weight of tissue extracted.

3.6.2 Estimation of total soluble proteins (mg g^{-1})

Total soluble protein content of leaf samples was estimated by simple protein dye binding assay of Bradford (1976) using bovine serum albumin (BSA) as the standard. Hundred milligram of CBB 250 was dissolved in 50 ml of 95% ethanol and 100 ml of 85% (w/v) ortho phosphoric acid was added to this. The resulting solution was diluted to a final volume of 200 ml with distilled water. 0.1g leaf sample was taken from third fully opened leaves and ground to a thin paste and soluble protein was extracted with 10 ml of phosphate buffer (pH 7.8).

The extract was centrifuged at 5000 rpm for 10 minutes. A known volume (5ml) of diluted dye binding solution was added to 20µl of the supernatant. The solution was mixed well and allowed to develop a blue colour by keeping for at least 5 minutes but not longer than 30 minutes and the absorbance was measured at 596 nm. The protein content was calculated using the BSA standard in the range of (10-100µg). The protein content was expressed as mg/g FW.

3.6.3 Estimation of proline (mg g^{-1})

Acid ninhydrin method by Bates (1973) was done for the estimation of proline.. 0.5g fresh leaves were homogenized in10 ml of 3% aqueous sulphosalicylic acid followed by centrifuging at 6000 rpm for 15 minutes. 2 ml aliquot of supernatant was mixed with an equal amount of acetic acid and ninhydrin and heated in boiling water bath for 1hour. The reaction was terminated on ice bath and extracted with 4ml of toluene. The extract was vortexed for 20 seconds and the chromatophore containing toluene was used for reading absorbance at 520 nm with toluene as blank. A standard curve was drawn using concentration versus absorbance. The concentration of proline was determined from the graph and expressed as

Micro moles per gram tissue = [(micro gram proline / ml) × ml toluene) /115.5]× (5/g sample)

Where 115.5 is the molecular weight of proline.

3.6.4 Estimation of super oxide dismutase (SOD) (mg g^{-1})

Estimation of SOD was done as per the method described by Kakkar *et al.* (1984). 0.5 g sample was taken and ground with 3ml of potassium phosphate buffer, centrifuged at 2000 rpm for 10 minutes and the supernatants were used for the assay. The assay mixture contained 1.2ml of sodium pyrophosphate buffer, 0.1ml of PMS, 0.3 ml of NBT (Nitroblue tetrazolium salt), 0.2 ml of the enzyme preparation and water to make a total volume of 2.8ml.Reaction was initiated by

the addition of 0.2ml of NADH. The mixture was incubated at 30 ^{0} Celsius for 90 seconds and arrested by the addition of 1ml of glacial acetic acid. This mixture was then shaken with 4 ml of n-butanol, allowed to stand for 10 minutes and centrifuged. Intensity of chromogen in the butanol layer was measured in spectrophotometer at 560 nm. One unit of enzyme activity is defined as the amount of enzyme that gave 50% inhibition of NBT reduction in1 minute.

3.6.5 Estimation of phenol (mg g^{-1})

Estimation of phenols was done by Folin-Ciocalteau method (Mayr *et al.*, 1995). 0.5g of leaf samples was taken and reflexed in 10 ml 80% methanol for 20 min. The tissue was ground thoroughly in a mortar and pestle and filtered through a double layered cheese cloth. The filtrate was centrifuged at 1000 rpm for 10 min. The supernatant was collected and made to a known volume using 80% methanol. 0.1 ml aliquot was drawn to a test tube and made up to 3 ml using 80% methanol. To this, 0.5 ml of Folin- Ciocalteau reagent and 2 ml 20% Na_2CO_3 were added.

It was kept in a boiling water bath for 5 minutes till a white precipitate was formed and was then again centrifuged at 5000 rpm for 5 minutes. The absorbance of the clear supernatant was read at 650 nm against the blank. Standard curve was prepared using different concentrations of catechol and expressed in catechol equivalents as microgram per gram leaf tissue on fresh weight basis.

3.7 STATISTICAL ANALYSIS

The data recorded were processed using the following statistical procedures.

3.7.1 Analysis of variance in pot culture study

The biometrical observations recorded from pot culture experiment were subjected to ANOVA (Panse and Sukhatme, 1985) for comparison among various treatments and to estimate variance components.

ANOVA for each character

Sources of variation	Degrees of freedom	Mean sum of squares	F value calculated
Replication	r-1	MSR	MSE/MSR
Treatment	t-1	MST	MST
Error	(r-1) (t-1)	MSE	
Total	rt-1		

Where r - Number of replications

t- Number of treatments

MSR - mean square of replications

MST = mean square of treatments

MSE = mean square of error

Critical difference $CD = t_{\alpha}\sqrt{\frac{2MSE}{r}}$

Where t_{α} = Students t table value at error degree of freedom at α level of significance

3.7.2 Analysis of variance in field experiment

The method described in (3.7.1) was followed. Other statistical analysis includes estimation of genetic component of variance, correlation analysis, path coefficient analysis, genetic divergence analysis and selection index were done in field experiment.

3.7.3 Estimation of genetic parameters

3.7.3.1 Genetic components of variance

The phenotypic and genotypic variances were calculated by utilizing the respective mean square values (Johnson *et al.*, 1955).

i. Genotypic variance (VG)

$$VG = \frac{MST - MSE}{r}$$

ii. Environmental variance (VE)

VE =MSE

iii. Phenotypic variance (VP)

VP= VG+ VE

3.7.3.2 Coefficient of variation

The genotypic and phenotypic variation were calculated by Burton (1952).

i) Phenotypic coefficient of variation (PCV)

$$PCV = \frac{\sqrt{VP}}{\bar{X}} \times 100$$

ii) Genotypic coefficient of variation (GCV)

$$GCV = \frac{\sqrt{VG}}{\bar{X}} \times 100$$

$\bar{X}$= mean of characters

Categorization of the range of variation was effected as proposed by Shivasubramanian and Menon (1973).

Category	Range
Low	< 10%
Moderate	10-20%
High	>20%

h^2 = heritability

The range of genetic advance as percent of mean was classified as suggested by Johnson *et al.* (1955).

Category	Range
Low	<10%
Moderate	10-20%
High	>20%

3.7.4 Correlation analysis

Phenotypic, genotypic and environmental correlation coefficients were calculated using the respective variance and covariance of the characters which showed significant variation in the ANOVA.

Phenotypic correlation coefficients, $r_{PXY} = \frac{C_0V_P(X,Y)}{\sqrt{V_P(X)\times V_P(Y)}}$

Genotypic correlation coefficients, $r_{GXY} = \frac{C_0V_G(X,Y)}{\sqrt{V_G(X)\times V_G(Y)}}$

Where CoV_P (X, Y) - phenotypic covariance between two traits X and Y

CoV_G (X, Y) - genotypic covariance between two traits X and Y

Vp (X) and Vp (Y) - phenotypic variance for X and Y respectively

V_G (X) and V_G (Y) - genotypic variance for X and Y respectively

3.7.5 Path coefficient analysis

To study the cause and effect relationship of yield and its component characters, direct and indirect effects were analysed using path coefficient analysis as suggested by Wright (1954).

3.7.3.3 Heritability

Heritability percentage in broad sense was estimated for various characters as per the formulae suggested by Johnson *et al.* (1955).

Heritability, $h^2 = \frac{VG}{VP} \times 100$

VG = Genotypic variance

VP = Phenotypic variance

As suggested by Johnson *et al.* (1955) heritability in broad sense estimates were categorized as,

Category	Range
Low	0-30%
Moderate	30-60%
High	>60

3.7.3.4 Genetic advance

Genetic advance is the measure of genetic gain under selection which depends upon standardized selection differential, heritability and phenotypic standard deviation (Allard, 1960). The genetic advance was calculated in percent by formulae (Johnson *et al.*, 1955).

Genetic advance $(GA) = K \times h^2 \sqrt{v_p}$

GA as percentage of mean $= \frac{GA}{\bar{X}} \times 100$

Where K = standardized selection differential (2.06 at 5% selection intensity)

The genotypic correlation between yield and selected component characters were subjected to path analysis and the direct effect of the character on yield as well as the indirect effect through other characters were estimated.

3.7.6 Genetic divergence analysis

Genetic divergence was measured using the technique D^2 statistics developed by Mahalanobis in 1928. Grouping of genotypes into clusters was done based on the relative distance (D^2) from each other, and was based on the method suggested by Tocher (Rao, 1952).

3.7.7 Selection index

A better way of exploiting genetic correlation with several traits having high heritability is to construct an index, called selection index, which was first proposed by Smith in 1936. Selection index combines information on all the characters associated with the dependent variable, say yield. Later on in 1943, Hazel developed a simultaneous selection model based on the approach of path analysis.

Results

4. RESULTS

The experiment entitled "Evaluation of sesame genotypes for tolerance to waterlogging" was conducted at Onattukara Regional Agricultural Research station, Kayamkulam. Preliminary screening for excess moisture was done as pot culture experiment with thirty genotypes in three replications. Based on this study, ten genotypes were selected. These ten genotypes were further evaluated in the field and observations were recorded on sixteen characters. Statistical analysis of the data was conducted and results are presented in this chapter.

4.1 POT CULTURE EXPERIMENT

Preliminary screening of thirty genotypes for tolerance to waterlogging was conducted during summer 2014-15. Waterlogging was imposed at three different duration ie. 24 hours, 48 hours and 72 hours. After the treatment, water was drained out and survival percentage was recorded (Table 2).

All the thirty genotypes survived 24 hours and 48 hours of waterlogging. Survival percentage varied from 12.2 (CUHY-57) to 100 percentage (*S.malabaricum*) under 24 h of waterlogging. The same genotypes recorded lowest (3.4%) and highest survival (100 %) under 48 h of waterlogging also. But seventeen numbers survived up to 72 hours of waterlogging and maximum survival was recorded by the wild species *Sesamum malabaricum*. Among the cultivated genotypes, maximum survival was recorded by Ayali (88.6%) and minimum by AT 282 (3.2%).

4.1.1 Performance of Sesame Genotypes in the Pot

Mean performance of different characters of the surviving 17 genotypes were studied. (Table 3). Maximum per plant yield was recorded by Thilak (4.24 g) followed by Ayali (4.21 g).

Table 2.Survival percentage of thirty genotypes after treatments

	Genotype	Period of waterlogging (h)							
Sl.No.		24	48	72	Sl.No.		24	48	72
1	Thilarani	85.4	55.3	45.2	16	PT 2	16.1	6.1	0.0
2	TC 289	14.8	4.8	0. 0	17	SV2	80.2	50.2	40.2
3	Ayali	96.4	60.3	50.1	18	GT 4	19.5	9.9	0.0
4	*Sesamum malabricum*	100.0	100.0	100.0	19	JLT 408	15.6	5.9	0.0
5	Thilak	83.4	52.0	42.4	20	Rama	92.8	59.4	49.2
6	AT 282	23.2	10.8	3.2	21	JTS 8	16.8	6.9	0.0
7	TKG 308	85.2	55.3	45.2	22	Subhra	26.8	12.6	5.2
8	GT 10	84.3	54.3	44.2	23	TKG 21	31.1	13.5	7.3
9	RT 369	24.4	11.7	4. 3	24	Surya	13.6	4.1	0.0
10	JLS 606-7-2	29.5	13.1	6.2	25	RT 362	17.1	7.1	0.0
11	MT 10-8-1	18.8	8.9	0.0	26	CUHY 57	12.2	3.4	0.0
12	OSC 208	34.4	14.8	9.4	27	OSC 560	17.7	7.8	0.0
13	TKG 22	81.6	51.6	41.4	28	OSC 207	90.6	58.6	48.6
14	JLS 9707-2	15.1	5.1	0.0	29	Thilathara	33.2	14.1	8.6
15	DS 5	18.1	8.1	0.0	30	DS 10	19.1	9.1	0.0

Table 3. Mean performance of survived sesame genotypes at different durations of waterlogging treatment

Sl.No.	Genotype	S.S.P			D.F.F			P.H (cm)			D.M			N.P.B		
		24 h	48 h	72 h	24 h	48 h	72 h	24 h	48 h	72 h	24 h	48 h	72 h	24 h	48 h	72 h
1	Thilarani	85.4	55.3	45.2	34.8	37.76	40.23	48.28	46.17	44.16	86.16	86.23	88.32	5.26	4.86	4.46
2	Ayali	96.4	60.3	50.1	38.52	40.13	42.24	52.37	48.72	45.13	84.39	86.41	88.69	5.53	5.12	4.75
3	*S.malabaricum*	100	100	100	31.43	33.36	35.16	36.95	34.53	32.62	126.68	128.47	131.3	4.96	4.49	4.43
4	Thilak	83.4	52	42.4	36.6	38.49	40.17	50.15	48.47	46.22	85.52	87.33	89.23	5.44	5.02	4.81
5	RT 369	23.2	10.8	3.2	33.51	35.67	37.41	31.2	27.36	24.39	94.25	96.47	98.43	4.48	4.39	4.23
6	TKG308	85.2	55.3	45.2	34.11	36.52	38.73	44.75	42.19	40.63	91.24	93.35	95.41	4.84	4.47	4.12
7	GT 10	84.3	54.3	44.2	30.41	32.51	34.69	34.9	32.86	30.87	90.53	92.4	93.51	4.25	4.22	4.1
8	JLS 606-2-2	24.4	11.7	4. 3	31.02	33.12	35.14	35.36	33.2	31.14	92.37	93.57	94.55	4.13	4.24	4.13
9	MT 10-8-1	29.5	13.1	6.2	29.66	31.51	33.12	34.37	32.82	30.63	92.46	93.05	94.86	4.14	4.1	3.98
10	RT 362	34.4	14.8	9.4	31.26	32.25	35.46	35.89	33.69	31.55	89.03	91.23	92.46	4.12	4.41	4.25
11	TKG 22	81.6	51.6	41.4	33.84	35.54	37.54	42.25	40.36	29.79	91.43	94.1	100.23	4.61	4.29	3.83
12	SV 2	80.2	50.2	40.2	29.34	31.28	33.03	33.82	31.78	30.66	94.03	94.03	95.6	4.16	4.08	3.84
13	Rama	92.8	59.4	49.2	28.41	30.22	32.03	32.9	30.81	28.94	89.33	90.63	91.65	3.6	3.72	3.64
14	JLS 9707-2	26.8	12.6	5.2	32.46	34.27	36.87	41.63	35.7	33.16	96.52	98.12	100.24	4.43	4.04	3.62
15	JLT 408	31.1	13.5	7.3	28.62	30.65	32.93	33.26	32.4	29.46	94.6	95.3	96.4	4.11	4.04	3.94
16	OSC207	90.6	58.6	48.6	34.7	36.64	38.43	47.26	45.49	43.66	87.8	89.49	91.03	5.14	4.77	4.41
17	PT 2	33.2	14.1	8.6	34.48	36.63	38.74	45.26	43.57	41.86	90.96	92.6	100.53	5.03	4.66	4.25
	C.D	2.72	1.52	1.87	1.37	1.49	1.37	1.64	1.70	1.67	3.75	3.95	3.43	0.18	0.15	0.17

S.S.P – Seedling survival %

DFF - Day's to fifty percentage flowering

DM - Days to maturity

RL - Root length

NC - Number of capsules plant^{-1}

S.P.C - Seeds capsule $^{-1}$

PH - Plant height

NPB - Number of primary branches plant^{-1}

NR - Number of roots plant^{-1}

RDW - Root dry weight

LC - Length of capsule

TSW - Thousand seed weight

SYP- Seed Yield plant^{-1}

Sl.No.	Genotype	N.C			L.C (cm)			N.S.C			R.L(cm)			R.D.W(g)		
		24 h	48 h	72 h	24 h	48 h	72 h	24 h	48 h	72 h	24 h	48 h	72 h	24 h	48 h	72 h
1	Thilarani	26.87	26.02	25.64	1.95	1.86	1.8	40.23	35.43	32.1	10.52	10.23	10.17	6.86	6.84	6.8
2	Ayali	27.49	27.03	26.72	2.17	2.16	2.1	42.66	41.06	38.53	12.28	11.85	11.81	6.87	6.82	6.81
3	*S.malabaricum*	22.48	22.17	21.91	2.36	2.33	2.30	32.38	30.45	28.45	7.23	6.85	6.42	7.03	6.92	6.81
4	Thilak	25.6	25.37	24.52	2.02	1.94	1.88	43.23	41.25	39.42	9.66	9.25	9.21	6.76	6.83	6.74
5	RT 369	23.21	22.91	22.71	1.75	1.69	1.63	35.15	33.56	31.69	8.52	8.14	7.74	6.77	6.73	6.7
6	TKG 308	24	23.74	23.45	1.47	1.32	1.1	36.18	34.06	32.36	9.47	9.14	8.83	6.78	6.65	6.57
7	GT 10	21.85	21.81	21.51	1.48	1.38	1.37	32.18	30.14	28.26	6.93	6.83	6.64	6.48	6.4	6.34
8	JLS 606-2-2	22.07	21.74	21.65	1.44	1.35	1.34	32.4	30.15	28.14	7.11	7.07	6.73	6.63	6.63	6.4
9	MT 10-8-1	21.73	21.66	21.6	1.46	1.39	1.35	30.65	28.69	26.4	7.02	6.72	6.42	6.45	6.34	6.3
10	RT 362	22.15	22.04	21.98	1.45	1.4	1.35	32.33	30.16	28.13	7.21	7.19	7.18	6.64	6.59	6.52
11	TKG 22	23.76	23.39	22.79	1.55	1.39	1.2	35.32	33.58	31.3	9.33	9.24	9.02	6.53	6.69	6.35
12	SV 2	21.63	21.6	21.57	1.47	1.4	1.35	30.5	28.66	26.46	6.61	6.31	6.21	6.41	6.33	6.25
13	Rama	21.4	21.54	21.52	1.52	1.54	1.59	28.73	30.04	28.33	6.41	6.21	6.14	6.34	6.24	6.65
14	JLS 9707-2	23.22	22.83	22.25	1.31	1.32	1.35	34.16	32.56	30.3	8.14	7.64	7.21	6.63	6.61	6.56
15	JLT 408	21.57	21.53	21.42	1.45	1.38	1.32	30.12	28.64	26.14	6.52	6.23	6.12	6.26	6.26	6.2
16	OSC 207	26.21	24.73	24.31	1.87	1.82	1.74	40.23	37.2	34.13	10.66	10.05	9.13	6.74	6.7	6.17
17	PT 2	25.54	23.7	21.84	1.65	1.61	1.35	38.45	36.45	34.32	11.47	11.42	11.37	6.98	6.92	6.64
	C.D	0.94	1.04	0.90	0.07	0.07	0.05	1.68	1.80	1.23	0.30	0.41	0.33	0.29	0.23	0.30

Sl.No.	Genotype	N.R			T.S.W (g)			S.Y.P (g)		
		24 h	48 h	72 h	24 h	48 h	72 h	24 h	48 h	72 h
1	Thilarani	27.03	30.43	34.46	2.55	2.54	2.52	4.37	4.28	4.11
2	Ayali	34.26	28.3	37.2	2.3	2.29	2.27	4.49	4.34	4.24
3	*S.malabaricum*	38.3	42.8	31.23	2.21	1.96	1.86	3.52	3.46	2.22
4	Thilak	34.53	32.3	27.23	2.46	2.45	2.43	4.42	4.31	4.21
5	RT 369	28	32.4	22.4	2.92	2.73	2.57	3.86	3.74	3.65
6	TKG 308	25.6	33.4	19.23	2.47	2.72	2.66	4.04	3.95	3.86
7	GT 10	23.4	27.16	21.2	2.61	2.59	2.57	2.82	2.76	2.65
8	JLS 606-2-2	24.6	30.26	17.2	2.63	2.61	2.59	2.97	2.82	2.75
9	MT 10-8-1	16.26	15.45	16.36	2.6	2.59	2.57	2.63	2.53	2.48
10	RT 362	15.36	14.43	15.9	2.64	2.62	2.57	3.02	2.84	2.57
11	TKG 22	31.3	34.43	25.63	2.87	2.85	2.74	3.96	3.85	3.75
12	SV 2	14.5	13.47	15.5	2.58	2.57	2.56	2.84	2.45	2.32
13	Rama	13.52	14.5	14.2	2.57	2.56	2.54	2.36	2.34	2.21
14	JLS 9707-2	23.1	27.36	30.4	3.02	2.97	2.93	3.74	3.59	3.53
15	JLT 408	13.21	13.66	13.33	2.57	2.56	2.55	2.46	2.32	2.16
16	OSC 207	26.83	31.4	36.36	2.63	2.46	2.43	4.92	4.12	4.01
17	PT 2	35.36	39.36	35.9	2.71	2.62	2.51	4.15	4.07	3.95
	C.D	1.10	1.03	1.22	0.10	0.09	0.10	0.15	0.13	0.16

4.1.2 Analysis of Variance for Yield and Yield attributes

Analysis of variance was done and presented in Table 4. The analysis of variance revealed that the genotypes recorded highly significant differences at 1% level of significance in all the three durations of waterlogging viz. 24 hours, 48 hours and 72 hours with respect to seedling survival percentage, plant height, days to 50% flowering, days to maturity, number of primary branches plant^{-1}, number of capsules plant^{-1}, seeds capsule^{-1}, root length, root dry weight, number of roots plant^{-1}, and seed yield plant^{-1}. Length of capsules and 1000 seed weight showed significant difference at 5% level of significance.

Selection of genotypes field screening was done based on the seedling survival percentage under waterlogged condition. Ten genotypes which recorded highest survival percentage at 72 hours of waterlogging were selected for field evaluation.

4.2 FIELD EXPERIMENT

Ten genotypes selected from the pot culture experiment were evaluated in the uplands at ORARS, Kayamkulam during 2015-16 rabi season (Plate.3). Observations were recorded and data were analysed.

4.2.1 Performance of Sesame Genotypes in the Field

Mean performances of ten genotypes for sixteen characters were studied and mean for different characters are presented in Table 5.

4.2.1.1 Seedling Survival %

All the genotypes differed significantly in seedling survival percentage. The wild genotype *Sesamum malabaricum* had the highest survival percentage (97.6 %) and it was significantly different from all other genotypes. The genotype Ayali recorded the maximum (88.61%) and SV 2 recorded the minimum seedling survival percentage (55.2 %). Five genotypes recorded survival percentage above

Table 4. Analysis of variance for yield and yield attributes of pot culture experiment

Characters	Mean sum of squares					
	Genotype	Error	Genotype	Error	Genotype	Error
	24 h	24 h	48 h	48 h	72 h	72 h
Survival %	2786.837**	2.69	2047.928**	0.844	2049.358**	1.272
Days to 50% flowering	24.689**	0.687	25.98**	0.815	26.281**	0.69
Plant height(cm)	141.025**	0.979	139.998**	1.054	171.618**	1.02
Days to maturity	262.643**	5.126	266.153**	5.681	286.247**	4.27
No. of primary branches plant^{-1}	0.925**	0.013	0.434**	0.009	0.352**	0.011
No. of capsules plant^{-1}	12.37**	0.323	8.887**	0.393	7.738**	0.295
Length of capsule (cm)	0.166*	0.02	0.19*	0.02	0.209*	0.01
No. of seeds capsule $^{-1}$	61.171**	1.029	49.955**	1.189	47.831**	0.556
Root length (cm)	10.625**	0.034	10.156**	0.061	10.324**	0.04
Root dry weight (g)	0.147**	0.032	0.154**	0.02	0.163**	0.034
No. of roots plant^{-1}	201.375**	0.446	263.969**	0.387	186.045**	0.549
1000 seed weight (g)	0.122*	0.04	0.145*	0.03	0.144*	0.04
Seed yield plant^{-1} (g)	1.911**	0.09	1.71**	0.07	1.755**	0.01

* Significant at 5% level

** Significant at 1% level

Table 5. Mean performance of sesame genotypes in field experiment

Genotype	S.S (%)	C	DFF	C	PH (cm)	C	DM	C	NPB	C	NC	C	LC (cm)	C	SPC	C
Rama	81.1	100	40.1	38.6	123.4	132.5	92.4	91.5	7.4	7.6	69.3	80	2.4	2.43	37.00	38
Thilak	65.5	100	42.0	40.3	128.1	139.2	89.1	87.8	7.8	8.2	80.0	86	2.51	2.54	40.00	42
OSC 207	78.1	100	39.5	36.5	119.8	132.1	93.1	91.6	7.5	7.8	66.0	74	2.36	2.38	36.00	38
S.malabaricum	97.6	100	46.6	45.3	101.4	111.3	134.5	132.5	5	5.5	41.3	52	2.94	2.96	26.66	28
SV2	55.2	100	36.2	34.3	103.2	110.3	100.1	96.2	5.7	5.9	46.6	56	2.15	2.16	30.00	32
TKG 308	72.0	100	37.2	35.8	109.7	114.5	98.3	95.7	6.7	7	51.3	60	2.25	2.28	31.00	34
GT 10	68.3	100	38.6	37.2	118.4	125.3	94.2	92.2	7	7.4	65.0	76	2.34	2.36	33.00	34
TKG 22	62.6	100	38.0	36.6	116.5	129.2	95.1	93.4	6.6	6.5	62.0	70	2.32	2.34	32.00	33
Thilarani	74.6	100	41.3	39.7	125.4	130.5	90.2	86.8	7.6	8.1	74.6	85	2.47	2.5	38.00	40
Ayali	88.6	100	43.3	40.2	131.7	136.8	90.1	85.4	8	8.3	97.6	102	2.72	2.74	42.00	44
Mean	74.3	-	40.3	-	111.7	-	97.5	-	6.93	-	65.3	-	2.36	-	34.5	-
C.D	2.08	-	0.92	-	6.32	-	1.18	-	1.15	-	6.76	-	0.004	-	0.62	-

S.S – Seedling survival %

C.D - Critical difference

PH- Plant height

NPB- Number of primary branches plant $^{-1}$

LC- Length of capsule

NR- Number of roots plant $^{-1}$

C- Control

RDW- Root dry weight

TSW- Thousand seed weight

YP- Yield plot $^{-1}$

SPC- Seed capsule $^{-1}$

DFF- Day's to fifty percentage flowering

DM- Days to maturity

NC- Number of capsules

OC- Oil content

OC- Oil content

RL- Root length

PP- Plants plot $^{-1}$

SYP- Yield plant $^{-1}$

Genotype	TSW (g)	C	NR	C	RL (cm)	C	RDW (g)	C	PP	C	SYP (g)	C	YP (g)	C	OC (%)
Rama	2.64	2.66	37.8	36.2	10.12	11.12	7.25	7.23	24.6	30	6.12	6.72	153.9	201.6	41.3
Thilak	2.55	2.57	41.5	40.2	11.63	12.13	7.31	7.27	20.0	30	7.30	7.84	145.9	235.2	48.6
OSC 207	2.75	2.76	37.2	35.3	9.73	10.21	7.15	7.12	23.6	30	5.61	6.13	131.3	183.9	48.1
S.malabaricum	2.41	3.13	46.4	45.2	6.4	7.14	7.39	7.35	29.3	30	2.92	3.45	85.38	103.5	32.5
SV2	3.01	3.08	34.5	32.3	7.30	8.13	7.0	6.95	17.3	30	3.65	4.1	63.03	123	47.6
TKG 308	2.92	2.98	35.8	33.2	7.81	8.42	7.02	7.0	22.0	30	4.23	4.1	93.20	123	40.9
GT 10	2.80	2.86	30.6	27.2	9.23	10.8	7.11	7.09	20.6	30	5.10	5.8	105.4	174	40.1
TKG 22	2.84	2.92	27.1	35.3	8.590	9.12	7.05	6.98	18.6	30	4.63	5.1	87.03	153	44.4
Thilarani	2.61	2.66	39.4	36.5	10.53	10.62	7.27	7.21	22.6	30	6.57	7.5	143.8	225	47.8
Ayali	2.52	2.55	43.3	40.1	12.20	13.2	7.37	7.31	27.0	30	7.46	8.13	201.4	243.9	45.4
Mean	2.70	-	37.36		9.36	-	7.19	-	22.6	-	5.36	-	123.06	-	46.67
C.D	0.09	-	5.99	-	0.57	-	0.03	-	1.02	-	0.39	-	14.98	-	0.58

the mean value (74.36%). Waterlogging was not imposed in the control plots and all the plants in these plots survived (100%).

4.2.1.2 Days to Fifty Percentage Flowering

Genotypes differed significantly for days to fifty percentage flowering. The wild species, *Sesamum malabaricum* recorded the maximum days to 50% flowering (46.6 days). Among the cultivated genotypes, SV 2 recorded minimum days for 50 % flowering (36.2 days) and it is significantly different from all other genotypes. Ayali recorded maximum days (43.3 days). General mean for this character was 40.31 days and four genotypes took more days than this. Control plants required lesser number of days for fifty percent flowering than treatments.

4.2.1.3 Plant Height (cm)

Plant height significantly differed among all the genotypes. Wild species *Sesamum malabaricum* recorded lowest plant height (101.4 cm).

Among *Sesamum indicum* genotypes, highest plant height was recorded by Ayali (131.7cm) and the two genotypes (Thilak and Thilarani) were on par with it. Lowest plant height was recorded for SV 2 (103.2 cm). Six genotypes had plant height above mean value (117.76 cm). Control plant recorded higher plant height than treatments.

4.2.1.4 Days to Maturity

Genotypes differed significantly high for days to maturity. Maximum duration was recorded by the wild species *Sesamum malabaricum* (134.5days) and among the cultivated genotypes maximum days to maturity was for SV 2 (100.1 days) and minimum was recorded by Thiiak (89.1 days). Two genotypes (Thilarani and Ayali) were on par with Thilak. Three genotypes viz. *S.malabaricum*, SV 2 and TKG 308 had above mean value for days to maturity (97.5 days). Control plants required lesser number of days to mature than treatments.

4.2.1.5 Number of Primary Branches Plant $^{-1}$

Highly significant difference was observed among all the genotypes for number of primary branches per plant. Among *Sesamum indicum* genotypes highest number of primary branches was recorded by Ayali (8) and five genotypes (Rama, Thilak, OSC 207, GT 10 and Thilarani) were on par with it. Lowest number of primary branches was for SV 2 (5.7).

The wild species *Sesamum malabaricum* recorded a lower value than that of SV 2 (5). Among ten genotypes six had number of primary branches plant^{-1} above mean value (6.93). Control plants recorded more number of primary branches than treatments.

4.2.1.6 Number of Capsules Plant $^{-1}$

Genotypes differed significantly for number of capsules plant^{-1}. Among *Sesamum indicum* genotypes highest number of capsules plant^{-1} was recorded by Ayali (97.6) and it was significantly different from all other genotypes. It was lowest for SV 2 (46.6).

The wild species *Sesamum malabaricum* recorded a lower value than that of SV 2 (41.3). Five genotypes had above general mean (65.3) and control plants had higher number of capsules than treatments.

4.2.1.7 Length of capsule (cm)

Significant difference was observed among all the genotypes for length of capsule, highest value recorded by Ayali (2.72 cm) and lowest by SV 2 (2.15 cm).

Sesamum malabaricum recorded the highest capsule length (2.94cm) and it is significantly different from all other genotypes. Five genotypes had length above general mean (2.36 cm) and control plants had slightly higher value than treatments.

4.2.1.8 Number of Seeds Capsule $^{-1}$

Genotypes differed significantly for number of seeds capsule^{-1}. Among the *Sesamum indicum* genotypes highest number of seeds per capsule was recorded by Ayali (42) and it is significantly different from all other genotypes. SV 2 recorded lowest number of seeds capsule^{-1} (30.0).

Wild species *Sesamum malabaricum* recorded the lowest number of seeds per capsule (26.66). Five genotypes had above general mean (34.56) and control plants had higher number of seeds per capsules than treated plants.

4.2.1.9 Thousand Seed Weight (g)

Significant difference was observed among all the genotypes for thousand seed weight. Wild species *Sesamum malabaricum* recorded lowest thousand seed weight (2.41 g). Among the *Sesamum indicum* genotypes highest 1000 seed weight was recorded by SV 2 (3.01 g) which was on par with TKG 308. 1000 seed weight lowest for Ayali (2.52 g). Five genotypes had above general mean (2.70 g) and thousand seed weight was more for control plants.

4.2.1.10 Number of Roots Plant $^{-1}$

Genotypes differed significantly for number of roots plant^{-1}. Among the *Sesamum indicum* genotypes highest number of roots was recorded by Ayali (43.3) and lowest by TKG 22 (27.1). *Sesamum malabaricum* had highest number of roots (46.4) and it was on par with four genotypes (Ayali, Thilarani, Rama and Thilak).

Number of roots plant^{-1} was less in control plants. Significant formations of adventitious roots were observed in *Sesamum malabaricum* on waterlogging (Plate 4).

(A) (B)

Plate 4 . A) Formation of adventitious roots after waterlogging in *Sesamum malabaricum* B) Control

4.2.1.11 Root Length (cm)

Significantly high difference was observed for length of root. Among the *Sesamum indicum* genotypes root length was highest for Ayali (12.2 cm) and it was on par with Thilak. Root length was lowest for SV 2 (7.30 cm).

Wild species *Sesamum malabaricum* had lowest root length (6.4 cm). Five genotypes recorded higher value than general mean (9.36 cm) and root length was more for control plants.

4.2.1.12 Root Dry Weight (g)

Wild species *Sesamum malabaricum* had highest root dry weight (7.39 g) and it was significantly different from all other genotypes. Five genotypes had above general mean value (7.19 g) and treated plants had more root dry weight than control.

4.2.1.13 Number of Plants Plot $^{-1}$

Significant difference was observed among all the genotypes for plants plot^{1} and was highest for wild species *Sesamum malabaricum* (29.3) and it was significantly different from all other genotypes.

Among the *Sesamum indicum* genotypes number of plants plot^{-1} was highest for Ayali (27) and lowest for SV 2 (17.3). Five genotypes have values above general mean (22.6) and control plots had more number of plants.

4.2.1.14 Yield Plant $^{-1}$ (g)

Genotypes differed significantly for yield per plant. Among cultivated species highest yield plant^{-1} was for Ayali (7.46 g) which was on par with Thilak. Lowest yield plant^{-1} was for SV 2 (3.65 g). Wild species *Sesamum malabaricum* recorded lowest yield (2.92 g). Five genotypes recorded yield plant $^{-1}$ above general mean (5.36 g) and control plants recorded more yield than treated plants.

4.2.1.15 Yield Plot $^{-1}$ (g)

Significant difference was observed among all the genotypes for yield plot^{-1}. Among cultivated species highest yield plot^{-1} was for Ayali (201.4g) and lowest for SV 2 (63.03 g). Wild species *Sesamum malabaricum* recorded yield plot^{-1} of 85.38 g. Five genotypes recorded higher yield plot^{-1} than general mean (123.06 g) and control plots had more yield per plot than treatments.

4.2.1.16 Oil content (%)

Genotypes differed significantly for oil content. Among *Sesamum indicum* genotypes highest oil content was recorded by Thilak (48.6%) and OSC 207 was on par with it. Oil content was lowest by GT 10 (40.1 %). *Sesamum malabaricum* recorded the lowest oil content (30.2%). Six genotypes had oil content above general mean value (43.67%).

4.2.1.17 Incidence of Pest and Disease

Mild damage of capsules by parrot was observed in all the genotypes under study. No other pest attack was observed. Mild incidence of phyllody was observed in Thilak and Thilarani (Plate 5). The crop was not affected by any other diseases.

4.2.2 Analysis of Variance for Yield and Yield Attributes

Selected ten genotypes were evaluated under field condition. Waterloggging was imposed 20 days after sowing for 72 h and observations on sixteen characters were recorded. Analysis of variance was done and presented in Table 6.

The analysis of variance revealed that the genotypes differed significantly from each other with respect to seedling survival percentage, plant height, days to 50% flowering, days to maturity, number of primary branches plant^{-1}, length of capsule, seeds capsule^{-1}, root length, root dry weight, number of roots plant^{-1},

(A)

Plate 5. A) Incidence of Phyllody

Table 6. Analysis of variance for various characters of sesame genotypes in field experiment

Characters	Mean sum of square		
	Replications	Genotypes	Error
Seedling survival %	4.73	475.36**	2.84
Days to 50% flowering	0.62	612.5**	0.421
Plant height (cm)	29.18	314.36**	13.60
Days to maturity	0.22	549.32**	0.505
No. of primary branches plant^{-1}	1.63	18.96**	0.81
No. of capsules plant^{-1}	30.10	831.91**	15.54
Length of capsule (cm)	0.00070	0.091*	0.004
Seeds capsule $^{-1}$	0.133	69.85**	0.132
1000 seed weight(g)	0.0035	0.119*	0.003
No. of roots plant^{-1}	14.68	99.62**	12.27
Length of root(cm)	0.0498	10.27**	0.11
Root dry weight(g)	0.00056	0.070**	0.0005
Plants plot $^{-1}$	0.100	41.39**	0.359
Yield plant^{-1} (g)	0.066	7.01**	0.051
Yield plot $^{-1}$ (g)	118.67	46811.42**	1373.5
Oil content (%)	0.013	77.026**	0.117

* Significant at 5% level

** Significant at 1% level

number of plants plot^{-1}, 1000 seed weight, seed yield plant^{-1}, seed yield plot^{-1} and oil content.

4.2.3 Anatomical study.

One of the adaptive mechanisms for waterlogging tolerance is the formation of aerenchymatic tissue in roots. Roots were collected after the waterlogging treatment. Anatomical studies of the roots of ten genotypes were conducted.

Significant formation of aerenchyma tissue was observed in the three genotypes viz. *Sesamum malabaricum*, Ayali and OSC 207 (Plate 6, 7, 8 respectively).

4.2.4 Biochemical study

Leaves were collected after the waterlogging treatment and estimated chlorophyll, total soluble protein, proline, super oxide dismutase and phenol. Result of the biochemical studies are presented below.

4.2.4.1 Chlorophyll content (mg g^{-1})

Waterlogging reduced the chlorophyll content in all the genotypes. Since t-statistic for chlorophyll content (7.21) after waterlogging was more than table t value (2.26), there was significant difference between waterlogging treatment and control for chlorophyll content. Total chlorophyll, chlorophyll a and chlorophyll b content were highest in Ayali (0.56 mg g^{-1}) and lowest in SV 2 (0.20 mg g^{-1}). Total chlorophyll, chlorophyll a and chlorophyll b content of sesame leaves were compared and presented in Table 7.

4.2.4.2 Total soluble protein (mg g^{-1})

Total soluble protein (TSP) content after waterlogging was compared with control and presented in Table 8 and it was found to be decreased by waterlogging. Since t-statistic for TSP content (10.35) after waterlogging

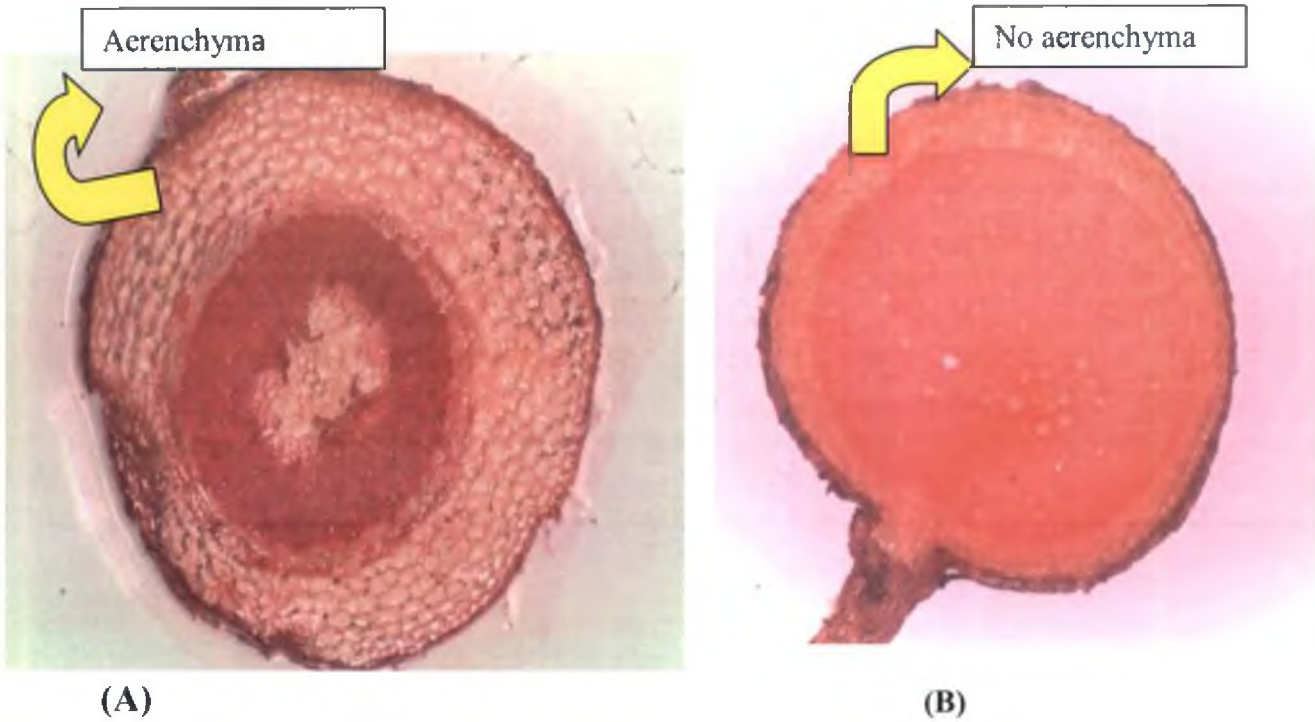


Plate 6 . A) Cross section of roots of *Sesamum malabaricum* after waterlogging B) Control

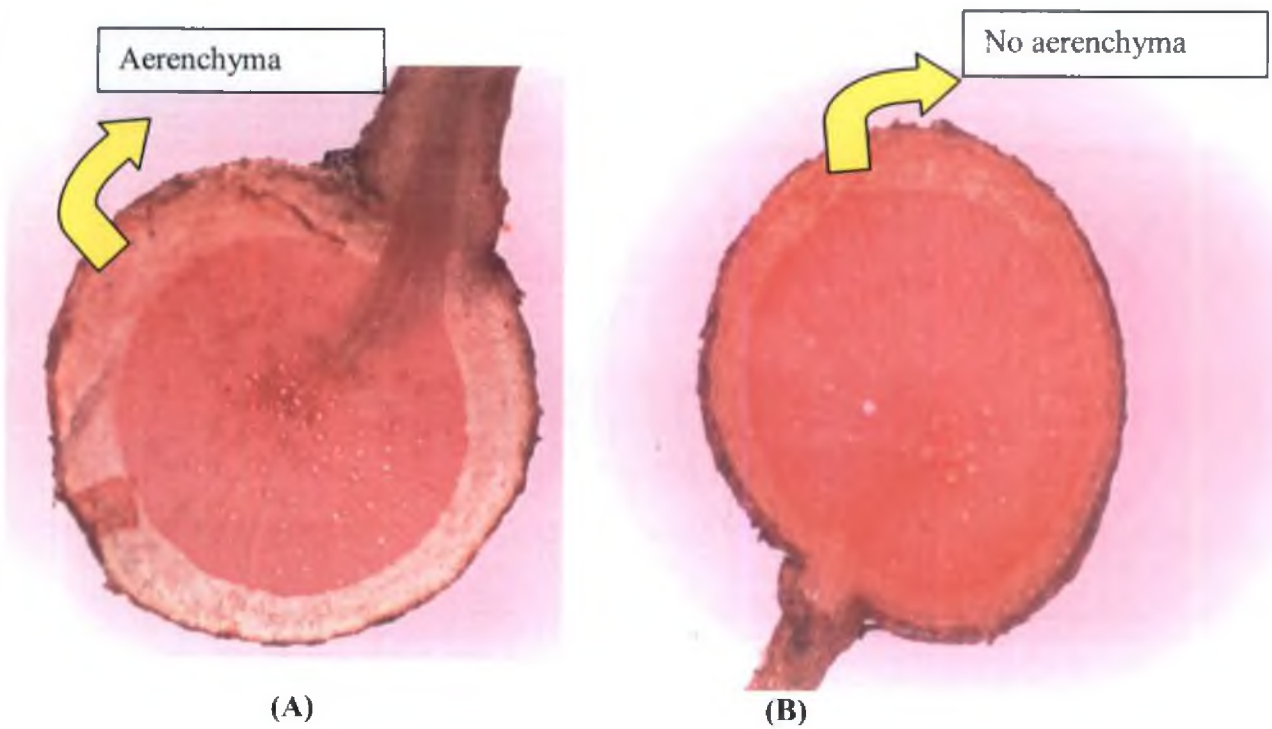


Plate 7. A) Cross section of roots of Ayali after waterlogging B) Control

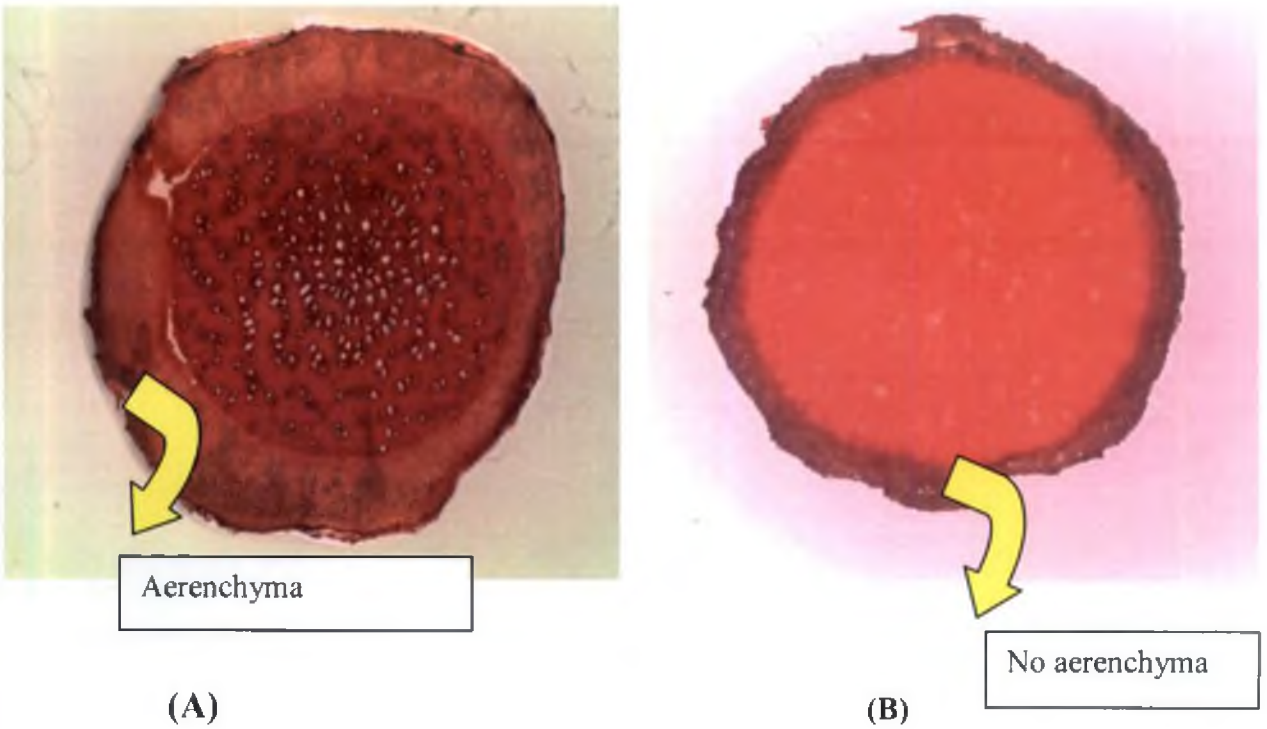


Plate 8. A) Cross section of roots of OSC 207 after waterlogging B) Control

Table 7. Chlorophyll content in sesame leaves after waterlogging (mg g^{-1})

Genotype	Chl a		Chl b		Total	
	After stress	Control	After stress	Control	After stress	Control
Rama	0.31	0.50	0.18	0.30	0.49	0.80
TKG 308	0.28	0.54	0.17	0.32	0.45	0.87
SV2	0.14	0.46	0.13	0.29	0.20	0.76
GT 10	0.29	0.70	0.14	0.42	0.44	1.13
S.malabaricum	0.33	0.62	0.19	0.43	0.53	1.06
Thilarani	0.34	0.41	0.19	0.39	0.54	0.80
OSC 207	0.31	0.41	0.16	0.23	0.48	0.65
TKG 22	0.25	0.32	0.14	0.25	0.40	0.58
Ayali	0.36	0.57	0.20	0.34	0.56	0.92
Thilak	0.27	0.49	0.15	0.30	0.43	0.80

t stat – 7.21 t table (9 d.f) – 2.26

Table 8. Concentration of TSP in sesame leaves after waterlogging (mg g^{-1})

Genotype	After stress	Control	% decrease over control
Rama	27.96	45.5	38.54
TKG 308	34.24	44.3	29.38
VR1(SV2)	78.81	95.2	20.79
GT 10	60.73	81.2	33.70
S.malabaricum	17.79	28.2	58.59
Thilarani	26.83	47.3	76.29
OSC 207	36.61	46.3	26.46
TKG 22	65.25	82.0	25.67
Ayali	23.21	37.6	61.99
Thilak	31.35	40.5	28.54

t stat – 10.35

Table 9. Concentration of proline in sesame leaves after waterlogging (mg g^{-1})

Genotype	After stress	Control	% increase over control
Rama	2.32	1.14	50.86
TKG 308	1.27	0.75	40.94
SV2	2.11	1.20	43.12
GT 10	1.83	0.72	60.65
S.malabaricum	2.41	1.21	49.79
Thilarani	1.60	0.51	68.12
OSC 207	1.82	0.7	61.53
TKG 22	0.97	0.52	46.39
Ayali	2.02	0.92	54.45
Thilak	2.18	0.96	55.96

t stat – 11.18

Table 10. Concentration of SOD in sesame leaves after waterlogging (mg g^{-1})

Genotype	After stress	Control	% increase over control
Rama	0.91	0.60	34.06
TKG 308	1.62	1.39	14.19
SV2	0.29	0.25	13.79
GT 10	0.80	0.65	18.75
S.malabaricum	1.89	1.34	29.10
Thilarani	1.71	1.42	16.95
OSC 207	0.95	0.74	22.10
TKG 22	0.31	0.25	19.35
Ayali	1.75	1.50	14.28
Thilak	1.05	0.78	25.71

t stat – 5.19

was more than table t value (2.26), there was significant difference between waterlogging treatment and control for TSP. In this study among the *Sesamum indicum* genotypes, SV 2 recorded the highest TSP (78.81 mg g^{-1}) and Ayali the lowest (23.2 mg g^{-1}). *Sesamum malabaricum* recorded the lowest TSP concentration (17.79 mg g^{-1}). Percentage decrease over control was highest for Thilarani and lowest for SV 2.

4.2.4.3 Proline (mg g^{-1})

Proline content was found to be increased by waterlogging. Proline content after waterlogging was compared with control and presented in Table 9. Since t-statistic for proline content (11.18) after waterlogging was more than table t value (2.26), there was significant difference between waterlogging treatment and control for proline. It was highest for the variety Rama (2.32 mg g^{-1}) and lowest for TKG 22 (0.97 mg g^{-1}). *Sesamum malabaricum* recorded highest proline content (2.41 mg g^{-1}).

Percentage of increase in proline content by waterlogging was highest for Thilarani and lowest for TKG 308 over control. Waterlogging increased the concentration of proline by 49.79 % over control in *Sesamum malabaricum.*

4.2.4.4 Super oxide dismutase (SOD) (mg g^{-1})

Since t-statistic for SOD content (5.19) after waterlogging was more than table t value (2.26), there was significant difference between waterlogging treatment and control for SOD. SOD was found to be increasing by waterlogging. Among *Sesamum indicum* varieties highest SOD content was recorded by Ayali (1.75mg/g) and lowest by SV 2 (0.29 mg g^{-1}). *Sesamum malabaricum* recorded highest concentration (1.89 mg g^{-1}).

Percentage increase of SOD was highest for Rama and lowest for SV 2. SOD content after waterlogging was compared with control and presented in Table 10.

4.2.4.5 Phenol (mg g^{-1})

Phenol content was estimated and presented in Table 11. It was found to be increased by waterlogging treatment. Since t-statistic for phenol content (3.32) after waterlogging was more than table t value (2.26), there was significant difference between waterlogging treatment and control. Among *Sesamum indicum* varieties total phenol content was highest for Rama (0.20 mg g^{-1}) and lowest for Thilak (0.03 mg g^{-1}). Percentage increase was lowest for SV 2. *Sesamum malabaricum* recorded the highest phenol concentration (0.25 mg g^{-1}).

4.2.5 Statistical analysis

4.2.5.1 Genetic Variability Parameters based on field experiment

Phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability (h^2) and genetic advance (GA) for sixteen characters were estimated (Table 12).

PCV, GCV and GA ranged from 7.0 to 34.8, 6.7 to 34.6 and .32 to 88.1% respectively. All these three parameters were highest for yield $plot^{-1}$ and lowest for root dry weight.

In the present investigation all the sixteen characters had high heritability and thirteen characters recorded heritability above 90%. Days to maturity had highest heritability (99.9%) and days to 50% flowering had lowest for (87.2%).

4.2.5.2 Correlation analysis

The correlation coefficients were estimated for all possible combinations for sixteen characters at phenotypic and genotypic levels and the result pertaining to character association is presented in Table 13 and Table 14 respectively. In the present study phenotypic correlation coefficients were higher than genotypic correlation coefficients for all characters.

Table 11. Concentration of Phenol in sesame leaves after waterlogging (mg g^{-1})

Genotype	After stress	Control	% increase over control
Rama	0.20	0.12	40
TKG 308	0.15	0.11	26.66
SV2	0.04	0.032	20
GT 10	0.06	0.04	33.3
S.malabaricum	0.25	0.17	32
Thilarani	0.04	0.035	12.5
OSC 207	0.12	0.10	23.07
TKG 22	0.07	0.05	28.57
Ayali	0.13	0.11	15.38
Thilak	0.03	0.025	37.5

t stat - 3.32

Table 12. Estimates of variability parameters for various characters of sesame genotypes

Characters	PCV	GCV	Heritability (%)	GA as % mean
Seedling survival %	17.0	16.8	98.2	44.1
Days to 50% flowering	8.1	7.5	87.2	18.7
Plant height (cm)	9.0	8.5	88.0	21.0
Days to maturity	13.8	13.8	99.9	36.5
No. of primary branches plant^{-1}	31.4	29.5	88.1	73.1
No. of capsules plant^{-1}	25.9	34.1	94.6	64.7
Length of capsule (cm)	7.4	7.3	98.4	19.2
Seeds capsule $^{-1}$	13.9	13.9	99.4	36.7
1000 seed weight(g)	7.3	7.3	97.8	.51
No. of roots plant^{-1}	17.2	17.2	95.7	11.9
Root length (cm)	19.9	19.9	92.8	4.7
Root dry weight (g)	7.2	6.7	97.8	.32
Plants plot $^{-1}$	16.5	16.5	96.8	9.6
Yield plant^{-1} (g)	28.7	28.4	97.9	74.1
Yield plot $^{-1}$ (g)	34.8	34.6	97.4	88.1
Oil content (%)	11.6	11.5	95.5	13.3

Table 13. Genotypic correlation among various characters of sesame

	S.S.P	D.F.F	P.H	D.M	N.P.B	N.C	L.C	S.P.C	T.S.W	N.R	L.R	R.D.W	N.P.P	O.C	Y.P	S.Y.P
S.S.P	1															
D.F.F	0.81**	1														
P.H	0.07	0.14	1													
D.M	0.48**	0.47**	-.73**	1												
N.P.B	0.14	.28*	.88**	-.65**	1											
N.C	0.12	.20*	.89**	-.71**	0.87**	1										
L.C	0.21	.26*	.70**	-.67**	.87**	.62**	1									
S.P.C	0.06	.15	.94**	-.77**	.93**	0.83**	0.72**	1								
T.S.W	-0.03	-0.12	-.72**	.78**	-.65**	-.81**	-.76**	-.51**	1							
N.R	0.65**	0.72**	.08	.35**	0.31**	0.18	0.21*	0.21*	-0.17	1						
L.R	0.07	0.19	.60**	-.75**	.72**	.68**	.82**	.76**	.52**	0.18	1					
R.D.W	0.78**	0.81**	.42**	.21*	.52**	.47**	.51**	.43**	-.42**	0.73**	0.46**	1				
N.P.P	.84**	0.77**	.08	.48**	.16	.11	.21*	.07	-.03	.67**	0.07	0.77**	1			
O.C	-.51**	-.36**	.54**	-.82**	.54**	.58**	.54**	.67**	-0.62**	-0.15	0.62**	-0.2	-.41**	1		
Y.P	0.43**	0.42**	0.85**	-.52**	.86**	.89**	.82**	.91**	-.85**	0.4**	0.89**	0.64**	0.43**	0.39**	1	
S.Y.P	0.04	0.15	.88**	-.76**	.90**	.93**	.86**	.92**	-.84**	0.18	.68**	0.45**	0.05	0.62**	.89**	1

Table 14. Phenotypic correlation among various characters of sesame

	S.S.P	D.F.F	P.H	D.M	N.P.B	N.C	L.C	S.P.C	T.S.W	N.R	L.R	R.D.W	N.P.P	O.C	Y.P	S.Y.P
S.S.P	1															
D.F.F	0.84**	1														
P.H	0.08	0.18	1													
D.M	0.49**	0.52**	-.79**	1												
N.P.B	0.16	0.31**	0.90**	-.69**	1											
N.C	0.13	0.24*	0.92**	-.72**	0..89**	1										
L.C	0.21	0.29**	0.72**	-.67**	0.85**	.65**	1									
S.P.C	0.06	0.16	0.94**	-.77**	.95**	0.84**	0.73**	1								
T.S.W	-.03	-0.13	0.74**	0.81**	-.67**	-.81**	-.77**	-.52**	1							
N.R	.79**	0.74**	0.13	0.44**	0.41**	0.24	0.28**	0.24*	-0.13	1						
L.R	.06	0.23	.62**	-.76**	.75**	.69**	.85**	.77**	.56**	0.22*	1					
R.D.W	0.79**	0.83**	0.46**	0.21*	0.56**	0.48**	0.53**	0.44**	-.42**	0.87**	0.47**	1				
N.P.P	.86**	0.85**	0.06	0.48**	0.17	0.14	0.2	0.07	-.04	0.82**	0.07	0.79**	1			
O.C	-.52**	-.39**	0.58**	-.81**	0.59**	0.63**	0.54**	0.68**	-.62**	-0.18	0.64**	-0.21	-0.5**	1		
Y.P	0.44**	0.42**	.87**	-.53**	.90**	0.94**	.84**	0.93**	-.89**	0.49**	0.92**	0.68**	0.45**	0.4**	1	
S.Y.P	0.05	0.18	.89**	-.77**	.91**	0.94**	.89**	.95**	-.85**	0.23*	.72**	0.46**	0.06	0.64**	.90**	1

Seedling survival percentage had positive and significant correlation with number of plants plot^{-1} (0.84), days to fifty percentage flowering (0.80), root dry weight (0.78) and number of roots plant^{-1} (0.65).

Plant height had significant positive correlation with number of primary branches (0.88), number of seeds capsule $^{-1}$ (0.94) and number of capsules plant^{-1} (0.89). A significant negative correlation existed between plant height and thousand seed weight (-0.72).

Days to maturity had a significant positive correlation with days to fifty percentage flowering (0.47). A significant negative correlation was noticed between days to maturity and plant height (-0.73). Root dry weight had significant positive correlation with seedling survival percentage (0.78), number of roots plant $^{-1}$ (0.73) and days to fifty percentage flowering (0.81).

Highly positive and significant phenotypic and genotypic correlations existed between yield plant $^{-1}$ and seeds capsule $^{-1}$ (0.94), number of capsules plant^{-1} (0.94), length of capsule (0.86), number of primary branches plant^{-1} (0.90) and plant height (0.88). There is significant and negative correlation between yield plant $^{-1}$ with thousand seed weight (-0 .77) and days to maturity (-0.76).

4.2.5.3 Path coefficient analysis

The genotypic correlation coefficient of seed yield per plant with yield contributing characters was partitioned into different components to find the direct and indirect contribution of each character to yield (Table 15). The characters with very high and very low correlation coefficient were omited to make path diagram.

4.2.5.3.1 Direct effects

Number of capsules plant^{-1} had highest positive direct effects (0.488) on seed yield plant^{-1}. Length of capsule (0.356) and plant height (0.26) also had positive direct effects on yield. Number of primary branches plant^{-1} (-0.408) had negative direct effect with seed yield plant^{-1}.

Table 15. Direct (diagonal) and indirect (off diagonal) effect of yield component characters on seed yield per plant

	P.H	D.M	N.P.B	N.C	L.C	T.S.W	N.R	L.R	R.D.W	O.C	S.Y.P
P.H	**0.2604**	0.3218	-0.3598	0.4350	0.3246	-0.0629	0.0160	-0.1195	0.0656	0.0004	0.8819
D.M	-0.1906	**-0.4396**	0.2679	-0.3432	-0.2392	-0.0673	0.0665	0.1490	0.0336	-0.0007	-0.7636
N.P.B	0.2294	0.2884	**-0.4085**	0.4257	0.3115	0.0568	0.0593	-0.1443	0.0819	0.0004	0.9009
N.C	0.2320	0.3090	-0.3562	**0.4883**	0.2225	0.0716	0.0343	-0.1360	0.0736	0.0005	0.9399
L.C	0.2375	0.2954	-0.3574	0.3051	**0.3561**	0.0663	0.0404	-0.1633	0.0801	0.0004	0.8609
T.S.W	0.1883	-0.3403	0.2667	-0.4023	-0.2716	**-0.0874**	-0.0325	-0.1037	-0.0667	-0.0005	-0.8498
N.R	0.0221	-0.1547	-0.1282	0.0888	0.0761	0.0149	**0.1891**	-0.0364	0.1152	-0.0001	0.1868
L.R	0.1573	0.3310	-0.2978	0.3354	0.2937	-0.0455	0.0347	**-0.1982**	0.0722	0.0005	0.6837
R.D.W	0.1096	-0.0949	-0.2144	0.2304	0.1829	0.0372	0.1396	-0.0916	**0.1563**	-0.0001	0.4547
O.C	0.1411	0.3622	-0.2238	0.2846	0.1940	0.0532	-0.0287	-0.1237	-0.0333	**0.0009**	0.6265

Residual effect = -0.01

4.2.5.3.2 Indirect effects

There is positive indirect effect of number of primary branches plant^{-1} through number of capsule plant^{-1} on seed yield plant^{-1} (0.425). There is a negative indirect effect of thousand seed weight (-0.402) through number of capsules plant^{-1} with seed yield plant^{-1}. The residual effect (R^2 = -0.01) was found to be low.

4.2.5.4 Genetic divergent analysis

D^2 analysis was done to measure the degree of divergence among the genotypes. The results of the analysis are presented below.

4.2.5.4.1. Group constellation: intra and inter cluster D^2

The relative distances (D^2 values) of genotype was used to group the ten genotypes into five clusters following the method suggested by Tocher (Rao, 1952). The cluster composition of genotypes was presented in Table 16.

Cluster I was the largest cluster with four genotypes followed by cluster II and cluster III having two genotypes each. Cluster IV and cluster V had only one genotype each.

The average intra and inter cluster distances based on the total D^2 values were presented (Table 17 and Fig 1). The intra cluster distances varied from zero to 1424.43 and highest intra cluster distance was for cluster I (1424.43) and minimum was for cluster IV and V.

Inter cluster distances ranged from 2100.76 to 7739.41. Highest inter cluster distance was present between cluster II and cluster V (7739.41) while the closer proximity existed between cluster I and cluster IV (2100.76).

For cluster I, the maximum divergence distance was with cluster V (5223.09) and minimum with cluster IV (2100.76). Intra cluster distance for cluster I was 1424.43. Cluster II with two genotypes had intra cluster distance of

Fig 1. Average intra and inter cluster distances among the five clusters.

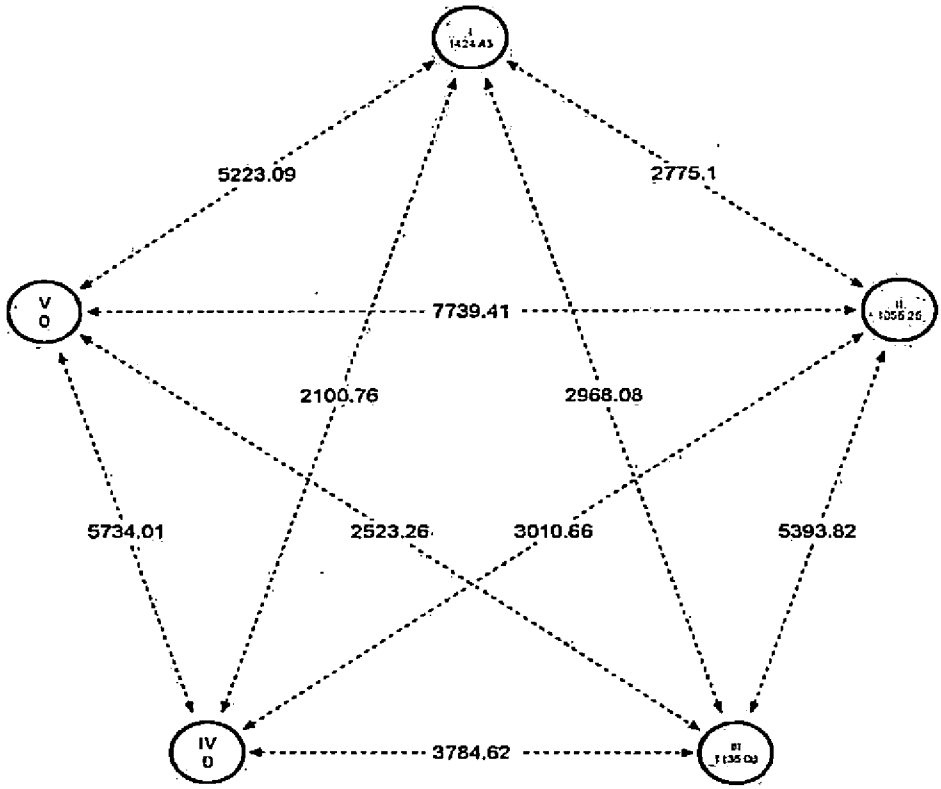


Table 16. Clustering pattern of sesame genotypes.

Clusters	Genotypes
I	Rama, TKG 22, OSC 207, GT 10
II	SV 2, TKG 308
III	Thilak, Thilarani
IV	*Sesamum malabaricum*
V	Ayali

Table 17. Average intra and inter cluster distances for the five clusters.

Clusters	I	II	III	IV	V
I	**1424.43**	2775.12	2968.08	2100.76	5223.09
II		**1055.25**	5393.82	3010.66	7739.41
III			**1135.08**	3784.62	2523.26
IV				**0.00**	5734.01
V					**0.00**

1055.25 and inter cluster distance was maximum with cluster V (7739.41) and minimum with cluster I (2775.1). The highest divergence of cluster III was recorded with cluster II (5393.82) and lowest with cluster V (2523.26).

The cluster IV and cluster V recorded maximum and minimum inter cluster values of 5734.01 (with cluster V) and 2100.76 (with cluster I) ; 7739.41(with cluster II) and 2523.2 (with cluster III) respectively.

4.2.5.4.2. Cluster means.

The cluster means obtained for sixteen characters in each cluster are presented in Table 18. Cluster IV showed the highest cluster mean for seedling survival percentage (97.6), days to fifty percentage flowering (46.67), days to maturity (134.57), length of capsule (2.94) , number of roots plant^{-1} (46.43) , root dry weight (7.39), and number of plants plot^{-1} (29.33). The maximum value of cluster mean for plant height (131.77), yield plant^{-1} (7.46), yield plot^{-1} (201.41), root length (12.2), number of seeds capsule^{-1} (42.2), number of capsules plant^{-1} (97.67) and number of primary branches plant^{-1} (8.3) was recorded in cluster V. Cluster III showed highest cluster mean for oil content (48.2) and cluster II showed highest cluster mean for thousand seed weight (2.97).

Cluster IV had lowest cluster mean for plant height (101.43) number of primary branches plant^{-1} (5.2), , number of capsules plant $^{-1}$ (41.33), number of seeds capsule^{-1} (26.67), seed yield plant^{-1} (2.91), thousand seed weight (2.41), root length (6.48) and percentage of oil content (32.53). Cluster II had lowest mean for seedling survival % (63.5), days to fifty percentage flowering (36.7), seed yield plot^{-1} (78.12), root dry weight (7.01), number of plants plot $^{-1}$ (19.67). Number of roots had lowest cluster mean in cluster I (33.23).

4.2.5.5 Selection index

Selection index was calculated based on all the biometric observations viz, seedling survival %, plant height, days to 50% flowering, days to maturity, number of primary branches plant^{-1}, number of capsule plant^{-1}, length of capsules,

Table 18. Cluster means for various characters

Character	I	II	III	IV	V
Seedling survival %	72.5	63.5	70.09	97.6	88.6
Days to 50% flowering	39.0	36.7	41.6	46.6	43.3
Plant height(cm)	119.5	106.5	126.8	101.4	131.7
Days to maturity	95.7	99.2	88.6	134.5	88.7
No. of primary branches plant^{-1}	7.05	6.2	7.7	5.2	8.3
No. of capsules plant^{-1}	65.5	49.2	77.3	41.3	97.6
Length of capsule (cm)	2.3	2.2	2.4	2.94	2.7
Seeds capsule $^{-1}$	34.5	30.5	39.3	26.6	42.2
1000 seed weight(g)	2.7	2.9	2.5	2.4	2.5
No. of roots plant^{-1}	33.2	35.1	40.4	46.4	43.3
Root length(cm)	9.4	7.5	11.08	6.4	12.2
Root dry weight(g)	7.1	7.01	7.2	7.39	7.3
Plants plot $^{-1}$	21.9	19.6	21.3	29.3	27.1
Yield plant^{-1} (g)	7.1	7.01	7.2	2.9	7.4
Yield plot $^{-1}$ (g)	119.4	78.1	144.9	85.3	201.4
Oil content (%)	43.5	44.2	48.2	32.5	45.4

number of seeds capsules^{-1}, root length, number of roots plant^{-1}, root dry weight, number of plants plot^{-1}, 1000 seed weight, yield plant^{-1}, yield plot^{-1} and oil content. The index value of each genotype was determined and the genotypes were ranked accordingly.

The selection index scores presented in Table 19 revealed that it was highest for Ayali (1319.6) followed by Rama (1208.4) and OSC 207 (1205.3). Selection index was lowest for SV 2 (1028.1). *Sesamum malabaricum* the wild species recorded a selection index score of 1134.7.

Table 19. Selection index scores of the ten sesamum genotypes

Genotype	Selection index score
Ayali	1319.6
Rama	1208.4
OSC 207	1205.3
Thilarani	1202.8
Thilak	1176.6
S. malabaricum	1134.7
GT 10	1117.1
TKG 22	1086.9
TKG 308	1079.7
SV 2	1028.1

Discussion

(A) (B)

Plate 9. Superior genotypes identified for waterlogging tolerance A) *Sesamum indicum* variety Ayali B) *Sesamum malabaricum*

5. DISCUSSION

Sesame is a plant breeder's dream crop because of its high variability. It is basically an arid crop and highly susceptible to excess moisture conditions. Occurrence of an unexpected rainfall causes a severe damge to crop and is one of the major threats faced by farmers nowadays.

The present study was envisaged to evaluate sesame genotypes for tolerance to excess soil moisture conditions, to elucidate the mechanism for tolerance and to find out the selection index for identification of tolerant genotypes. The results of the experiment conducted are discussed here.

5.1 EFFECTS OF WATERLOGGING ON THE IMPORTANT BIOMETRIC CHARACTERS

Waterlogging is one of the abiotic stresses which affect the plants in a negative way. Waterlogging will cause reduction in plant growth and development and eventually yield.

5.1.1 Survival percentage

Preliminary screening was done as pot culture experiment and waterlogging was imposed twenty days after sowing. Seedling test was conducted under controlled conditions because it is the most common method to screen waterlogging tolerance of crop plants and seedling stage is the most sensitive stage than flowering and maturity stage. It is presumed that if the plant survives in the seedling stage, it can survive the remaining stages also, as reported by Zhou *et al.* (2014) in rapeseed. Sarkar *et al.* (2016) also reported that waterlogging commonly damages the seedling establishment stage. In the present investigation a wide variation in survival percentage was observed among the genotypes. When the effect of waterlogging on seedling survival percentage was compared with control, all the genotypes survived under controlled condition. Seventeen genotypes survived waterlogging for 72 h and not a single plant of the rest of

thirteen genotypes could overcome this abiotic stress. Survival percentage is an important mean to assess the degree of flood tolerance as reported by Martin *et al.* (2006). Variability in this trait is directly related with genetic variability for flooding tolerance.

Flooding tolerance varies between crops,varieties, stages of crop, duration of flooding etc. Flooding did not lead to death of seedlings in Arabidopsis (Pigliucci and Kolodynska, 2002). while different degrees of damage is caused in sesame,This shows that sesame is very sensitive to waterlogging as in rapeseed (Zhou *et al.* ,2014). In this study tolerance level differed between wild species, *Sesamum malabaricum* and cultivated *Sesamum indicum*. *Sesamum malabaricum* recorded cent percent seedling survival. Wild relatives are proved to be source of stress tolerance (Moazzami *et al.*, 2006). .Intra specific variation for flood tolerance was also observed. Seventeen genotypes of *Sesamum indicum* survived waterlogging and twelve genotypes could not withstand the waterlogging condition. Similar varietal difference was reported by Zhou *et al.* (2014) in rapeseed. Highest survival percentage was recorded by Ayali (50.1%) and lowest by AT 282 (3.2%). Such intraspecific varietal difference was in conformity with the study of Noguchi and Morokuma (2007) in rice and Hussain *et al.* (2014) in cotton.

Duration of flooding also affected the seedling survival percentage. All the thirty genotypes survived 24 h and 48 h of waterlogging and only seventeen genotypes survived 72 h of waterlogging. Sarkar *et al.* (2016) also reported that duration of waterlogging can reduce crop growth in sesame.

Analysis of variance showed significant difference for all the characters ie, Survival survival %, Plant height, days to 50% flowering, days to maturity, number of primary branches plant^{-1}, seeds capsule^{-1}, root length, root dry weight, number of roots plant^{-1} , and seed yield plant^{-1}and significant difference for length of capsule and 1000 seed weight, This indicates that there is considerable amount

of genetic variation occures in the seventeen genotypes which can be made use of in future breeding programme.

Survival percentage was taken as the criteria for selection of waterlogging tolerant genotypes. The genotypes SV 2 (40.2%), TKG 22 (41.4%) , Thilak (42.4%), GT 10 (44.2%), Thilarani (45.2%), TKG 308 (45.2%), OSC 207 (45.2%) Rama (49.2%), Ayali (50.1%) and *Sesamum malabaricum* (100%) which recorded the highest percentage of survival at 72 h of flooding were selected for further field study.

In the field, genotypes showed significant difference in survival percentage. *Sesamum malabaricum* recorded the highest (97.6%) followed by Ayali (88.6%). The genotype SV 2 recorded the lowest survival percentage (55.2%). All the genotypes except *Sesamum malabaricum* recorded a higher survival value in the field than in pot culture experiment. This may be due to percolation of water in the soil under field condition. The degree of tolerance exhibited by the genotypes in the field is similar to that in the pot. This clearly denotes the presence of genetic influence in flood tolerance.

5.1.2 Plant height

Plant height was reduced by waterlogging in all the genotypes under study which was in conformity with the findings of Hossain and Salahuddin (2001) in sesame. The reduction in plant height may be due to the energy conservation for maintenance and survival. Wild species *Sesamum malabaricum* recorded lowest plant height.

Among *Sesamum indicum* genotypes, highest plant height was recorded by Ayali and lowest by SV 2. Fukao *et al.* (2006) reported that plant height is a useful adaptation for tolerance in rice under shallow flooded soil. In this study also the tolerant genotype Ayali recorded the highest plant height.

5.1.3 Days to 50% flowering and Days to maturity

In all the genotypes waterlogging delayed days to 50% flowering and maturity. Similar observation was made by Amri *et al.* (2014) in six bread wheat genotypes. The wild species, *Sesamum malabaricum* recorded the maximum days to 50% flowering. Among the cultivated types. Ayali recorded maximum days for 50% flowering and SV 2 recorded the minimum Maximum duration for maturity was recorded by the wild species *Sesamum malabaricum and* minimum by Thiiak which is the most desirable character for inclusion as a component in rice based cropping system.

5.1.4 Number of primary branches plant^{-1} and number of capsules plant^{-1}

Number of primary branches plant^{-1} and number of capsules plant^{-1} are important yield contributing characters. In the present study, among *Sesamum indicum* genotypes highest values for these character was recorded by Ayali and lowest by SV 2. The wild species *Sesamum malabaricum* recorded a lower value than that of SV 2. Same trend was seen in number of capsules plant^{-1} also. Number of primary branches plants^{-1} and capsules plant^{-1} were high in control for all the genotypes. Waterlogged condition reduced the number of primary branches plant^{-1} and number of capsules plant^{-1}. Similar results were reported by Ghoi *et al.* (1996) and Gutierrez *et al.* (1996).

5. 1. 5 Number of Seeds Capsule^{-1} and Thousand seed weight

Waterlogging condition reduced number of seeds capsule^{-1} and thousand seed weight in all the genotypes studied. Wild species *Sesamum malabaricum* recorded lowest thousand seed weight and among the *Sesamum indicum* genotypes highest thousand seed weight was recorded by SV 2 and lowest by Ayali.

5.1.6 Root morphology

Root characters like root length, root dry weight and number of roots plant^{-1} were the important indices for determining waterlogging tolerance. In the present investigation number of roots plant^{-1} and root dry weight were increased and root length was decreased by waterlogging. Among the *Sesamum indicum* genotypes Ayali recorded highest value for these characters and SV 2 the lowest. The wild species *Sesamum malabaricum* had higher root dry weight and number of roots than Ayali

In the present study tolerant genotypes had higher root dry weight and number of roots plant^{-1} than susceptible genotypes which was in accordance with Malik *et al.*(2009) and Marashi and Mojadham (2014) but contradictory to the findings of Zhang *et al.*(2015). The root length was decreased under waterlogging condition in all the genotypes which was in confirmatory with the findings of Zhang et *al.* (2015).

5.1.7 Seed yield and oil content

Seed yield is the most desirable character for breeders to develop a variety. Waterlogging tolerance drastically reduces the yield of plants. These may be due to reduced photosynthetic rate. Water logged condition creates loss of chlorophyll content and it eventually leads to reduction in photosynthetic rate, hence yield.

Present study also revealed that there was a reduction in yield in all the genotypes by waterlogging which may be due to the reduction in the yield attributing characters. These results are in accordance with the findings of Sparrow and Uren (1987) in cowpea, Marashi and Chinchanikar (2014) in wheat and Sarkar *et al.* (2016) in sesame.

Oil content is an important economic character for oil seed crop. Among *Sesamum indicum* genotypes oil content was highest for Thilak and lowest for GT 10. But the genotype Ayali recorded highest oil yield followed by Thilak and

Thilarani. Wild sesame species, *Sesamum malabaricum* recorded low oil content (30.2%). Akhila and Beevy (2015) also reported that wild species are low in oil content and *Sesamum malabaricum* has an oil content of 21.67%.

5.2 ANATOMICAL STUDY

Aerenchyma tissue is helpful for exchange of oxygen from aerobic shoot to anaerobic root. Formation of aerenchyma is an important adaptive character for waterlogging tolerance. Plants with well developed aerenchyma will overcome adverse condition quickly.

In the present investigation root anatomy was studied for the ten genotypes. Three genotypes viz. the wild species *Sesamum malabaricum*, and the varieties Ayali and OSC 208 of *Sesamum indicum* had significant formation of aerenchyma compared to others. This implies that these genotypes had more waterlogging tolerance than others. Similar results were reported by Malik *et al.* (2009) in wheat.

5.3 BIOCHEMICAL STUDY

The comprehensive actions of biochemical compounds are important factor for determining waterlogging tolerance. Plants automatically synthesize certain antioxidants and osmoregulants under abiotic stresses to combat the stress condition.

In the present investigation, the concentration of chlorophyil, proline, SOD, phenol and total soluble protein of the fresh leaves was estimated immediately after waterlogging and compared with control.

Waterlogging reduced the chlorophyll content in all the genotypes Among ten genotypes Ayali recorded highest content of chlorophyll a, chlorophyll b and total chlorophyll while SV 2 recorded the lowest value.

Fig 2. Chlorophyll content in sesame leaves after waterlogging (mg g $^{-1}$)

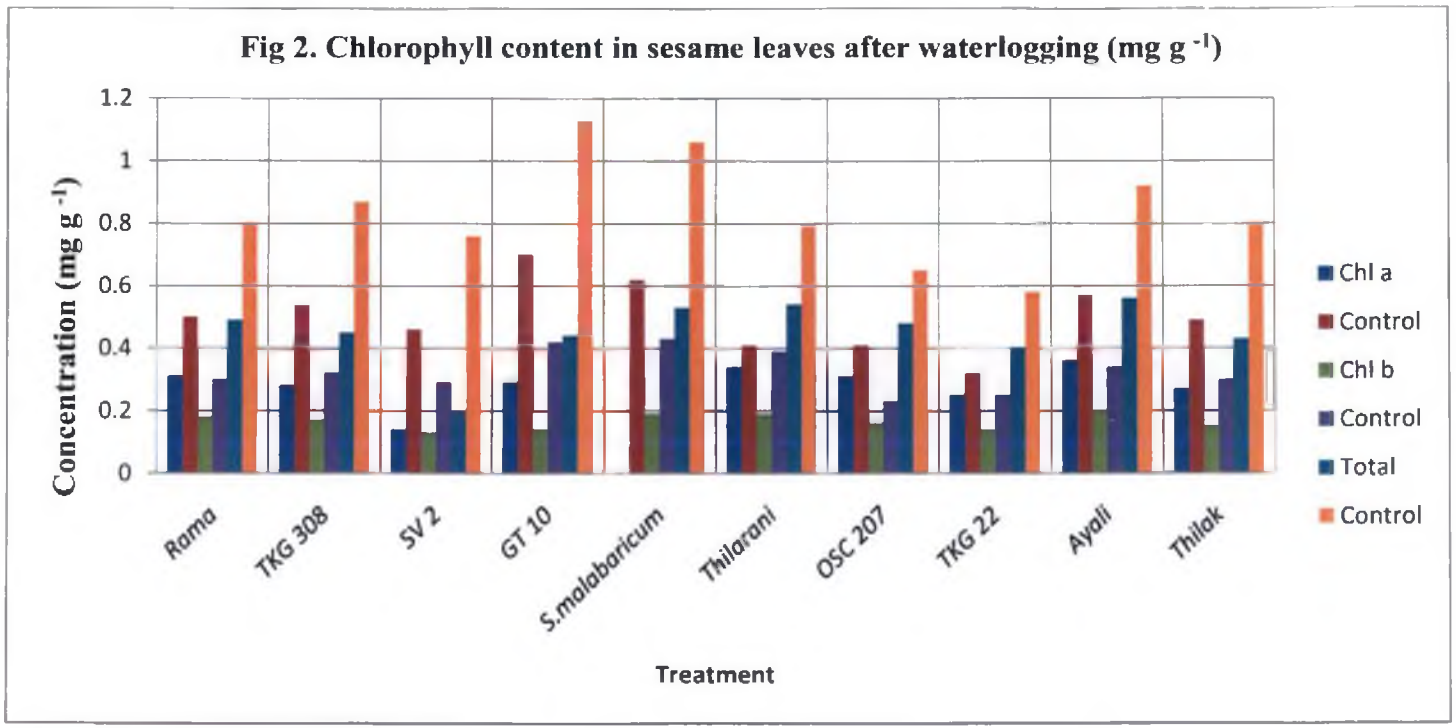


Fig 3. Concentration of TSP in sesame leaves after waterlogging (mg g^{-1})

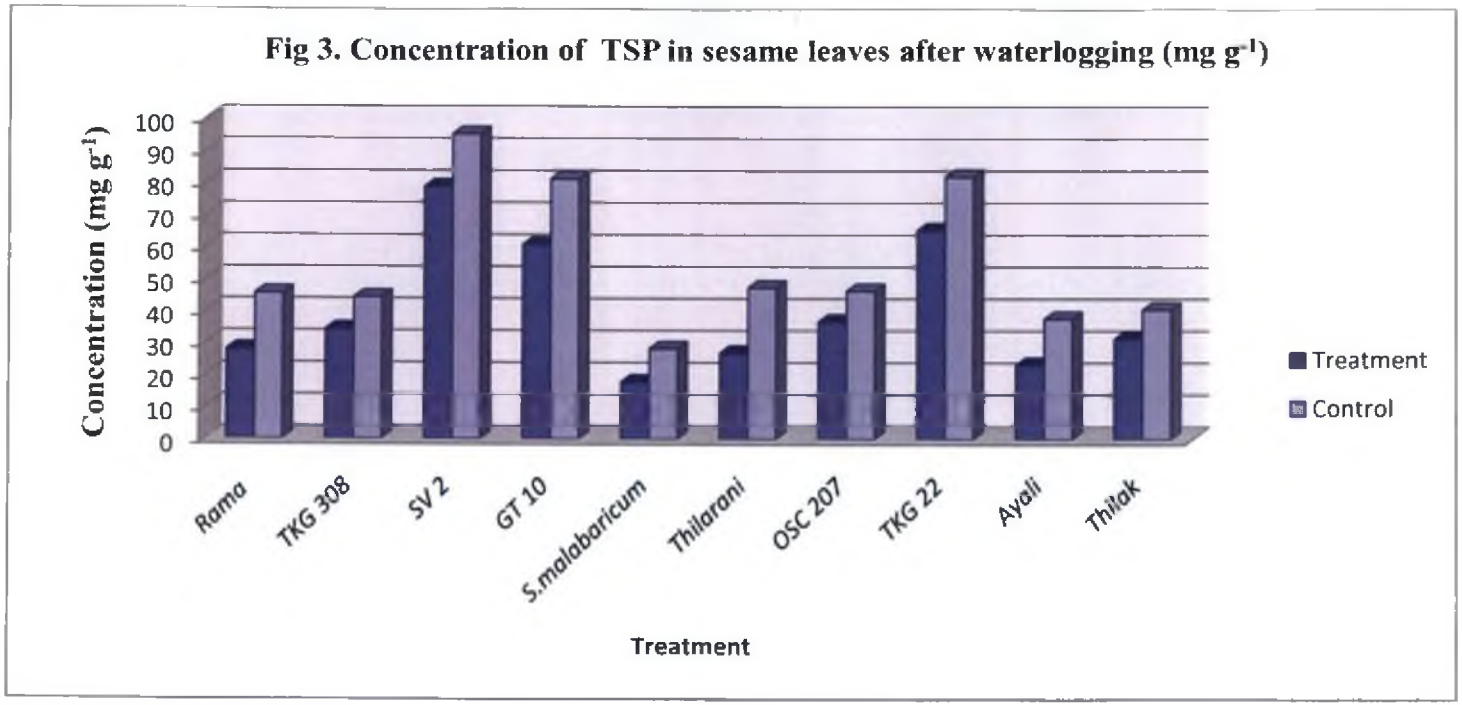


Fig 4. Concentration of proline in sesame leaves aftter waterlogging (mg g $^{-1}$)

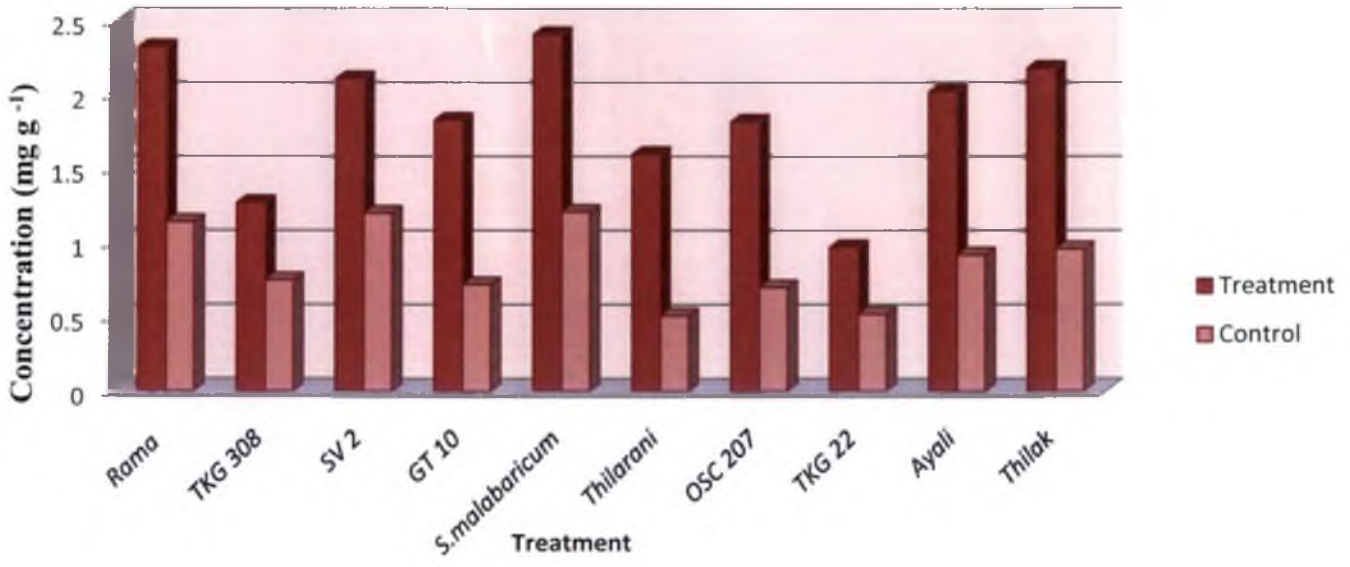


Fig 5. Concentration of SOD in sesame leaves after waterlogging (mg g^{-1})

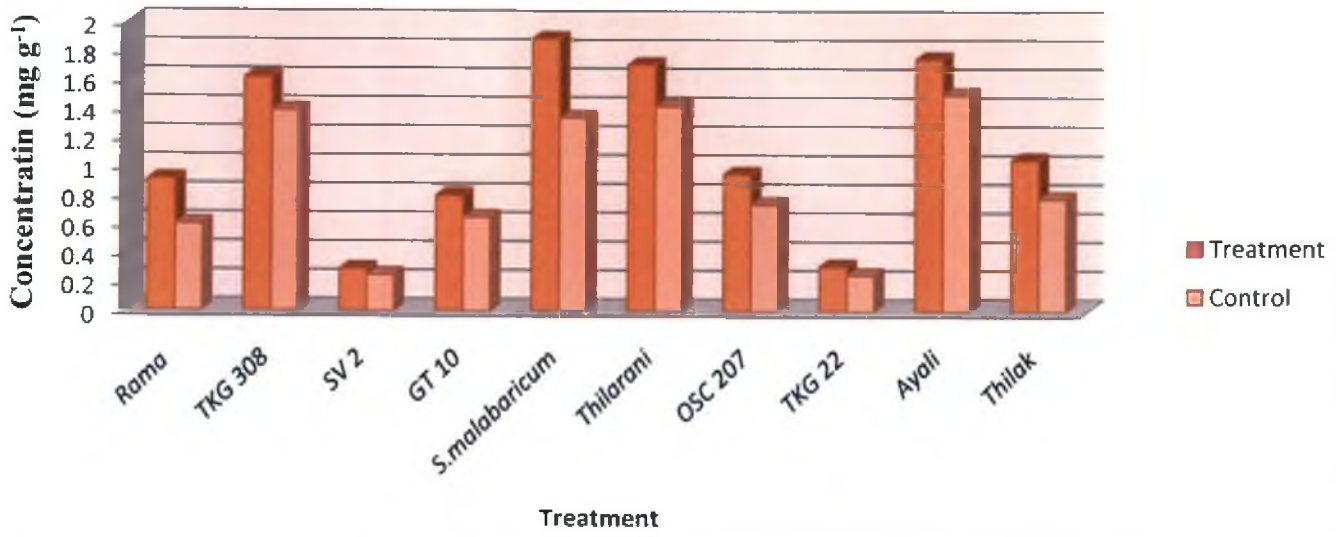
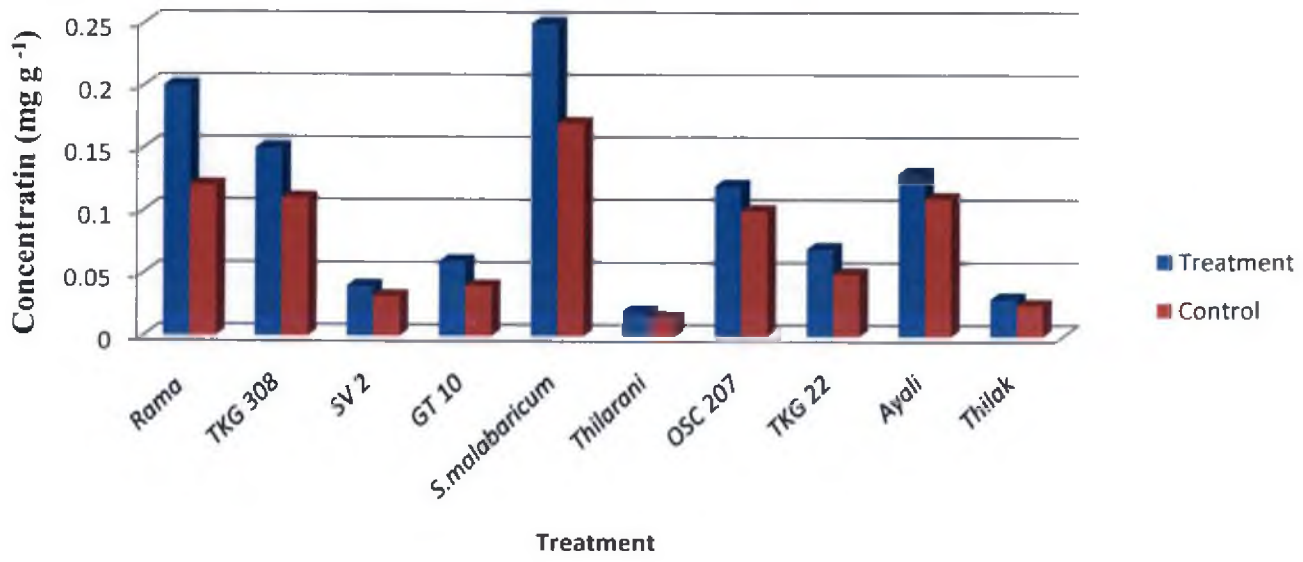


Fig 6. Concentration of phenol in sesame leaves after waterlogging (mg g $^{-1}$)



Similar observations were made by Wang *et al.* (1999) in sesame. Maximum chlorophyll retension was exhibited by tolerant variety Ayali.

Waterlogging induces the production of the osmoregulant such as proline in the plant system. In the present study also waterlogging increased the concentration of proline and it was highest for the variety Rama and lowest for TKG 22 in *Sesamum indicum*. *Sesamum malabaricum* recorded highest proline content. The report of Xing and Cai (1998) is in agreement with this result.

Stress condition causes the formation of free radicals in the plant system causing senescence of plants. Which will lead to the production of antioxidants to protect the plant system. SOD is one of the antioxidant produced during stress situations. Increase in SOD activity under flooding stress is an indication of an increased production of reactive oxygen species. In the present investigation also it was found that SOD content increased in all the genotype under waterlogged condition. Studies of Yan *et al.* (1996) and Xu *et al.* (2012) in sesame reported similar results.

Phenol is a secondary metabolite which acts as another antioxidant at stress situation. Present study revealed that waterlogging caused an increased concentration of phenol in plants. *Sesamum malabaricum* recorded the highest phenol concentration. This finding is in conformity with the observation of Nikam (2007) in *Chlorophytum borivillianum*. But phenol concentration has no role in imparting waterlogging tolerance in other genotypes.

Total soluble proteins (TSP) are macromolecules involved in almost all cellular activities. Present study revealed that, total soluble protein was reduced under water logged condition in all the genotypes. Water logged condition caused anaerobic condition in the plant system and the plant utilize might have the reserve protein for respiration and hence TSP concentration was reduced. These results are in agreement with the reports of Alves *et al.* (2012) in yellow lapacho.

In general concentration of proline, SOD and phenol increased under waterlogged condition while chlorophyll and TSP decreased. In the present study also same trend was recorded in all the genotypes. Wild species *Sesamum malabaricum* and cultivated variety Ayali recorded the maximum increase in proline, SOD and phenol which confirmed the waterlogging tolerance expressed by both the genotypes.

5.4 STATISTICAL ANALYSIS

5.4.1 Variability studies

Analysis of variance revealed significant difference among the genotypes for all the characters studied, which indicated the presence of substantial amount of variability. So selection could be effective for improvement of those characters. Similar results have also been reported by Solanki and Gupta (2001) and Valarmathi and Saravana (2004) in sesame.

In the present investigation highest PCV and GCV was recorded for seed yield plant^{-1} followed by number of primary branches per plant and number of capsules plant^{-1}. These result are in accordance with the study of Vasline *et al.* (2000), Saravanan and Nadarajan (2003), Parameshwarappa *et al.* (2009)., Sumathi and Muralidharan (2009) , Bindu *et al.*(2014) and Abhijatha (2014) in sesame.

5.4.1.1 Heritability and Genetic advance

The ratio of genotypic variance to phenotypic variance is known as heritability and it is the heritable portion of phenotypic variance. It is a good index of the transmission of characters from parents to offspring. The estimates of heritability help the plant breeder in selection of elite genotypes from diverse genetic populations. Improvement in the mean genotype value of selected population over the parental one is known as genetic adavance. High value of

genetic advance shows that character is governed by additive genes and selection will be effective for improvement of such trait.

These results were in agreement with Solanki and Gupta (2001). Genetic advance was highest for seed yield. Characters like number of seeds capsule^{-1}, number of capsules plant^{-1} and number of primary branches plant^{-1} had high genetic advance. Similar results were reported by Narain *et al.* (2004) and Babu *et al.* (2005) in sesame. In the present study high heritability was observed for all the characters studied.

High heritability with genetic advance is usually more helpful in predicting genetic gain under selection than heritability estimates alone. Present study implied that number of primary branches plant^{-1}, number of capsules plant^{-1}, days to maturity, number of seeds capsule^{-1}, plant height and seed yield had high heritability and genetic advance. Similar results were reported by Sumathi and Muralidharan (2009) in sesame. So selection based on the above described characters will be effective for improvement of sesame.

5.4.2 Correlation study

The statistics which measures the degree and direction of association between two or more variables is known as correlation. Correlation studies provide information about yield and yield contributing characters and this will helpful for selection from diverse populations.

In the present investigation there was high genotypic and phenotypic correlation of seed yield with seeds capsules^{-1}, number of capsules plant^{-1}, length of capsule, number of primary branches plant^{-1} and plant height. Similar results were reported by Tomar *et al.* (1999) and Parameshwarappa *et al.* (2009) in sesame.

1000 seed weight showed significant negative correlation with seed yield plant^{-1}. Similar results were published by Pawar *et al.* (2002) and Sankar and Kumar (2003) in sesame.

Days to maturity showed significantly positive correlation with number of capsules plant^{-1}, length of capsule and number of seeds capsule^{-1}. Reports of Ramireddy and Sundaram (2002) also supported such positive correlation in sesame.

5.4.3 Path analysis

Path analysis measures the direct and indirect contribution of various independent characters on a dependent character. The path analysis reveals whether the association of these characters with yield is due to their effect on yield or is a consequence of their indirect effects via other component characters.

In this study, a path coefficient analysis was used to separate the genotypic correlation coefficients of seed yield plant^{-1} with plant height, days to maturity, number of primary branches plant^{-1}, length of capsule, root length, root dry weight, number of roots plant^{-1}, 1000 seed weight and oil content into direct and indirect effects. The path analysis revealed that seed yield plant^{-1} was positively and directly affected by number of capsules plant^{-1}, plant height and length of capsule. The greater influence of these traits reflects their importance as yield components. Similar results were reported by Ganesh and Sakila (1999) and Kurdistani *et al.* (2011) in sesame.

Days to maturity and number of primary branches plant^{-1} showed a direct negative effect on yield plant^{-1} and it is analogous to the report of Siddiqui *et al.* (2005). Number of roots and root dry weight had direct positive effect while root length had negative direct effect on seed yield plant^{-1}.

According to this study it can be concluded that to increase seed yield under high moisture conditions for assessment of selection criteria in sesame, characters like number of capsule plant^{-1}, plant height, length of capsule and number of roots plant^{-1} be increased. The residual effect (R^2 = - 0.01) indicated that the characters selected are ideal in breeding programmes for waterlogging

tolerance and the remaining character have only minor contribution in the variability of seed yield.

5.4.4 Genetic divergence and clustering

In plant breeding genetic diversity plays an important role because hybrids between lines of diverse origin display a greater heterosis than those between closely related parents.

The D^2 statistic measures the forces of differentiation at intra- and inter-cluster levels and determines the relative contribution of each component trait to the total divergence. Clustering using D^2 (genetic distance) matrix is useful for analyzing the divergence of the population to identify genotypic variability. Clusters separated by the largest D^2 (genetic distance) show the maximum divergence, while the genotypes in the same clusters or groups are less divergent.

In the present study maximum genetic variability will be produce by crossing of lines in cluster II and cluster V which recorded highest intercluster distance. Also inter cluster distances were greater in magnitude than intra cluster distances, indicating the presence of diversity among the clusters. Bandila *et al.* (2001) also made similar inferences in sesame.

5.4.5 Selection index

The selection index technique can theoretically determine the genotypic worth of individuals or families in an objective manner. In general, selection indices provide a useful method for quantifying selection potential as well as providing a good chance for more efficient selection.

In the present investigation selection index was calculated based on all the biometric observations viz: seedling survival %, plant height, days to 50% flowering, days to maturity, number of primary branches plant^{-1}, number of capsule plant^{-1}, length of capsules, number of seeds capsules^{-1}, root length, number of roots plant^{-1}, root dry weight, number of plants plot^{-1}, 1000 seed

weight, yield plant^{-1}, yield plot^{-1} and oil content. Maximum selection index score was recorded by Ayali followed by Rama and OSC 207 and these genotypes may be identified as suitable for cultivating under waterlogged conditions.

Summary

6. SUMMARY

The project "Evaluation of sesame genotypes tolerant to waterlogged conditions" was conducted at Onattukara Regional Agricultural Research Station, Kayamkulam during 2014-16. The germplasam was evaluated for waterlogging tolerance. Yield and yield contributing characters, anatomical and biochemical changes and statistical parameters were studied.

Preliminary screening for excess moisture was done using thirty accessions collected from the germplasam of Department of Plant Breeding and Genetics, Onattukara Regional Agricultural Research Station, Kayamkulam by pot culture experiments. Ten genotypes were selected from this study and evaluated in the field. The relationship among the yield and yield attributing traits was also worked out. Biochemical analysis of leaves and anatomical study of the roots were done after waterlogging treatment. The population was statistically analysed to study the correlation between various economic traits, direct and indirect effect of independent variables over dependent variable, genetic divergence of the genotype. Simultaneous selection indices were computed to identify the high yielding waterlogging tolerant genotypes.

The salient findings of the investigation are summarised below.

- Preliminary screening of thirty sesame genotypes revealed that significant variability occurred in tolerance to waterlogging. Ten genotypes which recorded comparatively high survival percentage under excess moisture conditions for 72 h viz, Thilak, Thilarani, *Sesamum malabaricum*, Ayali, Rama, TKG 308, TKG 22, SV 2, OSC 207 and GT 10 were selected.
- Analysis of variance of thirteen biometric characters viz. seedling survival %, plant height, days to 50% flowering, days to maturity, number of primary branches plant^{-1}, number of capsules plant^{-1}, length of capsules, number of seeds capsules^{-1}, root length, number of roots plant^{-1}, root dry weight, 1000 seed weight and yield plant^{-1} revealed significant differences

among genotypes for all the characters showing the presence of high genetic variability.

- The assessed germplasm had sufficient variability and scope for selection based on the characters like number of plants plot $^{-1}$, yield plot^{-1} and oil content.
- Among the selected genotypes highest survival percentage was recorded by the wild species *Sesamum malabaricum* while Ayali recorded the highest survival percentage in *Sesamum indicum* genotypes.
- Biochemical studies revealed that genotypes like *Sesamum malabaricum*, Ayali and OSC 207 have comparatively more production of antioxidants viz.SOD and phenol and osmoregulant proline after waterlogging.
- Anatomical study emphasized that among ten genotypes *Sesamum malabaricum*, Ayali and OSC 207 have significant formation of aerenchyamatous tissues in roots.
- Phenotypic coefficient of variation, genotypic coefficient of variation and genetic advance were highest for yield plot^{-1}.
- High heritability was recorded for all the characters. Days to maturity has highest heritability (99.9 %).
- Highly positive and significant phenotypic and genotypic correlations existed in seeds capsule^{-1}, number of capsules plant^{-1}, length of capsule, number of primary branches plant^{-1} and plant height with yield plant^{-1}. Yield plant^{-1} recorded significant and negative correlation between with thousand seed weight and days to maturity.
- Number of capsules plant^{-1} had highest positive direct effects on seed yield plant^{-1}. Length of capsule and plant height had positive direct effects on yield. Selection based on these characters would be effective for developing high yielding waterlogging tolerant varieties of sesame.
- Genotypes were grouped into five clusters based on genetic divergence study. Cluster I was the largest group with four genotypes, followed by cluster II and cluster III with two genotypes each. Cluster IV and cluster V

had only one genotype each. There was maximum intra cluster distance for Cluster I and maximum inter cluster distance for cluster II and cluster V.

- Simultaneous selection index was calculated on the basis of all the biometric observations studied. Selection index score was high in Ayali (1319.65), followed by Rama (1208.48) and OSC 207 (1202.85). It was lowest in SV 2(1028.14).
- Future line of work

  The present investigation had both preliminary screening of waterlogging tolerance followed by field experiment. From this study, *Sesamum malabaricum* was identified as a source of waterlogging tolerance, which can be used as a donor parent in future breeding programmes.

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EVALUATION OF SESAME GENOTYPES FOR TOLERANCE TO WATERLOGGING

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ABSTRACT

The project entitled "Evaluation of sesame genotypes for tolerance to Waterlogging" was undertaken with an objective to identify sesame genotypes which are tolerant to excess soil moisture conditions, to elucidate the mechanism for tolerance and to find out the selection index for identification of tolerant genotypes. Two experiments viz. pot culture and field experiments were conducted during 2014-16 at Onattukara Regional Agricultural Research Station, Kayamkulam.

Thirty genotypes were screened at seedling stage (20 days after sowing) by pot culture experiment to identify tolerant types for excess soil moisture by imposing flooding for 24 hours, 48 hours and 72 hours duration. All the genotypes survived 24 h and 48 h of waterlogging while 17 genotypes alone survived 72 h of waterlogging. Observations of the genotypes which survived 72 h of flooding were recorded and the results showed that the genotypes differed significantly for the characters under study. Ten genotypes viz. Ayali, *Sesamum malabaricum*, TKG 22, OSC 207, Thilak, Thilarani, GT 10, SV 2, TKG 308 and Rama which recorded the highest survival percentage were selected for field experiment

In the field experiment flooding was imposed for 72 h duration and biometrical characters were recorded and statistically analysed. All the characters showed significant difference among the genotypes. Among the ten genotypes highest yield per plant was recorded by the local variety, Ayali (7.46g) and lowest by *Sesamum malabaricum* (2.92g). Oil content was highest for Thilak (48.6 %) and lowest for wild species *Sesamum malabaricum* (32.5%).

To elucidate the mechanism for flood tolerance, anatomical and biochemical studies were conducted. Anatomical study revealed that there was significant formation of aerenchymatous tissue in the flood tolerant genotypes compared to the control. In the biochemical study, the total chlorophyll content was highest for Ayali (0.56 mg g^{-1}) and lowest for SV 2 (0.20 mg g^{-1}). The total soluble protein was highest for SV 2 (95.2 mg g^{-1}) and lowest for *Sesamum malabaricum* (28.2 mg g^{-1}). Proline content was highest for *Sesamum*

malabaricum (2.41 mg g^{-1}) and lowest for TKG 22 (0.97 mg g^{-1}). Superoxide dismutase (SOD) was highest for *Sesamum malabaricum* (1.89 mg g^{-1}) and lowest for SV 2 (0.29 mg g^{-1}). Phenol content was highest for *Sesamum malabaricum* (0.25 mg g^{-1}) and lowest for Thilak (0.03 mg g^{-1}).

Phenotypic coefficient of variation (34.8), genotypic coefficient of variation (34.6) and genetic advance (88.1) were highest for yield per plot. Heritability was highest for number of days to maturity (99.9 %.). There was a strong positive genotypic (0.92) and phenotypic correlations (0.95) between number of seeds per capsule and yield per plant. There was a negative correlation between oil content and days to maturity (-0.80). In path analysis direct effect number of capsules per plant recorded highest positive direct effect (0.488) with yield per plant. Plant height registered a positive indirect effect through number of capsules per plant (0.435). Ten genotypes were grouped in to five clusters by genetic divergence analysis. There was maximum intra cluster distance for cluster I (1424.43) and maximum inter cluster distance for cluster II and cluster V (7739.41). Selection indices were calculated on the basis of all the biometric observations and it was highest for Ayali (1319.65) followed by Rama (1208.48) and OSC 207 (1205.30).

In the present study, *Sesamum malabaricum*, a wild species and Ayali, a local cultivar of *Sesamum indicum* were identified to have tolerance to excess moisture conditions. The production of aerenchymatous tissue and comprehensive actions of proline, SOD and phenol production gave excess moisture tolerance in plants. These tolerant genotypes can be used as parents for further breeding programmes.