EFFECT OF EXOGENOUS APPLICATION OF GLYCINE BETAINE ON COTTON (*Gossypium hirsutum* L.) UNDER MOISTURE STRESS

THESIS

Submitted to
Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola
in partial fulfilment of the requirements
for the Degree of

MASTER OF SCIENCE
IN
AGRICULTURE
(AGRICULTURAL BOTANY)
(Plant Physiology)

By

PANDE HARSHAL SANJAY

DEPARTMENT OF AGRICULTURAL BOTANY
POST GRADUATE INSTITUTE, AKOLA

Dr. PANJABRAO DESHMUKH KRISHI VIDYAPEETH,
KRISHINAGAR PO, AKOLA (MS) 444104

Enrolment Number - DD/369

2012

DECLARATION OF STUDENT

I hereby declare that the experimental work and its interpretation of the Thesis entitled **"Effect of exogenous application of Glycine betaine on cotton (*Gossypium hirsutum* L.) under moisture stress"** or part thereof has neither been submitted for any other degree or diploma of any university, nor the data have been derived from any thesis / publication of any university or scientific organization. The sources of material used and all assistance received during the course of investigation have been duly acknowledged.

Place: Akola

(Pande Harshal Sanjay)

Date: 14/06/2012

Enrolment Number-DD/369

CERTIFICATE

This is to certify that the thesis entitled **"Effect of exogenous application of Glycine betaine on cotton (*Gossypium hirsutum* L.) under moisture stress"** submitted in partial fulfillment of the requirement for the degree of **"Master of Science in Agriculture (Agricultural Botany)"** of Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola is a record of bonafide research work carried out by **Pande Harshal Sanjay** under my guidance and supervision.

The subject of the thesis has been approved by the student's Advisory Committee.

Place: Akola

Date: 14/06/2012

(**Dr.T.H.Rathod**)
Chairman,
Advisory Committee

Countersigned

Associate Dean,
Post Graduate Institute,
Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola.

THESIS APPROVED BY THE STUDENT'S ADVISORY COMMITTEE INCLUDING EXTERNAL EXAMINER (AFTER VIVA-VOCE)

1. Chairman Dr.T.H.Rathod
2. Member Dr.B.R.Patil
3. Member Dr.D.V.Durge
4. Member Dr.S.J.Gahukar
5. Member Dr.S.W.Jahagirdar
6. External Member(S. B. Amarshettiwar

ACKNOWLEDGEMENT

Thus it completed!

Today, I am finishing my research project. It is really a unique experience I earned during last two years and I experienced the fact that any successful achievement is not possible without support, help, guidance and encouragement of many helping hands, inspiring minds and guiding brains and the same is true with me. So, I am trying to acknowledge all the personalities who made my research work possible.

My acknowledgement should be for the guiding person, Dr. T.H. Rathod, Associate Professor, Cotton Research Unit, Dr. P.D.K.V. Akola, Chairman of my advisory committee, who guide not only papers but in the real sense he is guide. His continuous guidance always directed my study towards success. Besides his support and friendly behaviors released my burdens in the hours of need. He taught me, scold me and ultimately made me liable to reach this moment. His share is of unique importance right from the selection of topic till the submission of thesis, positively.

It is my proud privilege to record my cordial thanks to all members of my advisory committee Dr. B.R. Patil, Senior Research Scientist, Cotton Research Unit, Dr. PDKV. Akola, Dr. D.V. Durge, Associate Professor and Head of Dept. of Agril Botany, College of Agriculture, Dr. PDKV. Akola, Dr. S.J. Gahukar Associate Professor, Officer Incharge Biotechnology Section, Department of Agril Botany, Dr. PDKV. Akola and Dr S. W. Jahagirdar Associate Professor, Department of Agril.Economics and statistics for their timely help and keen interest in the pursuit of present investigation.

I am grateful to Dr. R.S. Nandanwar Head Of Dept, Department Of Agril Botany, Dr. PDKV. Akola and Prof. D.B. Tamgadge, Associate Dean, PGI, Dr. PDKV, Akola for providing necessary facilities for carrying out present investigation and rendering valuable guidance from time to time.

I also take this opportunity to express my sincere and humble thanks to Dr. D.B. Dhumale, Professor, Department of Agril Botany, Dr. PDKV. Akola, Shri. S.B. Amarshettiwar, Associate Professor, Department of Agril Botany, Dr. PDKV. Akola , Shri. G.R. Shamkuwar, Associate Professor, Department of Agril Botany, Dr. PDKV. Akola, Shri. W.K. Shembekar, Assistant Professor, Department of Agril Botany, Dr. PDKV. Akola, and Shri. M.R. Wandhare, Assistant Professor, Department of Agril Botany, Dr. PDKV. Akola, and all the Staff members of Department of Agril Botany, PGI, Dr. PDKV, Akola for their valuable advice and timely co-operation in numerous ways in which they helped me during the tenure of my course work and present investigation.

I am cheered to place my gratitude to University Librarian Shri. A.B. Bhosale and staff member of computer centre for their help in processing the data for statistical analysis.

Special thanks must be recorded to Umale kaka, Damodar kaka,Dept. of Agril. Botany, PGI, Dr. PDKV, Akola. and Thakare kaka, Pramila Mawashi CRU, Dr. PDKV Akola, for their very valuable co-operation during my research and laboratory work.

I express my heartful thanks to my colleagues Avinash, Meena, Trupti, Vallabh, Amol, Nilesh, Avadhoot for their help and constant encouragement during course my studies.

Its proud fact, I always have support, love, affection, guidance, inspiration, encouragement and feeling that I am not alone, from my friends forever and its not justice to say thank to all them. Yet, I reward my affection to my beloved friends Sachin, Swapnil, Ujwal, Sharad, Sameer dada, Mangesh, Saurabh, Amod, Darshan, Prathamesh, Bhagyashri, Shraddha for their kind help and close cooperation during this journey.

My father Shri. Sanjay Waman Pande and mother Sau. Rekha Sanjay Pande are the supreme souls for me. They not only showed me the world but add the essence of being within me. Their each act and thought is for me and my well being. The exact thing to say thanks to them is only to bow humbly in front of them. They are ultimate reason and cause of my each activity. Not only words but my intellectuality is just insufficient to repay their deeds. My beloved, Smt. Nila Ajji, Smt. Meera Ajji and uncle Shri. Sunil Gopalrao Pande, were always a source of inspiration for me. My small and cute sister Gayatri, who broaden joys in my life and I owed her smile in my life.

I will be failing in my duty if I do not record my gratitude and indebtedness to Deshpande Family and Kawishwar Family, whose encouragement, kind blessing, while traveling on this path of education enable me to accomplish my study. I will never be able to repay their debts.

I am very much thankful to all the authors and researchers, whose articles helped me in organizing my research work in proper line and utilize proper tools for interpretation of the results.

Finally, I express my sense of gratitude towards the farm labours without whom, I could not have completed my research work. As well as I was fortunate enough to receive the kind cooperation from almost everyone in one way or the other during my stay at this institute. It is extremely difficult to thank all of them individually by name this short coming may please be pardoned.

Place : Akola. (Pande Harshal Sanjay)

Date : 14/06/2012. Enrolment No. DD/369

Table of Contents

(A) List of Tables

(B) List of Figures

(B) List of Plates

D) Abbreviations

Abbreviations		Expanded Form
%	-	Per cent
/	-	Per
µg g fresh wt^{-1}	-	microgram per gram fresh weight
µg	-	microgram
µM	-	micromolar
µmoles g fr. wt^{-1} hr^{-1}	-	microgram per gram fresh weight per hour
A.P.	-	Andhra Pradesh
Agric	-	Agriculture
Agril.	-	Agricultural
Agron.	-	Agronomy
C.D.	-	Critical difference
cm	-	Centimeter
cm^2	-	square centimeter
CRU	-	Cotton Research Unit
CSI	-	Chlorophyll stability index
d.f.	-	Degree of freedom
DAG	-	Days After Germinaion
DAS	-	Days after sowing
Dev.	-	Development
et al.	-	et alia (and others)
Fig.	-	Figure
FCRD	-	Factorial Completely Randomized Design
g	-	Gram
G.	-	*Gossypium*
GB	-	Glycine betaine.
ha.	-	Hectare
i.e.	-	id est (that is)
J.	-	Journal
mg	-	milligram
mg/cm^2	-	milligram per square centimeter

mM	-	millimolar
MS	-	Maharashtra State
No.	-	Number
N.S.	-	Non significant
NR	-	Nitrate reductase
PDKV	-	Panjabrao Deshmukh Krishi Vidyapeeth
PGI	-	Post Graduate Institute
PKV	-	Panjabrao Krishi Vidyapeeth
Prof.	-	Professor
Publi.	-	Publication
Res.	-	Research
RWC	-	Relative water content
Sci.	-	Science
SCY	-	Seed cotton yield
SE (m$\pm$)	-	standard error of mean
SLW	-	Specific leaf weight
Sr. No.	-	Serial number
USA	-	United State of America
Viz.,	-	Videlicet (Namely)
Wt.	-	Weight
w/w	-	weight by weight
w/v	-	weight by volume

F) **THESIS ABSTRACT**

a. Title of the thesis	: **"Effect of exogenous application of Glycine betaine on cotton (*Gossypium hirsutum* L.) under moisture stress"**
b. Name of student	: Pande Harshal Sanjay
c. Name and Address of Major Advisor	: Dr. T.H. Rathod Associate Professor, Cotton Research Unit, Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola (MS) 444104.
d. Degree to be awarded	: M.Sc. (Agri.)
e. Year of award of degree	: 2012
f. Major subject	: Agricultural Botany (PlantPhysiology)
g. Total number of pages in the thesis	: 80
h. Total number of words in thesis abstract	: 461
i. Signature of the student	:
j. Signature, name and address of forwarding authority	:

Head,
Department of Agril. Botany,
PGI, Dr. Panjabrao Deshmukh Krishi Vidyapeeth,
Akola (M.S.)

ABSTRACT

An experiment was conducted to investigate "Effect of exogenous application of Glycine betaine on cotton (*Gossypium hirsutum L.*) under moisture stress" on growth, yield and to asses physio-chemical parameters indicating moisture stress tolerance in cotton. Experiment was conducted at the experimental field of CRU,

PGI, Dr. PDKV, Akola, during *Kharif* season of 2011-12. The experiment was laid out in FCRD, with 18 treatment combinations comprising different levels of two factors. Factor A consists two Bt-cotton cultivars **V_1** (Bramha BG I) and **V_2** (Mallika BG I) and nine different levels of Factor B, i.e. **S_1** (control) and eight treatments of Glycine betaine **S_2** [2.5% (w/w) GB seed treatment], **S_3** [5% (w/w) GB seed treatment], **S_4** [1% (w/v) GB foliar spray at 15 DAG], **S_5** [2% (w/v) GB foliar spray at 15 DAG], **S_6** [2.5% (w/w) GB seed treatment + 1% (w/v) GB foliar spray at 15 DAG], **S_7** [2.5% (w/w) GB seed treatment + 2 % (w/v) GB foliar spray at 15 DAG], **S_8** [5% (w/w) GB seed treatment + 1% (w/v) GB foliar spray at 15 DAG], **S_9** [5% (w/w) GB seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 DAG]. The experiment was conducted under open shade conditions. The pot [polythene bag (90cm x 60cm x 60cm)] culture method was used. Two identical sets (one for recording morphological and biochemical observations and another for phonological parameters, yield and its desirable traits) was arranged, comprising of all treatments and three replications each. After 15 DAG, moisture stress was maintained by providing irrigation to the pots (polythene bags) by the interval of 21 days.

The parameters like leaf area (1334.12 cm^2), SLW (3.536 mg/cm^2), Dry matter accumulation (53.571 g/plant), CSI (68.437), RWC (83.13%), Proline content (54.78µg/g), NR activity (1.8061 µmoles/g/hr), Seed cotton yield (28.86 g/plant) and Harvest index (37.70 %) maintained the higher values than control in respect of factor B (treatment S_9) They might be associated with the moisture stress tolerance in cotton. The exogenous application of Glycine betaine successfully averted the negative effects of moisture stress on growth and yield of cotton. The Glycine betaine treatment S_9 found superior amongst all the treatments and recorded high harvest index (37.70 %) which might be resulted into elevating the negative effects of moister stress as compared to control (19.43 %). The Variety V_2 (Mallika BG I) found promising than V_1 (Bramha BG I) under moisture stress. V_2 (19.48 g/plant) recorded higher seed cotton yield as compared to V_1 (18.56 g/plant). There is a need to standardize the dosage and concentration of Glycine betaine along with the stage of spray and number of sprays required for elevating moisture stress in cotton. However, the data recorded implies only one season crop growth which needs further confirmation on multilocation basis.

CHAPTER I

INTRODUCTION

1.1 Background Information

Cotton is a soft, fluffy, staple fibre that grows in a boll, or protective capsule, around the seeds of cotton plants of the genus *Gossypium*. The plant is a shrub native to tropical and subtropical regions around the world, including the Americas, Africa, India and Pakistan. The fibre most often is spun into yarn or thread and used to make a soft, breathable textile, which is most widely used natural fibre cloth in clothing today. The botanical purpose of cotton is to aid in seed dispersal.

According to the Foods and nutrition Encyclopedia, the earliest cultivation of cotton discovered thus for in the Americas occurred in Mexico, some 8000 years ago. The indigenous species was *Gossypium hirsutum*, which is today the most widely planted species of cotton in the world constituting about 89.9% of production worldwide. The greatest diversity of wild cotton species is found in Mexico, followed by Australia and Africa.

Cotton is used to make a number of textile products. These include terrycloth for highly absorbent bath towels and robes; denim for blue jeans; chambray, popularly used in the manufacture of blue work shirts (from which we get the term "blue-collar"); and corduroy, seersucker, and cotton twill. Socks, underwear, and most T-shirts are made from cotton. Bed sheets often are made from cotton. Cotton also is used to make yarn used in crochet and knitting. Fabric also can be made from recycled or recovered cotton that otherwise would be thrown away during the spinning, weaving, or cutting process. While many fabrics are made completely of cotton, some materials blend cotton with other fibers, including rayon and synthetic fibers such as polyester. It can either be used in knitted or woven fabrics, as it can be blended with elastine to make a stretchier thread for knitted fabrics, and apparel such as stretch jeans. In addition to the textile industry, cotton is used in fishnets, coffee filters, tents, gunpowder (nitrocellulose), cotton paper, and in bookbinding. The first Chinese paper was made of cotton fiber.

The cottonseed which remains after the cotton is ginned and used to produce cottonseed oil, which, after refining, can be consumed by humans like any other vegetable oil. The cottonseed meal that is left generally is fed to ruminant livestock; the gossypol remaining in the meal is toxic to monogastric animals. Cottonseed hulls can be added to dairy cattle rations for roughage. During the American slavery period, cotton root bark was used in folk remedies as an abortifacient, that is, to induce a miscarriage.

Cotton linters are fine, silky fibers which adhere to the seeds of the cotton plant after ginning. These curly fibers typically are less than 1/8" (3 mm) long. The term also may apply to the longer textile fiber staple lint as well as the shorter fuzzy fibers from some upland species. Linters are traditionally used in the manufacture of paper and as a raw material in the manufacture of cellulose. In the UK, linters are referred to as "cotton wool". This can also be a refined product (*absorbent cotton* in U.S. usage) which has medical, cosmetic and many other practical uses. The first medical use of cotton wool was by Dr. Joseph Sampson Gamgee at the Queen's Hospital (later the General Hospital) in Birmingham, England.

The five leading exporters of cotton in 2009 are (1) the United States, (2) India, (3) Uzbekistan, (4) Brazil, and (5) Pakistan. The largest non producing importers are Korea, Russia, Taiwan, Japan, and Hong Kong.

India is second largest producer as well as consumer and third largest exporter of the Cotton in the world. This accounts for about 10% of US Cotton-dominant imports. India is, however, overshadowed by China, which is the world's largest producer, consumer and importer of Cotton. A major part of the raw Cotton is used in textile industry, which is second largest industry in India, employing over 35 million people. This fast moving industry has given a boost to the domestic market and consumer spending on apparels has increased.

During 2011-12 in India area under cotton was 121.91 lakh ha, with production of 345 lakh bales and productivity 481.23 lint Kg/ ha. In Maharashtra, area was 40.95 lakh ha with production 69 lakh bales and productivity 280 lint kg/ha. In Vidharbha area was 15.6 lakh ha with production of 35.5 lakh bales and productivity 228 lint Kg/ ha. (Anonymous 2012).

Above 70 percent of cotton growing area in India is under rainfed condition, which is characterized by uneven rainfall coupled with moisture stress, inadequate fertilizer application and plant protection measures. In rainfed condition, cotton crop frequently encounters variety of stresses that adversely affects growth, development and productivity. Stresses can be biotic, imposed by other organism or abiotic, excess or deficit in the physical or chemical environment. Among the environmental conditions, drought or moisture stress is the major constraint to the cotton production. There is an urgent need to concentrate on the research to overcome abiotic stresses causing limitations to increase the yield.

1.2 Importance and need of study

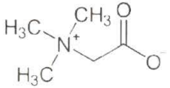
In recent years lint yields of cotton production belt have become stagnant with little or no improvement as newer varieties have been used. The impact is not only on the lint yield but also on the all of components that undergoes in to the development of the yield. In addition to yield stagnation problem, there has been considerably instability in lint yield and fibre quality for some of the newer cotton cultivars currently in production. The phenomenon co-insides with the increased use of transgenic cottons (Bt gene, glyphosate resistant gene, Bt stacked) and weather patterns that have been hotter and dryer than normal (Pettigrew, 2004_a). The introduction of transgenic cotton varieties into large scale production has occurred within the last decade. The possibility of the uncertainty of gene insertion position for a given transformation event utilizing current transformation technologies, DNA could be inserted in to chromosomal region containing a gene and there by disrupt a gene function. This possibility, combined with the current cotton yield stagnation and instability problems, has lead to the speculation of transgenic cotton lines are more sensitive to abiotic stresses (Pettigrew, 2004_b)

Plant scientists are making concerted efforts in identifying genotypes with better drought tolerance coupled with relatively higher yield. The progress in developing crop cultivars for tolerance to abiotic stresses particularly drought has been slow, because of lack of knowledge of mechanisms of tolerance, poor understanding of inheritance of tolerance, low heritability and lack of efficient techniques for screening germplasm (Kush, 1998).

Plant morphological features and biochemical parameters have been useful tools routinely used in understanding the mechanism of drought tolerance and its relation to growth and development of crops (Weising et al., 1995). The mechanism of drought tolerance depends upon the species, genotypes and the development stage of plant. A strategy that tolerant plant often uses to overcome the water deficit is the accumulation of solutes (osmotic adjustment) in the cell help to maintain plant water status, particularly under drought (Cassandra and Oosterhius, 1999). Accumulation of osmoprotectants in higher plants and other organisms is a well known phenomenon representing metabolic adaptation to salinity, drought and high temperature stress (Wyn Jones *et al.*, 1984).

Osmoprotectants are small molecules that can benefit osmotically stressed cells in two ways i.e., by acting as nontoxic cytoplasmic osmolytes to raise osmotic pressure and by protecting enzymes and membranes against damage by salt levels (Wyn Jones *et* *al.* 1984). These osmolytes include sugar and sugar alcohols (polyols), manitol, proline, cholinage, glycine, a number of quaternary ammonium compounds (betaines) and tertiary ammonium compounds (Roy and Basu, 2009).

Glycine betaine [$(CH_3)_3N^+$-CH_2-COO^-] is one of the major osmolyte that accumulates in variety of plant species in response to environmental stresses such as drought, salinity, extreme temperature, U.V. radiations and heavy metals. Many studies have indicated a positive relationship between accumulation of glycine betaine and plant stress tolerance (Ashraf and Foolad, 2007).



Glycine betaine

Glycine betaine is abundantly present in chloroplast where it plays a vital role in adjustment and protection of thylakoid membrane, thereby maintaining photosynthetic efficiency (Robinson and Jones, 1986; Genard et al., 1991). In higher plants, glycine betaine is synthesized in chloroplast from serine via ethanolamine, choline, and betaine aldehyde (Hanson and Scott, 1980; Rhodes and Hanson, 1993). Choline is converted to betaine aldehyde, by choline monooxygenase (CMO), which is then converted to glycine betaine by betaine aldehyde dehydrogenase (BADH). Although other pathways such as direct *N*-methylation of glycine is also known, the pathway from choline to glycine betaine has been identified in all glycine betaine accumulating plant species (Weretilnyk et al., 1989).

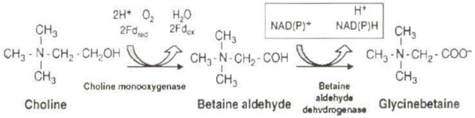


Fig. : Biosynthetic pathway of glycine betaine in higher plants.

1.3 Objectives

1. To assess physio-chemical parameters indicating moisture stress tolerance in cotton.
2. To find out suitable concentration of Glycine betaine for elevating moisture stress in cotton
3. To study the effect of exogenous application of Glycine betaine on growth and yield of cotton.

1.4 Hypothesis

The findings of the present investigation might be helpful to minimize the losses due to moisture stress in cotton by application of desirable outcome.

1.5 Scope and limitations of study

Considering the importance of this crop every effort is being made to minimizing the losses due to moisture stress in cotton by using various biochemical aids and agronomical practices.

Recent studies have indicated that glycine betaine foliar spray enhanced osmotic adjustment and improved drought and heat stress tolerance (Cassandra and Oosterhius, 1999). The doses of glycine betaine were finalized under this investigation and it may provide guideline to the cotton growers to adopt suitable remedial measures for reducing loss and sustaining seed cotton yield. However, the time period adopted to carry out the project was bounded, to find out more information about research topic. It will need further multilocation confirmation.

CHAPTER II

REVIEW OF LITERATURE

An exhaustive review of literature is essential in any research endeavor. It makes researcher up to date with the theoretical knowledge and findings of research topic in the field of investigation. Review of past literature makes the researcher aware about the methods, procedures and techniques available and used as well as outcomes of conclusions at the past studies. It provides clues and guidance throughout the research processes.

Abiotic stresses are considered as major factors limiting growth and yield of cotton in semi-arid environment (Simpson, 1981). The yield losses due to abiotic stresses ranged from 5 to 75 per cent depending on time intensity and duration of stress during the crop period. Moisture deficit can depress cotton (*G. hirsutum L.*) lint yield in all cotton production regions (Pettigrew, 2004_b). The effect of stress on growth and yield parameters is through its effect on various physiological and developmental processes. A thorough understanding of effects of drought at different levels is absolutely essential. It becomes essential to give importance for drought management practices to overcome the yield loss (Patil, 2005).

The most cotton physiological drought stress researches has been conducted in arid production regions, growth chambers or green houses. With objective to document the effects of moisture stress on the physiology of cotton grown (Pettigrew, 2004_b), in pots (Singh *et al.*, 1996, Basal *et al.*, 2005, Pawar *et al.*, 2010).

One of the most common responses in plants is over production of different types of compatible organic solutes (Serraj and Sinclair, 2002).However, not all plants accumulates Glycine Betaine in sufficient amounts to help averting adverse effects of abiotic stresses. In some plants increased resistance to abiotic stresses has been achieved by exogenous application of various organic solutes. There are many reports demonstrating positive effects of exogenous application of glycine betaine on plant growth and final crop yield under drought stress. Exogenous application of glycine betaine is an economically feasible approach to counteract adverse effects of environmental stress on crop productivity (Ashraf and Foodlad, 2007).

Considering the negative effects of moisture stress on cotton, research programme "Effect of exogenous application of glycine betaine on cotton (*Gossypium hirsutum L.*) under moisture stress" was undertaken and the following review is presented to highlight the salient features related for research under different subheads.

2.1 Effect of moisture stress and exogenous application of GB on morphological parameters

May and Milthorpe (1962) revealed that growth of plants was not much affected due to small reduction in relative turgidity as it does not greatly influenced the metabolic processes. They also reported that although the water stress leads to reduction in total growth of plants it influences the growth of different organ differently.

Laude (1971) concluded that a reduction in size of plants is generally noted and is termed as stunting or growth retardation. The height of plant is particularly affected due to water stress. He also reported that moderate water stress does reduce vegetative growth and ultimately yield.

Patterson (1989) reported that water stress reduced the plant height, leaf number, leaf area and total dry weight of cotton in comparison with well watered control.

Singh *et al.* (1996) studied the variation in genotypes for biomass accumulation per plant and leaf area per plant in upland cotton (*Gossypium hirsutum L.*) under drought condition pot culture technique was used. Simulated drought was created at flowering and boll development stage by withholding of water. High genotypic variability was found for biomass accumulation and leaf area per plant. He recorded range of biomass accumulation lies between maximum (60.66 g/plant) and lowest (44.18 g/plant) and for leaf area it was recorded maximum (1125.30 cm^2/ plant) to minimum (306.82 cm^2/plant).

Agboma *et al.* (1997) had shown the resultant increase in phytomass and leaf area of soybean by glycine betaine application @ 3 kg ha^{-1} under moisture stress condition.

Gorham *et al.* (1998_a) reported, significant increase in total dry matter production of cotton under water deficit field condition when glycine betaine applied at squaring stage @ 3 or 6 kg/ ha.

Mottaram (1999) reported, resultant increase in plant height (85 to 110 mm) over control with the use of glycine betaine @ 3.5 kg/ha.

Xing and Rajashekar (1999) concluded that the unfavorable effect of water stress on shoot and dry matter accumulation was not affected by the glycine betaine treatment.

Taiz and zeiger (2005) revealed that Decrease in leaf area is an early adaptive response given by the plants to the water deficit.

Farooq *et al.* (2008) showed that, 50 mM, 100 mM and 150 mM Glycine betaine foliar spray at five leaf stage resulted in to increase in dry matter of rice seedlings under water deficit conditions.

Hussain *et al.* (2009) reported that, foliar application of Glycine betaine on sunflower significantly increased biological yield in water deficit condition.

Mahmood *et al.* (2009) reported that pre-sowing seed treatment of 50 mM GB increased shoot fresh biomass and leaf area per plant in wheat under water deficit condition.

Patil *et al.* (2009) studied the effect of exogenous application of glycine betaine @ 0.3% on cotton. Recorded resultant increase in plant height, leaf area index and total dry matter accumulation of cotton plant under drought. He also reported increase in specific leaf weight of cotton by foliar applied Glycine betaine.

2.2 Effect of moisture stress and exogenous application of GB on phenological parameters

Marani (1973) reported that the stress imposed at the end of flowering cause earliness and also increased lint fineness in cotton.

Quisenberry and Roask (1976) concluded that, when timely rainfall does not occur during the growth season, and crop is forced to make its yield on stored moisture results in to early maturity. They also reported positive correlation of early maturity with high yield under water deficit.

Chaudhary and Syed (1978) observed delay in flowering in moisture stressed sorghum they further observed that due to moisture stress, thousand grain weight, fodder yield was reduced as compared to well watered control.

Naidu *et al.* (1995) revealed that, Glycine betaine seed treatment resulted in earlier flowering under rainfed condition in cotton.

Rajeshwari (1995) found that, earliness is associated with drought tolerance in cotton under rainfed condition.

Adarsha *et al.* (2003) reported that earliness appears to have a distinct advantage under rainfed condition. Since early maturity got less exposure to stress compared to late maturity.

Kumar and Kurju (2003) reported that, water deficit at anthesis and during advance stages of plant growth delay in further flowering and seed development.

Thind (2007) observed that, drought may make cotton cut out early or it may extend early growth cycle in plants, depending upon timing of dry periods.

2.3 Effect of moisture stress and exogenous application of GB on biochemical parameters

2.3.1 Effect of moisture stress and exogenous application of GB on Chlorophyll stability index

Rajeshwari (1995) found that, chlorophyll stability index is associated with drought tolerance in cotton under rainfed condition.

Patil *et al.* (2011) reported significant increase in the chlorophyll stability index with glycine betaine foliar spray @ 0.3% (66.12) over control (58.12) under moisture stress condition.

Kar *et al.* (2005) reported that PKV Hy-2 (0.76) and PKV Hy-4 (0.74) has recorded greater chlorophyll stability over cultivar Sabita (0.64) and surya (0.64) under stress conditions.

Patil (2005) reported that chlorophyll stability index was greater at 100 DAS (70.84) followed by 140 DAS (66.12) and 60 DAS (64.29) when plants was subjected to glycine betaine foliar spray @ 0.3% applied to the cotton plants under drought condition.

2.3.2 Effect of moisture stress and exogenous application of GB on Stomatal frequency

Kale (2009) found decrease in stomatal frequency with the increase in growth stages of pigeon pea. To reduce the water stress during water stress condition stomata get closed and stomatal frequency decreased. It also reported that genotypes with less or decreased stomatal frequency indicates higher drought tolerance.

Singh *et al.* (1996) reported that, in *Gossypium hirsutum* L. the range of stomatal frequency at harvest lies in between 26.50 to 54.30 per mm^2.

Xu and Zhou (2008) suggested that, stomatal density increased with decreasing potential under moderate water deficit, but it declines under severe moisture stress.

2.3.3 Effect of moisture stress and exogenous application of GB on Relative water content

Singh (1992) observed r reduction in relative water content in all genotypes of upland cotton from 3rd day of moisture stress. He also reported that, at 6th day of moisture stress relative water content reduced about 40 to 55 %.

Rajeshwari (1995) found that, relative water content is associated with drought tolerance in cotton under rainfed condition.

Singh *et al.* (1996) studied the effect of moisture stress condition on the 10 genotypes of *Gossypium hirsutum* L. in earthen pots found that, the relative water content in leaf of stressed cotton was between (41.80 %) to (61.77 %) at boll development stage.

Xing and Rajashekar (1999) reported that exogenous application of GB @ 10 mM in beans resulted in favorable water status of plants as indicated by a slower decrease in leaf water potential and delayed wilting during water stress as compared to the untreated plants.

Patil *et al.* (2011) has recorded relative water content significantly higher in 0.3% GB spray treatment (60.62%) over control (51.37%).

2.3.4 Effect of moisture stress and exogenous application of GB on Proline content

Patil *et al.* (2011) reported increase in leaf proline content of cotton with Glycine betaine foliar application @ 0.3% (76 µg/g fresh wt.) than control (68 µg/g fresh wt.) under rainfed condition.

Farooq *et al.* (2008) found significant increase in leaf free proline levels by foliar application of Glycine betaine under water deficit sunflower plants. 100 mM spray of glycine betaine recorded highest levels of free proline under moisture stressed sunflower plants.

Hussain *et al.* (2008) reported significant increase in leaf proline content of sunflower with exogenous application of Glycine betaine under water stress.

Pawar *et al.* (2010) noticed increase in leaf proline content with the increase in endogenous Glycine betaine levels of cotton under moisture deficit.

2.3.5 Effect of moisture stress and exogenous application of GB on Nitrate reductase activity

Sung (1981) reported decline in nitrate reductase activity, is different for the two cultivars, even though the enzyme response to the leaf water potential was same. He also reported the curvilinear relationship between nitrate reductase activity and water stress.

Becker and Fock (1986) found severe inhibition of nitrate reductase activity under water stressed maize leaves.

Singh (1992) revealed that, nitrate reductase activity may be used as parameter for drought tolerance in cotton during seedling growth stage under moisture stress condition. He also reported that, nitrate reductase activity associated with relative water content and proline content.

Arora and Gupta (2003) revealed that rapid fluctuations of nitrate reductase activity that occurs when plants are placed in dark or exposed to heat tor drought. They also reported that the fluctuations can be brought about by changes in relative rates of synthesis or activation and breakdown or deactivation.

2.3.5 Effect of moisture stress and exogenous application of GB on Photosynthetic efficiency

Koti (1997) observed that, Glycine betaine treatment increased the Fv/Fm ratio under drought stress which in turn resulted in an increase in photosynthetic rate.

Xing and Rajashekar (1999) reported that, water stress adversely affects the photosynthetic activity of plants indicated by chlorophyll florescence (Fv/Fm). They also reported increase in chlorophyll fluorescence by application of glycine betaine under water stressed bean plants.

Pettigrew (2004_b) observed that, the dark adapted / chlorophyll fluorescence (Fv/Fm) is less in dryland cotton (0.781) than irrigated cotton (0.789). He also reported genotypic variation, Paymaster 1220 BR (0.729) recorded maximum chlorophyll fluorescence (Fv/Fm) whereas, MD 51 ne normal (0.687) recorded lowest among all eight genotypes of *G. hirsutum* L. tested.

2.4 Effect of moisture stress and exogenous application of GB on yield and yield attributes

Shimshi and Marani (1971) found that, reduction in number of bolls per plant and lint yield when water stress is imposed at boll development in cotton.

Marani (1973) concluded that, effect of moisture stress on cotton at different growth stages caused reduction in yield.

Giri and Upadhyay (1980) reported that, sympodial branches and picked bolls per plant are important yield components.

Khorgade and Ekbote (1980) indicated that, number of sympodia per plant is an important factor determining direct influence on yield and suggested that selection based on the number of sympodia, boll number and boll weight may be helpful in evolving high yielding varieties of upland cotton.

Channaveeraiah (1983) indicated that, medium sized, 22 sympodial branches were considered as one of the important characters among other morphological characters for high yield in cotton.

Barketali *et al.,* (1982) noticed high positive correlation between number of sympodial branches and seed cotton yield.

Basu and Bhat (1987) reported a high positive correlation between number of sympodial branches and harvest index in cotton.

Naidu *et al.* (1995) revealed that, Glycine betaine seed treatment resulted in stronger stem improved branching and greater number of bolls per plant which increased yield by 22% over control under rainfed condition in cotton.

Agboma *et al.* (1997) indicated that, exogenously applied glycine betaine (3 kg ha^{-1}) has potential to confer drought tolerance properties of soybean and reduce yield losses associated with water stress. They also reported the positive association of increased seed yield with more large seed size and greater number of seeds filled underwater stress.

Gorham *et al.* (1998_a) reported that, seed cotton yields were highest with 3 to 6 kg/ha glycine betaine applied at squaring. It also increased number of bolls per plant, also had more nodes at maturity than control.

Gorham *et al.* (1998_b) reported that, all glycine betaine treatments produced higher yields and are associated with higher boll numbers.

Mottaram (1999) reported that, the use of glycine betaine under water deficit improves the number of bolls causes increase in yield.

Pettigrew (2004_a) conducted a field experiment on eight diverse genotypes of cotton to study the effects of moisture deficit stress on boll distribution, lint yield and fibre length from 1998 to 2001 under both dry land and irrigated conditions. Irrigations caused to produce more vegetative growth and delay maturity compared to dry land plants. During the years when sufficient moisture deficits occurred, the lint yield of dry land plants was reduced 25%, primarily because of a 19% reduction in number of bolls, while irrigated plants produced more bolls at higher plant nodes and at the more distal positions on the sympodial branches and also produced approximately 2% longer fibre than the dry land plants.

Kar *et al.* (2005) assessed Performance of five cotton hybrids (PKV Hy 2, PKV Hy 4, Sabita, Surya and Ankur), and the control cultivar (MCU-5) for seed cotton yield under drought conditions at Bhubaneswar, Orissa, India. The study revealed that moisture deficit adversely affected yield performance. The yield decreased conspicuously in all the varieties in response to water stress imposed at flowering stage. Amongst the cotton hybrids, PKV Hy 4 and PKV Hy 2 were noted to be relatively more drought tolerant than others, based on the assessment of their yield performance.

Sarwar *et al.* (2006) found positive relation between Glycine betaine accumulation and seed cotton yield of cotton under water deficit condition. Whereas, correlation between boll weight and boll number with Glycine betaine accumulation also found positive under water deficit.

Patil *et al.* (2009) studied the effect of exogenous application of glycine betaine @ 0.3% on cotton. Recorded resultant increase in number of monopodia, number of sympodia, number of bolls per plant and seed cotton yield per plant over control under drought condition. It also reported genotypic variation amongst all the genotype tested for above mentioned parameters.

Hussain *et al.* (2008) found that, foliar application of Glycine betaine averted negative effect of drought on yield of sunflower. It also increased 1000 achene weight in sunflower under water stress condition.

Hussain *et al.* (2010) reported that, foliar applied 100 mM Glycine betaine recorded significant increase in test weight of sunflower over control.

2.5 Effect of moisture stress and exogenous application of GB on fibre properties

Marani (1973) reported that, the stress imposed at the end of flowering increased lint fineness in cotton.

Karademir et al. (2011) found that, fibre properties like fibre strength, fibre length, fibre fineness and fibre elongation were negatively affected, whereas, fibre elongation was not affected by water stress in cotton.

CHAPTER III

MATERIAL AND METHODS

An experiment was conducted to investigate "Effect of exogenous application of Glycine Betaine on cotton (*Gossypium hirsutum L.*) under moisture stress"

Details of the materials used and the methods adopted during the course of investigations are explained here under.

3.1 Experimental details and technical programme

3.1.1 Material

The experimental material consisted of two different cultivars of Bt cotton, which was procured from Senior Research Scientist (Cotton), Cotton research Unit, Dr. PDKV, Akola (M.S.). Glycine betaine was provided by Department of Agricultural botany, Dr. PDKV, Akola (M.S.).

3.1.2 Location

The investigation was carried out at the experimental site of Cotton Research Unit, Central Research Station, Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola located at 304.415 meter altitude, $20^{O}31'$N latitude and 72^{O} 02 longitude during *kharif* season of 2011-2012.

3.1.3 Experimental details

The experiment was conducted under open shade conditions. The pot (polythene bag) culture was used. Each pot (polythene bag) was filled by 20 kg pot mixture i.e. Soil + FYM + Sand (4:2:1). Two identical sets (one for recording morphological and biochemical observations and another for phonological parameters, yield and its desirable traits) was arranged, comprising of all treatments and three replications each. The weather data for the year 2011-2012 is enclosed as Appendix.

Crop	:	Cotton (*Gossypium hirsutum* L.)
Experimental design	:	Factorial Completely Randomized Design
Number of set (s)	:	Two (2)
No of replication (s)	:	Three (3) (in each set)
Number of treatments	:	Eighteen (18)
Treatment details	:	**Factor 'A': Variety**

V_1 : Bramha BG (I)

V_2 : Mallika BG (I)

Factor 'B': Concentation of glycine betaine

S_1: Control.

S_2: 2.5% (w/w) Glycine betaine seed treatment.

S_3: 5% (w/w) Glycine betaine seed treatment.

S_4: 1% (w/v) Glycine betaine foliar spray at 15 days after germination.

S_5: 2% (w/v) Glycine betaine foliar spray at 15 days after germination.

S_6 : 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.

S_7 : 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.

S_8 : 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.

S_9: 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.

Treatment combinations	:	$V_1 S_1$	$V_2 S_1$
		$V_1 S_2$	$V_2 S_2$
		$V_1 S_3$	$V_2 S_3$
		$V_1 S_4$	$V_2 S_4$
		$V_1 S_5$	$V_2 S_5$
		$V_1 S_6$	$V_2 S_6$
		$V_1 S_7$	$V_2 S_7$
		$V_1 S_8$	$V_2 S_8$
		$V_1 S_9$	$V_2 S_9$
Method of sowing	:	Dibbling	
Pot size (polythene bag)	:	90cm x 60cm x 60cm	
Date of sowing	:	12^{th} July, 2011	

Glycine Betaine treatment

Each pot (polythene bag) was sown by five seeds. As per treatment details Glycine Betaine seed treatment was done before sowing, and accordingly foliar spray was applied to the plants at the time of initiation of stress (15 days after germination). Carboxymethyl cellulose (5% solution) was used as a sticking agent for seed treatment, whereas Tween-20 (0.01% solution) was used as a surfactant for foliar spray.

Irrigation

For first 15 days after germination pots (polythene bags) were irrigated daily and the irrigation level was maintained at field capacity. After 15 DAG, moisture stress was maintained by providing irrigation to the pots (polythene bags) by the interval of 21 days. At the time of irrigation the pots (polythene bags) are irrigated up to the field capacity. However, the polythene bags were punctured at lower periphery to improve the drainage.

Plant protection

Drenching of soil in pots (polythene bags) by COC @ 2% and Carbandenzim @ 1.5% as per recommendation. Foliar spray of Dimethoate @ 2% carried out for the control of sucking pest as per pest as per recommendation.

Plan of Experimental Layout

3.2 Observations recorded

3.2.1 Morphological observations

3.2.1.1 Plant height (cm)

Observations on plant height were recorded in centimeter at squaring, 50% flowering, 50% boll bursting and at harvest stage. The height of the all five plants from each replication and treatments was recorded in centimeter with scale from the base of a plant to top most developing node. From these 5 plants per treatment and replications mean height was calculated and recorded.

3.2.1.2 Leaf area (cm^2)

Leaf area (cm^2) was recorded at squaring, 50% flowering, 50% boll bursting and at harvest stage. All leaves from an observational plant are detached and scanned with the help of instrument "CI - 202 automatic leaf area meter, CID (INC), USA." The summation of the values for each treatment in each replication were done and recorded.

3.2.1.3 Specific leaf weight (g/cm^2)

The specific leaf weight (g/cm^2) for each treatment in each replication at squaring, 50% flowering, 50% boll bursting and at harvest stage was worked out by using following formulae.

$$SLW = \frac{\text{Leaf dry weight (g/plant)}}{\text{Leaf area (cm}^2\text{/plant)}}$$

3.2.1.4 Dry matter production per plant (g/ plant)

For dry matter study, single plant from each treatment and replication was uprooted periodically at squaring, 50% flowering, 50% boll bursting and harvest stage. The plant samples were washed with the tap water carefully in order to remove soil and dust particles adhered to it. The samples were allowed to dry at room temperature separately. The plant samples were then placed in the big size brown paper perforated bags. After drying the sample on open air basis the plant sample was finally dried in hot air oven at 70°C to get the constant weight. The plant samples were kept in oven upto achieving the constant weight of the samples. The total dry matter was recorded as gram/plant.

3.2.2 Phenological observations

3.2.2.1 Days to 50% flowering

Data of the flowering of 50 per cent of plants in each treatment per replication was recorded and average was carried out.

3.2.2.2 Days to 50% boll bursting

Data of boll bursting of the 50 per cent of plants in each treatment were recorded and days were calculated.

3.2.2.3 Days to maturity

The days required for maturity was counted from days of emergence and the mean was noted treatment and replication wise.

3.2.3 Biochemical observations

3.2.3.1 Chlorophyll stability index

Chlorophyll stability index was recorded at squaring, 50% flowering, 50% boll bursting stages of plant growth. This method is based on pigment changes induced by heating. The chlorophyll destruction commences rapidly at critical temperature of 55-56°C. For measuring Chlorophyll Stability Index (CSI), third fully opened leaf from the top was used.

Leaf samples of 0.2 g were collected in two sets. One set was placed in the test tubes containing 50 ml of distilled water and these tubes were kept in hot water, both maintained at 56°C for 30 minutes. Another set was kept at room temperature to serve as control. The leaf samples were then incubated in 7.0 ml Dimethyl sulphoxide (DMSO) at 65°C for 30 minutes. At the end of the incubation period, supernatant was decanted and the volume was made upto 10.0 ml with DMSO. The absorbance of the extract was read at 645 and 663nm in a spectrophotometer using DMSO as blank. The chlorophyll content was calculated by using the following formula and expressed in mg per g fresh weight.

$$\text{Total chlorophyll} = 20.2\,(A645) + 8.02\,(A663) \times \frac{V}{1000 \times W \times a}\ (\text{mg/g fresh wt.})$$

Where,

A = Absorbance at specific wave length (645 and 663 nm)

V = Final volume of the chlorophyll extract (ml)

W = Fresh weight of the sample (g)

a = Path length of light (1 cm)

From this, CSI was calculated by the formula as given below

$$CSI = \frac{CS}{CC} x100$$

Where,

CS = Total chlorophyll content of stressed plant (mg/g fresh wt.)

CC = Total chlorophyll content of control plant (mg/g fresh wt.)

3.2.3.2 Stomatal frequency

Stomatal frequency was recorded at squaring, 50% flowering, 50% boll bursting stages of plant growth. For stomatal frequency, stage micrometer on the stage of the microscope was mount. Coincides the stage divisions with a square that was in the eyepiece and count like divisions for the length and breadth of a known the area of square. Count the number of stomata within the area of square. Computed the stomatal frequency in mm^2 per unit of area with the help of scanned area and stomatal number (Dhopte and Livera, 1989).

3.2.3.3 Relative water content (%)

Relative water content was recorded at squaring, 50% flowering, 50% boll bursting stages of plant growth. Relative water content of leaf gives an idea of water retaining ability of plant in leaf. Third leaf from top was selected from each treatment and replication. The leaves were labeled and were placed in polythene bags separately to avoid water loss. Leaves were weighted on electronic balance. Their fresh weight was taken and then transferred to petridish containing water. The leaves were kept floating on water for 4 hrs and after that their weight was recorded. The weight of leaves was recorded after keeping it in hot air oven at $70^{O}C$ (Barrs and Weatherley, 1962).

Relative water content was calculated with the equation of

$$RWC(\%) = \frac{\text{Fresh weight - oven dry weight}}{\text{Turgid weight - oven dry weight}} x100$$

3.2.3.4 Proline (µg g fresh wt.$^{-1}$)

Proline content from the leaf tissues of each treatment from each replication were estimated at squaring, 50% flowering, 50% boll bursting stage, following is the method as suggested by Bates *et al.* (1973). The leaf sample of 0.5 g was homogenized in 10 ml of 3 percent sulphosalicylic acid. The homogenate was filtered through a double layered filter paper. A 2 ml of the filtrate was taken in a test tube to which 2 ml of acid ninhydrin reagent (2.5 g of ninhydrin was dissolved in 40 ml of 6M orthophosphoric acid and 60 ml of glacial acetic acid), 2 ml of glacial acetic acid was added. The test tubes containing the mixture were placed in boiling water bath for one hour. The test tubes were then cooled by keeping them in an ice bath. The contents were transferred to a separating funnel and 4 ml of toluene was added and mixed vigorously. The coloured toluene fraction was separated and measured at 520 nm in a spectrophotometer. A blank was maintained with all the reactants except the leaf extract. Proline content in leaf tissue was calculated by using the formula:

$$\text{Proline}\ (\mu\mu \text{ g fresh wt}^{-1}) = \frac{34.11 x OD520 x V}{2 x f}$$

Where,

V = Total volume of extract

f = Grams of fresh leaf

2 = Volume of extract taken

3.2.3.5 Nitrate reductase activity (μ moles of nitrite formed g^{-1} hr^{-1})

In vivo nitrate reductase activity from the leaf tissues of each treatment from each replication were estimated at squaring, 50% flowering, 50% boll bursting stage, was determined following the method of Srivastava (1974). 0.3 g fresh material was cut into small pieces and put in 5 ml tubes filled with incubation medium (0.1 M phosphate buffer at 7.5 pH and 200 mM KNO3 and 0.5% n-propanol) and left in dark for two hours at 30° C. Thereafter, to 1 ml aliquot, 1 ml sulfanilamide (1% in 3N HCl) and 1 ml 0.02% naphthyl-ethylene-diamine-hydrochloride (NNED-HCL) was added and shaken thoroughly. After keeping for twenty five minutes for colour development absorbance was read at 540 nm. NR activity was calculated employing standard curve of nitrate and expressed in μ moles of nitrite formed g^{-1} of fresh weight hr^{-1} basis.

3.2.3.6 Photosynthetic efficiency

The photosynthetic efficiency (Fv/Fm) was measured by recording the chlorophyll flurosence value by using the instrument "Chlorophyll Flurometer (OS-30p Optisciences, USA)". It was recorded at squaring, 50% flowering, 50% boll bursting stages of plant growth.

Determination of dark adaptation time

For determination of dark adaptation time maximum dark period was measured by placing 20 clips on the third leaf of 20 cotton plants. Shutter plates were closed on leaves as such. The photosynthetic efficiency (ratio of Fv/Fm) was recorded at every 2 min time interval (100 percent light level was used).

Measurement of photosynthetic efficiency

For cotton crop 12 min maximum dark period was recorded. After that the photosynthetic efficiency of third leaf was measured by giving 12 min dark adaptation time period. After 12 min the sensor unit was brought over leaf clip and fitted so that the day light was excluded. Then a shutter plate in leaf clip is open to expose the leaf ready for illumination and measurement by sensor unit. Run the instrument and record the observation of Fv/Fm. photosynthetic efficiency was recorded on three plants per treatment replication wise and mean values were calculated.

3.2.4 Yield and yield attributes

3.2.4.1 Number of monopodia per plant

The monopodial branches bearing at least one functional sympodial branch were counted separately on each plant and the average value was recorded at the time of harvest.

3.2.4.2 Number of sympodia per plant

Fruiting branches arising from the main stem were counted separately on each plant at harvest and average value was recorded.

3.2.4.3 Number of bolls per plant

From the initiation of first picking the number of bolls per plant was counted and recorded upto last picking and the summation of number of bolls at each picking gave number of bolls per plant.

3.2.4.4 Single boll weight (g)

The weight of single boll (g) was calculated by taking the mean of five bolls weight (g).

3.2.4.5 Boll weight per plant

Number of bolls harvested from all five observational plants under each treatment and replication at all picking were weighted to calculate mean boll weight (g).

3.2.4.6 Seed cotton yield per plant (g)

The seed cotton obtained from all five plants from each treatment and replication were weighted separately and mean seed cotton yield (g) was calculated and recorded.

3.2.4.7 Test weight (g)

The test weight of seed were recorded by weighing 100 seeds from each treatment in each replication.

3.2.4.8 Harvest index (%)

Harvest index (%) was computed as per equation given by Donald (1962).

$$\text{Harvest Index (\%)} = \frac{\text{Economic yield. (g) / plant}}{\text{Biological yield (g) / plant}} \times 100$$

3.2.5 Fibre properties

3.2.5.1 2.5% Span length (mm)

2.5 per cent span length is the distance from the test specimen clamp line up to which 2.5 per cent fibres extended in the test beared of equipment. The span length of each treatment was measured in 'mm' by using 'Digital Fibro-graph 530'.

Test specimen was prepared by using fibro sampler and fibre comb, then loaded fibro comb was placed in comb holder of equipment and brushed to remove loose fibers and strengthen the fibres. The lens house was lowered and 2.5% span length was directly measured.

3.2.5.2 Uniformity ratio

Uniformity ratio is the ratio of 50 per cent span length to 2.5 per cent span length.

3.2.5.3 Fibre strength (g/tex)

It is the force required to break of fibre of unit linear density. Fibre strength was determined by using HVI. It was expressed in grams per tex.

3.2.5.4 Elongation (%)

Fibre elongation was determined by using HVI. It is expressed in terms of percentage. Also called as a percent elongation.

3.2.5.4. Fibre fineness (µg/inch)

It is the average weight per unit length of fibre. It is used in determining the fibre fineness. Linear density of fibre is expressed in micrograms per inch.

3.3 Statistical analysis

The data collected on various observations, during the course of investigation were statistically analyzed by Factorial

Completely Randomized Design (FCRD) as suggested by Panse and Sukhatme (1967). The 'F' tests whenever revealed significant, the critical difference were worked out at 5 per cent level of significance for treatment comparisons. The analysis was carried out at Computer Centre, Directorate of Research, Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola.

Analysis of variance

Source of Variation	d.f.	Sum of squares	Expected mean sum of squares	Calculated 'F'	Tabulated 'F'
Factor A (Varieties)					
Factor B (Glycine betaine treatment)					
Interaction (A x B)					
Error					
Total					

CHAPTER IV

RESULTS AND DISCUSSION

An experiment was conducted to investigate "Effect of exogenous application of Glycine betaine on cotton (*Gossypium hirsutum L.*) under moisture stress" was carried out at the experimental field of Cotton Research Unit, Post Graduate Institute, Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola during *Kharif* season of 2011-12 with the following objectives.

1 To assess physio-chemical parameters indicating moisture stress tolerance in cotton.
2 To find out suitable concentration of Glycine betaine for elevating moisture stress in cotton
3 To study the effect of exogenous application of Glycine betaine on growth and yield of cotton.

The observations were recorded on various aspects viz., morphological, phenological and biochemical parameters, yield and yield attributing characters and fibre quality. The observations recorded during the course of investigation are presented and discussed in this chapter under appropriate headings and sub headings.

4.1 Morphological parameters

4.1.1 Plant height (cm)

Plant height is an important morphological character in cotton which provides seat for nodes and internodes from where monopodial and sympodial branches emerges, and thus play an important role in determining the morphological framework relating to productivity (Eaton, 1955).

The data on plant height were recorded at different growth stages as influenced by exogenous application of Glycine betaine is presented in Table 1 and depicted in Fig. 1. An increase in plant height of cotton throughout growth period (up to maturity) of crop was recorded under moisture stress during the course of observation.

Table 1. Effect of exogenous application of Glycine betaine on Plant height of cotton under moisture stress.

Treatment(s)	Plant Height (cm)			
	Square initiation	50% flowering	50% boll bursting	Maturity
FACTOR "A" (VARIETY)				
V_1 - Bramha BG (I)	20.63	64.74	85.28	85.77
V_2 - Mallika BG (I)	20.77	65.12	85.71	86.03
'F' test	NS	NS	NS	NS
SE(m)±	0.07	0.18	0.21	0.20
CD at 5%	-	-	-	-
FACTOR "B" Glycine betaine Treatment(s)				
S_1 – Control	15.85	51.24	70.06	70.32
S_2 -2.5% (w/w) Glycine betaine seed treatment.	17.58	54.68	75.31	75.65
S_3 - 5% (w/w) Glycine betaine seed treatment.	18.39	58.11	78.74	79.12
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	19.20	60.28	80.91	81.28
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	19.83	62.66	83.29	83.76
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	22.09	68.92	89.55	90.05
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	24.41	75.85	96.65	96.95
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	23.62	74.40	95.03	95.53
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	25.33	78.21	99.92	100.47
'F' test	Sig	Sig	Sig	Sig
SE(m)±	0.14	0.39	0.44	0.43
CD at 5%	0.40	1.12	1.26	1.24
INTERACTION "AXB"				
'F' test	NS	NS	NS	NS
SE(m)±	0.20	0.55	0.62	0.61
CD at 5%	-	-	-	-

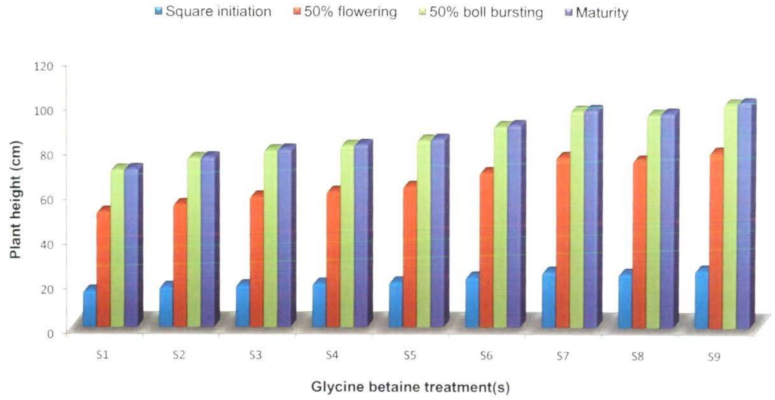


Fig.1: Effect of exogenous application of Glycine betaine on Plant height of cotton under moisture stress.

At square initiation stage, in case of factor A statistical difference for plant height found non significant. While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (25.33 cm) recorded significantly maximum plant height among all the treatments followed by S_7 (24.41 cm), S_8 (23.62 cm) and S_6 (22.09 cm), whereas, minimum plant height was observed in S_1 (15.85 cm). However, the interaction effect between varieties and treatments found non significant at this stage of plant growth.

At 50 percent flowering stage, in case of factor A statistical difference between plant height found non significant. While in case of factor B, all treatments differed significantly for plant height at this stage. The treatment S_9 (78.21cm) recorded significantly maximum plant height among all the treatments, followed by S_7 (75.85 cm), S_8 (74.40 cm) and S_6 (68.92 cm), whereas, least plant height was observed in S_1 (51.24 cm). However, the interaction effect between varieties and treatments found non significant at this stage of plant growth.

At 50 percent boll bursting stage, in case of factor A statistical difference for plant height found non significant. While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (99.92cm) recorded significantly maximum plant height among all the treatments followed by S_7 (96.65 cm), S_8 (95.03 cm) and S_6 (89.55 cm), whereas, least plant height was observed in S_1 (70.06 cm). However, the interaction effect between varieties and treatments again found non significant at this stage of plant growth.

At maturity, cotton plant recorded its maximum plant height, but the per cent increase was very less as compared to other growth stages. It might be due to the fact that vegetative growth during this phase gets ceased due to carbohydrate demand by bolls. It results into stoppage of initiation and elongation of main stem nodes and internodes (Thind, 2007). In case of factor A statistical difference for plant height found non significant. While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (100.47) recorded significantly maximum plant height among all the treatments followed by S_7 (96.95 cm), S_8 (95.53 cm) and S_6 (90.05 cm), whereas, least plant height was observed in S_1 (70.32 cm). However, the interaction effect between varieties and treatments found non significant at this stage of plant growth.

Greater plant height is related to better yield and drought tolerance in cotton (Shakoor, 2010). The resultant increase in plant height in cotton plants under moisture stress with exogenously applied Glycine betaine may be the result of maintenance of photosynthetic activity owing to Glycine betaine application by means of osmotic adjustment. The organelles and cytoplasmic activities takes place at about a normal pace and help plant to perform better in terms of growth, photosynthesis and assimilate partitioning (Hussain *et al.*, 2009). These findings are similar to the findings of Mottaram (1999) and Patil *et al.* (2009) in cotton.

4.1.2 Leaf area (cm^2)

Leaf area per plant depends upon the number and size of leaves beared by plant. The leaf size is influenced by moisture. Leaf area is one of the essential factors for determining the dry matter production (Shewale, 2007).

The data on leaf area were recorded at different growth stages as influenced by exogenous application of Glycine betaine is presented in Table 2 and depicted in Fig. 2. There was an increase in leaf area up to 50 percent boll bursting and picked off then after.

At square initiation stage, in case of factor A i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in leaf area. V_2 (387.86 cm^2) found to be superior over V_1 (385.97 cm^2). While in case of factor B, the statistical difference found significant. The treatment S_9 (412.15 cm^2) recorded significantly maximum leaf area amongst all the treatments followed by S_7 (404.73 cm^2), S_8 (399.74 cm^2) and S_6 (395.90 cm^2), whereas, lowest leaf area was recorded in S_1 (358.72 cm^2) the control. However, the interaction effect between varieties and treatments found non significant.

Table 2. Effect of exogenous application of Glycine betaine on leaf area per plant of cotton under moisture stress.

Treatment(s)	Leaf area (cm^2)			
	Square initiation	50% flowering	50% boll bursting	Maturity
FACTOR "A" (VARIETY)				
V_1 - Bramha BG (I)	385.97	802.43	1191.41	1141.78
V_2 - Mallika BG (I)	387.86	805.98	1196.49	1146.66
'F' test	Sig	Sig	Sig	Sig
SE(m)±	0.18	0.37	0.55	0.53
CD at 5%	0.52	1.06	1.59	1.52
FACTOR "B" Glycine betaine Treatment(s)				
S_1 – Control	358.72	742.56	965.32	925.12
S_2 -2.5% (w/w) Glycine betaine seed treatment.	371.09	757.03	1074.98	1030.20
S_3 - 5% (w/w) Glycine betaine seed treatment.	374.74	783.20	1135.64	1088.34
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	380.62	791.76	1179.62	1130.48
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	384.56	807.58	1212.50	1161.99
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	395.90	819.52	1237.48	1185.93
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	404.73	845.52	1305.19	1250.83
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	399.74	835.45	1300.71	1246.53
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	412.15	855.21	1334.12	1278.56
'F' test	Sig	Sig	Sig	Sig
SE(m)±	0.38	0.79	1.18	1.13
CD at 5%	1.10	2.25	3.37	3.23
INTERACTION "AXB"				
'F' test	NS	NS	NS	NS
SE(m)±	0.54	1.11	1.66	1.59
CD at 5%	-	-	-	-

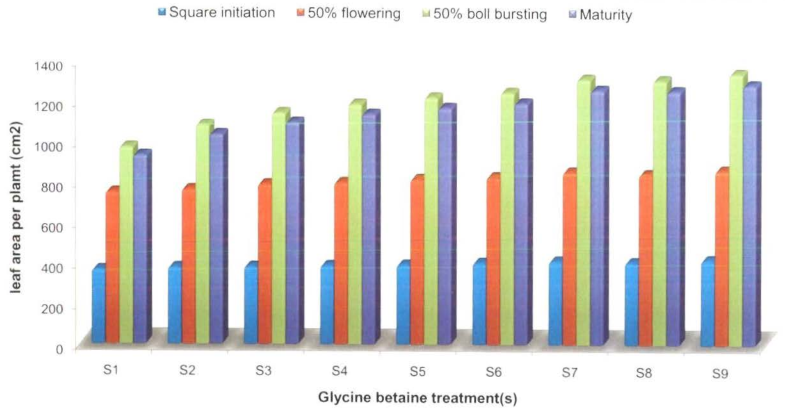


Fig.2: Effect of exogenous application of Glycine betaine on leaf area per plant of cotton under moisture stress

At 50 percent flowering stage, in case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in leaf area. V_2 (805.98 cm^2) found statistically superior over V_1 (802.43 cm^2). While in case of factor B, the statistical difference found significant. The treatment S_9 (855.21cm^2) recorded significantly maximum leaf area among all the treatments followed by S_7 (845.52 cm^2), S_8 (835.45 cm^2) and S_6 (819.52 cm^2) whereas lowest leaf area was recorded in S_1 (742.56 cm^2) the control. However, the interaction effect between varieties and treatments found non significant.

The maximum leaf area in cotton plant under moisture stress were recorded at 50 percent boll bursting stage, in case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in leaf area. V_2 (1196.49 cm^2) found to be superior over V_1 (1191.41 cm^2). While in case of factor B, the statistical difference found significant. The treatment S_9 (1334.12 cm^2) recorded significantly maximum leaf area among all the treatments followed by S_7 (1305.19 cm^2), S_8 (1300.71 cm^2) and S_6 (1237.48 cm^2), whereas lowest leaf area was recorded in S_1 (965.32 cm^2) the control. However, the interaction effect between varieties and treatments found non significant.

At maturity, leaf area of plant is found to be declined as compared to previous growth stage i.e. 50 percent boll bursting. This might be due to fact that, increase in canopy leaf age and increased self shedding and senescence along with the increase in boll demand for carbohydrates results in restricted any new leaf development which cause in leaf area reduction (Thind, 2007). In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in leaf area. V_2 (805.98 cm^2) found to be superior over V_1 (802.43 cm^2). While in case of factor B, the statistical difference found significant. The treatment S_9 (855.21cm^2) recorded significantly maximum leaf area among all the treatments followed by S_7 (845.52 cm^2), S_8 (835.45 cm^2) and S_6 (819.52 cm^2), whereas, lowest leaf area was recorded in S_1 (742.56 cm^2) the control. However, the interaction effect between varieties and treatments found non significant.

Significant increase in leaf area due to exogenous application of Glycine betaine on cotton plant under moisture stress as compared to control, may be the result of promotion of higher accumulation of compatible solutes like proline which then enhance the osmoregulation ability of crop under water stress condition (Farooq *et al.*, 2009). Osmoregulation enables plants to: (1) continue normal leaf elongation but at a reduced rate, (2) adjust their stomatal and photosynthetic functions, (3) postpone leaf senescence, under adverse climatic conditions Boyer, (1982). Osmoregulation (osmotic adjustment) in the leaves of cotton (*Gossypium hirsutum* L.) was reported by Acevedo *et al.*, (1979); Oosterhuis *et al.*, (1987). The present findings are in line with the findings of Agboma et al., (1997) in soybean, Patil (2005) in cotton and Mahmood *et al.* (2009) in wheat.

4.1.3 Specific leaf weight (SLW) (mg/cm^2)

Specific leaf weight is the integral structure of the leaf and is known to have correlation with photosynthetic rate (Rasulou and Asrorov, 1991).

The data on specific leaf weight were recorded at different growth stages as influenced by exogenous application of Glycine betaine is presented in Table 3 and depicted in Fig. 3.

Maximum SLW was recorded at the stage of 50 percent flowering amongst all the growth stages. Then after it declines with the subsequent growth stages and minimum SLW was recorded at the maturity stage under moisture stressed cotton crop.

At square initiation stage, in case of factor A statistical difference for SLW found significant. V_2 (3.162 mg/cm^2) found to be superior over V_1 (3.147 mg/cm^2). While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (3.333 mg/cm^2) recorded significantly maximum SLW among all the treatments

Table 3. Effect of exogenous application of Glycine betaine on specific leaf weight (SLW) of cotton under moisture stress.

Treatment(s)	Specific leaf weight (mg/cm^2)			
	Square initiation	50% flowering	50% boll bursting	Maturity
FACTOR "A" (VARIETY)				
V_1 - Bramha BG (I)	3.147	3.347	3.050	2.813
V_2 - Mallika BG (I)	3.162	3.363	3.064	2.828
'F' test	Sig	Sig	Sig	Sig
SE(m)±	0.001	0.002	0.003	0.001
CD at 5%	0.004	0.007	0.008	0.003
FACTOR "B" Glycine betaine Treatment(s)				
S_1 - Control	2.888	3.134	2.839	2.583
S_2 -2.5% (w/w) Glycine betaine seed treatment.	3.041	3.228	2.944	2.720
S_3 - 5% (w/w) Glycine betaine seed treatment.	3.062	3.250	2.963	2.738
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	3.125	3.317	3.025	2.795
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	3.155	3.347	3.052	2.820
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	3.238	3.435	3.132	2.894
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	3.294	3.495	3.186	2.945
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	3.255	3.454	3.150	2.910
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	3.333	3.536	3.225	2.980
'F' test	Sig	Sig	Sig	Sig
SE(m)±	0.003	0.005	0.006	0.002
CD at 5%	0.008	0.015	0.016	0.006
INTERACTION "AXB"				
'F' test	NS	NS	NS	NS
SE(m)±	0.004	0.007	0.008	0.003
CD at 5%	-	-	-	-

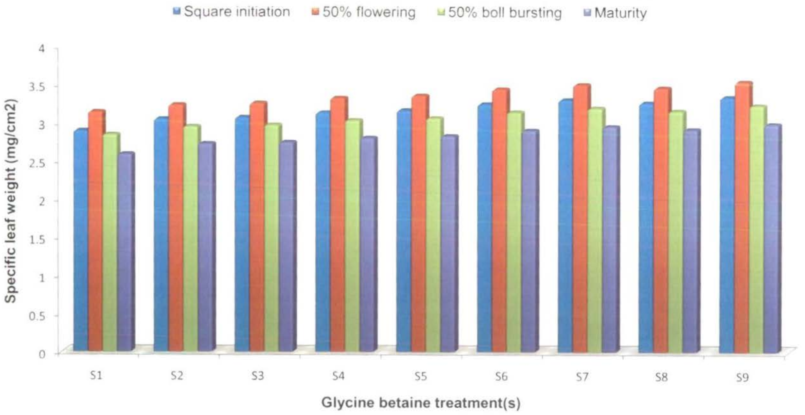


Fig.3: Effect of exogenous application of Glycine betaine on specific leaf weight of cotton under moisture stress

followed by S_7 (3.294 mg/cm^2), S_8 (3.255 mg/cm^2) and S_6 (3.238 mg/cm^2). The least SLW was observed in S_1 (2.888 mg/cm^2) the control. However, the interaction effect between varieties and treatments found non significant.

Maximum SLW were recorded under moisture stress at 50 percent flowering stage. In case of factor A statistical difference for SLW found significant. V_2 (3.363 mg/cm^2) found to be superior over V_1 (3.347 mg/cm^2). While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (3.536 mg/cm^2) recorded significantly maximum SLW among all the treatments followed by S_7 (3.495 mg/cm^2), S_8 (3.454 mg/cm^2) and S_6 (3.435 mg/cm^2). The least SLW was observed in S_1 (3.134 mg/cm^2) the control. However, the interaction effect between varieties and treatments found non significant.

At 50 percent boll bursting specific leaf weight found to be declined with the age of crop, which could be attributed to reduced vegetative growth of plant (Patil, 2005). In case of factor A statistical difference for SLW found significant. V_2 (3.064 mg/cm^2) found superior over V_1 (3.147 mg/cm^2). While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (3.333 mg/cm^2) recorded significantly maximum SLW among all the treatments followed by S_7 (3.294 mg/cm^2), S_8 (3.255 mg/cm^2) and S_6 (3.238 mg/cm^2). The least SLW was observed in S_1 (2.888 mg/cm^2) the control. However, the interaction effect between varieties and treatments found non significant.

Least specific leaf weight under moisture stress were recorded at the stage of maturity. In case of factor A statistical difference for SLW found significant. The variety V_2 (2.828 mg/cm^2) found superior over V_1 (2.813 mg/cm^2). While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (2.980 mg/cm^2) recorded significantly maximum SLW among all the treatments over S_7 (2.945 mg/cm^2), S_8 (2.910 mg/cm^2) and S_6 (2.894 mg/cm^2)

whereas least SLW was noted in S_1 (2.583 mg/cm^2). The interaction effect between varieties and treatments found non significant.

An increase in specific leaf weight due to exogenous application of Glycine betaine on cotton plant under moisture stress as compared to control might be result of either enhanced layer of mesophyll cells and/or increased thickness of conducting vessels (Landiver et al., 1983). The decline in the SLW from later reproductive growth stages could be attributed to the reduction in leaf area expansion (Bharadwaj and Singh, 1988). The reduced rate of leaf expansion results in to reduced size of individual leaves (Thind, 2007) Present results are in conformity with the results of Patil, (2005) in cotton. The varital difference for SLW also reported by Patil *et al.* (2009) in cotton.

4.1.4 Dry matter production/ plant (g)

Water stress reduces the plant height, leaf number, leaf area which results in to reduction in total dry weight of cotton (Patterson, 1989). More biomass accumulation under water deficit condition can be considered as a physiological trait for drought tolerance in cotton (Quisenberry *et. al.*,2011; Hatfield *et al.*, 1987).

The data on dry matter production per plant were recorded at different growth stages as influenced by exogenous application of Glycine betaine is presented in Table 4 and depicted in Fig. 4. An increase in dry matter production per plant of cotton throughout growth period (up to maturity) of crop was recorded under moisture stress during the course of observation.

At square initiation stage, in case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in dry matter production per plant. V_2 (9.974 g) found statistically superior over V_1 (9.910 g). While in case of factor B, the statistical difference found significant. The treatment S_9 (11.169 g) significantly produced maximum dry matter per plant amongst all the

Table 4. Effect of exogenous application of Glycine betaine on dry matter production/plant of cotton under moisture stress.

Treatment(s)	Dry matter production (g/ plant)			
	Square initiation	50% flowering	50% boll bursting	Maturity
FACTOR "A" (VARIETY)				
V_1 - Bramha BG (I)	9.910	25.438	40.420	45.429
V_2 - Mallika BG (I)	9.974	25.587	40.654	45.691
'F' test	Sig	Sig	Sig	Sig
SE(m)±	0.006	0.015	0.024	0.027
CD at 5%	0.017	0.043	0.068	0.076
FACTOR "B" Glycine betaine Treatment(s)				
S_1 - Control	8.426	21.530	29.893	33.597
S_2 -2.5% (w/w) Glycine betaine seed treatment.	9.179	23.115	35.056	39.400
S_3 - 5% (w/w) Glycine betaine seed treatment.	9.332	24.075	37.283	41.903
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	9.673	24.837	39.525	44.423
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	9.864	25.570	41.002	46.083
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	10.420	26.625	42.939	48.260
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	10.839	27.963	46.083	51.793
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	10.579	27.293	45.384	51.007
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	11.169	28.608	47.665	53.571
'F' test	Sig	Sig	Sig	Sig
SE(m)±	0.012	0.032	0.050	0.056
CD at 5%	0.036	0.091	0.144	0.162
INTERACTION "AXB"				
'F' test	NS	NS	NS	NS
SE(m)±	0.018	0.045	0.071	0.080
CD at 5%	-	-	-	-

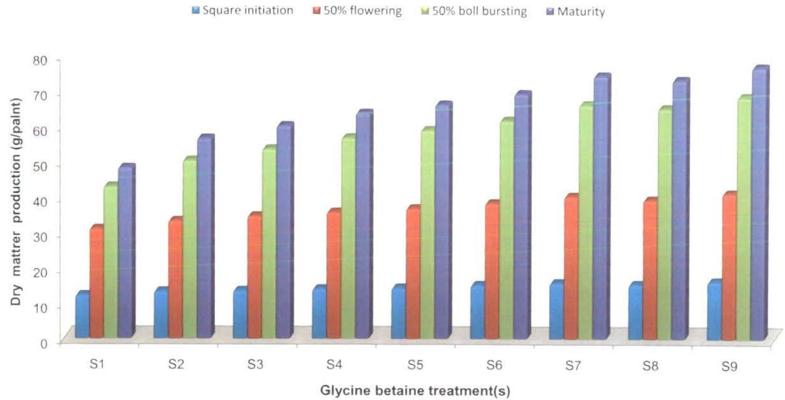


Fig.4: Effect of exogenous application of Glycine betaine on dry matter production per plant of cotton under moisture stress

treatments than S_7 (10.839 g), S_8 (10.569 g) and S_6 (10.420 g), whereas lowest dry matter per plant was produced in S_1 (8.426 g) the control.Interaction effect between varieties and treatments found non significant.

At 50 percent flowering stage, in case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in dry matter production per plant. V_2 (25.587 g) found superior over V_1 (25.438 g). While in case of factor B, the statistical difference found significant. The treatment S_9 (28.608 g) significantly produced maximum dry matter per plant amongst all the treatments than S_7 (27.963 g), S_8 (27.293 g) and S_6 (26.625 g), whereas, lowest dry matter per plant was produced in S_1 (21.530 g) the control. However, the interaction effect between varieties and treatments found non significant.

At 50 percent boll bursting stage, in case of factor A, variety V_2 (40.654 g) found superior than V_1 (40.420 g). While in case of factor B, the statistical difference found significant. The treatment S_9 (47.665 g) significantly produced maximum dry matter per plant amongst all the treatments followed by S_7 (46.083 g), S_8 (45.384 g) and S_6 (42.939 g). The lowest dry matter per plant was produced in S_1 (29.893 g) the control. However, the interaction effect between varieties and treatments found non significant.

Maximum production of dry matter per plant in cotton under moisture stress were recorded at maturity, in case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in dry matter production per plant. V_2 (45.691 g) found superior over V_1 (45.429 g). While in case of factor B, the statistical difference found significant. The treatment S_9 (53.571 g) significantly produced maximum dry matter per plant amongst all the treatments followed by S_7 (51.793 g), S_8 (51.007 g) and S_6 (48.260 g). The lowest dry matter per plant was produced in S_1 (33.597 g) the control. The interaction effect between varieties and treatments found non significant.

Exogenously applied Glycine betaine cause resultant increase in dry matter production per plant of cotton under moisture stress as compared to control. This might be due to fact that exogenously applied Glycine betaine helps in maintaining photosynthesis activity, starch metabolism and increased antioxidant enzyme activities which results in improvement and accelerated growth of plant (Ma et al.,2007). It also helps in active accumulation of compatible solutes (Farooq et al., 2008, Hussain et al., 2009, Patil *et al.*, 2011) which helps to maintain water potential gradient and reestablishment of cell turgor (Hsiao, 1973). This process of osmotic adjustment enables plant to achieve better dry matter and yield production under adverse climatic conditions (Boyer, 1982). The present results are in accordance with the results of Gorham *et al.* (1998_a); Patil *et al.*, (2009) in cotton and Farooq *et al.* (2008).

4.2 Phenological parameters

4.2.1 Days to 50 percent flowering

The data on, days to 50 percent flowering is presented in Table 5 and depicted in Fig. 5. The difference between number of days to 50 percent flowering in case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), found statistically non significant. In case of factor B (Glycine betaine treatment) treatment S_7 (64.67) took minimum no of days for 50 percent flowering. It found to be statistically at par with S_9 (65.17) followed by S_8 (65.83). Whereas treatment S_1 (70.67) the control, took maximum no of days for 50 percent flowering. The treatments [S_7 and S_9], [S_6 and S_5], [S_4 and S_3] [S_3 and S_2] [S_2 and S_1] were recorded at par days to 50 percent flowering.

4.2.2 Days to 50 percent boll bursting

The data on, days to 50 percent boll bursting is presented in Table 5 and depicted in Fig. 5. In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), found statistically non significant.

Table 5. Effect of exogenous application of Glycine betaine on phenological characters of cotton under moisture stress.

Treatment(s)	Days After Sowing (DAS)		
	50% flowering	50% boll bursting	Maturity
FACTOR "A" (VARIETY)			
V_1 - Bramha BG (I)	67.67	130.44	155.85
V_2 - Mallika BG (I)	67.96	130.63	156.04
'F' test	NS	NS	NS
SE(m)±	0.11	0.09	0.08
CD at 5%	-	-	-
FACTOR "B" Glycine betaine Treatment(s)			
S_1 – Control	70.67	134.50	159.17
S_2 -2.5% (w/w) Glycine betaine seed treatment.	70.33	133.67	159.00
S_3 - 5% (w/w) Glycine betaine seed treatment.	69.67	132.67	158.17
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	69.00	131.67	157.00
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	67.83	130.83	156.00
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	67.17	129.17	155.00
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	64.67	127.17	153.00
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	65.83	128.83	154.00
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	65.17	126.33	152.17
'F' test	Sig	Sig	Sig
SE(m)±	0.24	0.18	0.17
CD at 5%	0.68	0.53	0.48
INTERACTION "AXB"			
'F' test	NS	NS	NS
SE(m)±	0.33	0.26	0.24
CD at 5%	-	-	-

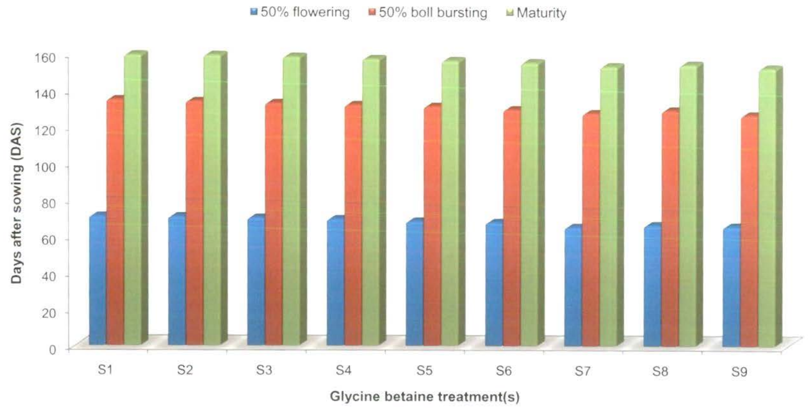


Fig.5: Effect of exogenous application of Glycine betaine on phenological parameters of cotton under moisture stress

In case of factor B (Glycine betaine treatment) treatment S_9 (126.33) took minimum no of days for 50 percent boll bursting, followed by S_7 (127.17), S_8 (128.83) and S_6 (129.17). Whereas treatment S_1 (70.67) the control, took maximum no of days for 50 percent boll bursting.

4.2.3 Days to maturity

The data on days to maturity is presented in Table 5. and depicted in Fig. 5. The difference for number of days to maturity in case of factor A, i.e. found statistically non significant. In case of factor B (Glycine betaine treatment) treatment S_9 (152.17) took minimum no of days to maturity, followed by S_7 (153), S_8 (154) and S_6 (155). Whereas treatment S_1 (159.17) the control, took maximum no of days for attaining maturity. treatment S_2 and S_1 recorded at par days to attaining maturity.

Water stressed condition may have extended early growth cycles in cotton plant (Thind, 2007). Exogenously applied Glycine betaine under water deficit found helpful in maintaining requisite tissue water status of plant which helps in improving photosynthesis activity, starch metabolism and increased antioxidant enzyme activities, results in improvement and accelerated growth (Ma *et al.*, 2007) which might have played important role in achieving early maturity as compared to control. Early maturity is correlated with high yield in upland cotton *Gossypium hirsutum* L.) (Quisenberry and Roask, 1976).

Adarsha *et al.* (2003) indicated that, earliness appears to have a distinct advantage under water stress condition in cotton. Chaudhary and Syed (1978) found similar results for delay in flowering due to moisture stress. Water deficit during advance stages delay in flowering and grain filling in rice (Kumar and Kujru, 2003).

4.3 Biochemical parameters

4.3.1 Chlorophyll stability index (CSI)

The data on chlorophyll stability index at different growth stages of cotton under moisture stress is presented in Table 6 and depicted in Fig. 6.

Table 6. Effect of exogenous application of Glycine betaine on chlorophyll stability index (CSI) of cotton under moisture stress.

Treatment(s)	chlorophyll stability index (CSI)		
	Square initiation	50% flowering	50% boll bursting
FACTOR "A" (VARIETY)			
V_1 - Bramha BG (I)	52.044	57.383	46.169
V_2 - Mallika BG (I)	52.334	57.945	46.657
'F' test	Sig	Sig	Sig
SE(m)±	0.020	0.039	0.032
CD at 5%	0.058	0.113	0.092
FACTOR "B" Glycine betaine Treatment(s)			
S_1 – Control	44.102	46.082	35.727
S_2 -2.5% (w/w) Glycine betaine seed treatment.	48.869	49.735	40.174
S_3 - 5% (w/w) Glycine betaine seed treatment.	49.845	51.743	41.795
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	51.605	55.458	44.799
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	52.110	56.551	45.681
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	54.536	61.936	50.032
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	56.432	66.323	53.572
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	54.875	62.710	50.657
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	57.326	68.437	55.283
'F' test	Sig	Sig	Sig
SE(m)±	0.043	0.083	0.068
CD at 5%	0.123	0.239	0.196
INTERACTION "AXB"			
'F' test	NS	NS	NS
SE(m)±	0.060	0.118	0.096
CD at 5%	-	-	-

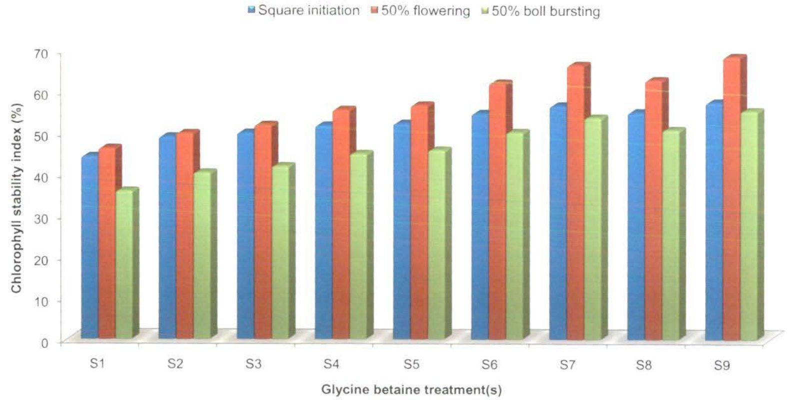


Fig. 6: Effect of exogenous application of Glycine betaine on chlorophyll stability index of cotton under moisture stress

At square initiation stage, in case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in chlorophyll stability index. V_2 (52.334) found superior over V_1 (52.044). While in case of factor B, the statistical difference found significant. The treatment S_9 (57.326) significantly recorded maximum chlorophyll stability index amongst all the treatments followed by S_7 (56.432), S_8 (54.875) and S_6 (54.536), whereas, the lowest chlorophyll stability index was recorded in S_1 (44.102) the control. However, the interaction effect between varieties and treatments found non significant.

Maximum CSI was recorded at 50 percent flowering stage amongst all three observational growth stages. In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in chlorophyll stability index. V_2 (57.945) found superior over V_1 (57.383). While in case of factor B, the statistical difference found significant. The treatment S_9 (68.437) significantly recorded maximum chlorophyll stability index amongst all the treatments followed by S_7 (66.323), S_8 (62.710) and S_6 (61.936), whereas lowest chlorophyll stability index was recorded in S_1 (46.082) the control. However, the interaction effect between varieties and treatments found non significant.

At 50 percent boll bursting stage, in case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), recorded significant difference in chlorophyll stability index. V_2 (46.657) found to be superior over V_1 (46.169). While in case of factor B, the statistical difference found significant. The treatment S_9 (55.283) significantly recorded maximum chlorophyll stability index amongst all the treatments followed by S_7 (53.572), S_8 (50.657) and S_6 (50.032), whereas lowest chlorophyll stability index was recorded in S_1 (46.169). However, the interaction effect between varieties and treatments found non significant.

The reduction in chlorophyll stability index might be attributed to decreased chlorophyll content in leaves under water stress condition which can be caused due to lesser chlorophyll membrane integrity under water stress condition (Patil, 2005). Glycine betaine sustains the integrity of thalakoid membranes (Genard *et al.*, 1991) which might have helped to increase in CSI of cotton plants under moisture stress as compared to control.

Chetti *et al.* (2002) reported that, the property of CSI is associated with drought resistance and hence the criteria for measuring drought tolerance. Present findings are in accordance with the findings of Patil *et al.* (2011) in cotton. The varital difference in CSI under water deficit was also reported by Kar *et al.* (2005) and Patil *et al.* (2011) in cotton.

4.3.2 Stomatal frequency (per mm^2)

The data recorded for stomatal frequency in cotton under moisture stress is presented in table 7. The data indicated that factor A (varieties, V_1 and V_2) factor B (Glycine betaine treatment) and interaction effect found statistically non significant in all the three observational growth stages (square initiation, 50 percent flowering and 50 percent boll bursting) in cotton under moisture stress. However, the data recorded implies to only one season crop growth, which needs further confirmation on multilocation basis.

4.3.3 Relative water content (%)

The data recorded for relative water content at different growth stages of cotton under moisture stress is presented in Table 8 and depicted in Fig. 7. Plate 1a and 1b shows the effect of exogenous application of Glycine betaine on cotton plant water status under moisture stress.

Table 7. Effect of exogenous application of Glycine betaine on stomatal frequency of cotton under moisture stress.

Treatment(s)	Stomatal frequency (per mm^2)		
	Square initiation	50% flowering	50% boll bursting
FACTOR "A" (VARIETY)			
V_1 - Bramha BG (I)	81.30	61.94	45.54
V_2 - Mallika BG (I)	81.33	60.33	45.71
'F' test	NS	NS	NS
SE(m)±	0.69	0.87	0.23
CD at 5%	-	-	-
FACTOR "B" Glycine betaine Treatment(s)			
S_1 – Control	80.67	58.21	44.95
S_2 -2.5% (w/w) Glycine betaine seed treatment.	81.50	63.00	45.45
S_3 - 5% (w/w) Glycine betaine seed treatment.	82.50	58.33	45.58
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	78.83	61.00	46.21
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	81.67	63.56	45.71
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	80.67	64.67	45.08
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	80.50	62.50	45.58
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	85.83	58.47	46.09
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	79.67	60.50	45.96
'F' test	NS	NS	NS
SE(m)±	1.46	1.84	0.49
CD at 5%	-	-	-
INTERACTION "AXB"			
'F' test	NS	NS	NS
SE(m)±	2.06	2.61	0.69
CD at 5%	-	-	-

Table 8. Effect of exogenous application of Glycine betaine on relative water content (RWC) of cotton under moisture stress.

Treatment(s)	Relative water content (%)		
	Square initiation	50% flowering	50% boll bursting
FACTOR "A" (VARIETY)			
V_1 - Bramha BG (I)	75.80	60.02	50.56
V_2 - Mallika BG (I)	76.20	60.33	50.83
'F' test	Sig	Sig	Sig
SE(m)±	0.02	0.02	0.02
CD at 5%	0.08	0.06	0.05
FACTOR "B" Glycine betaine Treatment(s)			
S_1 –Control	66.83	52.91	44.57
S_2 -2.5% (w/w) Glycine betaine seed treatment.	70.86	56.10	47.27
S_3 - 5% (w/w) Glycine betaine seed treatment.	72.28	57.22	48.21
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	74.83	59.25	49.92
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	75.56	59.82	50.40
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	79.08	62.61	52.75
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	81.83	64.79	54.58
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	79.57	63.02	53.07
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	83.13	65.82	55.45
'F' test	Sig	Sig	Sig
SE(m)±	0.05	0.04	0.03
CD at 5%	0.17	0.13	0.11
INTERACTION "AXB"			
'F' test	NS	NS	NS
SE(m)±	0.08	0.06	0.05
CD at 5%	-	-	-

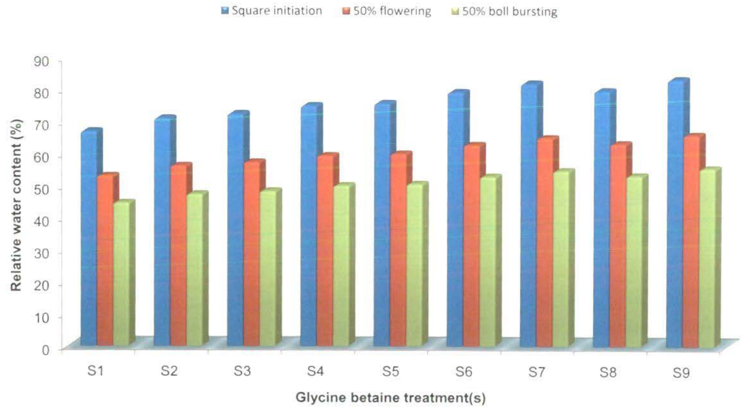


Fig. 7: Effect of exogenous application of Glycine betaine on relative water content cotton under moisture stress

V_1S_1

V_1S_2

V_1S_3

V_1S_4

V_1S_5

V_1S_6

V_1S_7

V_1S_8

V_1S_9

Plate 1(a). Effect of Glycine Betaine on cotton (Cv. Bramha BG I) at 21st day of moisture stress

Plate 1(b). Effect of Glycine Betaine on cotton (Cv. Mallika BG I) at 21st day of moisture stress

At square initiation stage, in case of factor A which comprises of varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), the statistical difference for relative water content found significant. V_2 (76.20%) found superior over V_1 (75.80%). While in case of factor B, all treatments differed significantly for RWC. The treatment S_9 (83.13%) recorded significantly maximum RWC among all the treatments followed by S_7 (81.83%), S_8 (79.57%) and S_6 (79.08%), whereas, least RWC was observed in S_1 (66.83%) the control. However, the interaction effect between varieties and treatments found non significant.

At 50% flowering stage, in case of factor A which comprises of varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), the statistical difference for relative water content found significant. V_2 (76.20%) found superior over V_1 (75.80%). While in case of factor B, all treatments differed significantly for RWC. The treatment S_9 (83.13%) recorded significantly maximum RWC among all the treatments followed by S_7 (81.83%), S_8 (79.57%) and S_6 (79.08%), whereas, least RWC was observed in S_1 (66.83%). However, the interaction effect between varieties and treatments found non significant at this stage of plant growth.

At 50% boll bursting stage, in case of factor A which comprises of varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), the statistical difference for relative water content found significant. V_2 (50.83%) found superior over V_1 (50.56%). While in case of factor B, all treatments differed significantly for RWC. The treatment S_9 (55.45%) recorded significantly maximum RWC among all the treatments followed by S_7 (54.58%), S_8 (53.07%) and S_6 (52.75%), whereas, least RWC was observed in S_1 (44.57%). However, the interaction effect between varieties and treatments found non significant.

The relative water content (RWC) describes internal water status of the plant. Increase in RWC at all three observational stages with exogenous application of Glycine betaine as compared to control is may be attributed to the promotion of higher osmolytes accumulation like proline etc. (Farooq *et al.*, 2008, Hussain *et al.*, 2009, Patil *et al.*, 2011) which helps to maintain water potential gradient and reestablishment of cell turgor under water deficit. (Hsiao, 1973).present results are similar to the findings of Farooq *et al.*, (2008) in rice, Hussain *et al.*, (2009) in sunflower, Patil *et al.*, (2011) in cotton. The varital difference in RWC also been reported by Patil *et al.*, (2011) in cotton.

4.3.4 Proline (µg g fresh wt.$^{-1}$)

The data recorded for proline content at different growth stages of cotton under moisture stress is presented in Table 9 and depicted in Fig. 8.

At square initiation stage, in case of factor A which comprises of varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), the statistical difference for proline content found significant. V_2 (46.72 µg/g) found superior over V_1 (46.46 µg/g). While in case of factor B, all treatments differed significantly. The treatment S_9 (54.78 µg/g) recorded significantly maximum proline content among all the treatments followed by S_7 (52.96 µg/g), S_8 (52.16 µg/g) and S_6 (49.35 µg/g) and the least proline content was observed in S_1 (34.36 µg/g) the control. However, the interaction effect between varieties and treatments found non significant.

At 50% flowering stage, in case of factor A which comprises of varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), the statistical difference for proline content found significant. V_2 (34.28 µg/g) found superior over V_1 (34.09 µg/g). While in case of factor B, all treatments differed significantly. The treatment S_9 (40.20 µg/g) recorded significantly maximum proline content among all the treatments followed by S_7 (38.87 µg/g), S_8 (38.27 µg/g) and S_6 (36.21 µg/g) and the least proline content was observed in S_1 (25.21 µg/g) the control. However, the interaction effect between varieties and treatments found non significant.

Table 9. Effect of exogenous application of Glycine betaine on proline content in cotton under moisture stress.

Treatment(s)	Proline (µg g fresh wt.$^{-1}$)		
	Square initiation	50% flowering	50% boll bursting
FACTOR "A" (VARIETY)			
V_1 - Bramha BG (I)	46.46	34.09	23.03
V_2 - Mallika BG (I)	46.72	34.28	23.16
'F' test	Sig	Sig	Sig
SE(m)±	0.03	0.02	0.01
CD at 5%	0.08	0.06	0.04
FACTOR "B" Glycine betaine Treatment(s)			
S_1 – Control	34.36	25.21	19.49
S_2 -2.5% (w/w) Glycine betaine seed treatment.	40.29	29.56	20.92
S_3 - 5% (w/w) Glycine betaine seed treatment.	42.85	31.44	21.79
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	45.43	33.33	22.48
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	47.12	34.58	23.15
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	49.35	36.21	24.10
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	52.96	38.87	25.31
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	52.16	38.27	24.71
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	54.78	40.20	25.90
'F' test	Sig	Sig	Sig
SE(m)±	0.06	0.04	0.03
CD at 5%	0.17	0.12	0.08
INTERACTION "AXB"			
'F' test	NS	NS	NS
SE(m)±	0.08	0.06	0.04
CD at 5%	-	-	-

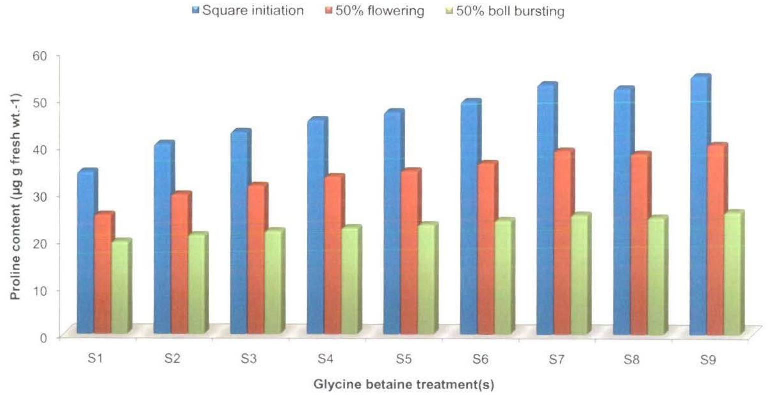


Fig. 8: Effect of exogenous application of Glycine betaine on proline content in cotton under moisture stress

At 50% boll bursting stage, in case of factor A which comprises of varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), the statistical difference for proline content found significant. V_2 (23.16 µg/g) found statistically superior over V_1 (23.06 µg/g). While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (25.90 µg/g) recorded significantly maximum proline content among all the treatments followed by S_7 (25.31 µg/g), S_8 (24.71 µg/g) and S_6 (49.35 µg/g) and the least proline content was observed in S_1 (24.10 µg/g) the control. However, the interaction effect between varieties and treatments found non significant.

Proline content has been shown to accumulate upon desiccation in leaves of many plant species. It has been suggested by Jones and Turner, (1978) that accumulation of proline could make useful contribution to the osmotic adjustment. If it is confined to cytoplasm, the direct evidences for this are lacking and several other roles have also been suggested by Blum and Ebercon (1976) for proline accumulation in addition to osmotic adjustment. Proline plays an important role as carbon and nitrogen metabolism, detoxification of NH3, preserving the hydration of proteins in dehydrated tissues thereby contributing to the survival of cellular functions. Exogenous application of Glycine betaine increased levels of proline as compared to control under moisture deficit cotton might be due to fact of significant increase in Δ^{1}- pyrroline-5-carboxylate synthetase (P5CS) activity which attributes to the synthesis of proline under stress condition (Kavikishor *et al.*, 1995) found positively correlated with increase in Glycine betaine levels in cotton plant under water deficit condition (Pawar *et al.*, 2010).

4.3.5 Nitrate reductase (NR) activity (µmole g fresh wt.$^{-1}$ hr^{-1})

The data recorded for nitrate reductase (NR) activity at different growth stages of cotton under moisture stress is presented in Table 10 and depicted in Fig. 9.

Table 10. Effect of exogenous application of Glycine betaine on Nitrate reductase (NR) activity of cotton under moisture stress.

Treatment(s)	Nitrate reductase (NR) activity (µmole g fresh wt.$^{-1}$ hr^{-1})		
	Square initiation	50% flowering	50% boll bursting
FACTOR "A" (VARIETY)			
V_1 - Bramha BG (I)	0.9831	1.1904	0.9841
V_2 - Mallika BG (I)	1.0077	1.2495	1.0081
'F' test	Sig	Sig	Sig
SE(m)±	0.0050	0.0196	0.0081
CD at 5%	0.0143	0.0563	0.0232
FACTOR "B" Glycine betaine Treatment(s)			
S_1 – Control	0.9058	0.7501	0.7874
S_2 -2.5% (w/w) Glycine betaine seed treatment.	0.9642	0.8629	0.8470
S_3 - 5% (w/w) Glycine betaine seed treatment.	0.9572	0.8864	0.8585
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.9951	1.0681	0.9422
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.9917	1.1095	0.9602
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	1.0035	1.3555	1.0616
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	1.0403	1.6410	1.1663
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	1.0270	1.4999	1.1166
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	1.0737	1.8061	1.2253
'F' test	Sig	Sig	Sig
SE(m)±	0.0106	0.0416	0.0172
CD at 5%	0.0303	0.1194	0.0493
INTERACTION "AXB"			
'F' test	NS	NS	NS
SE(m)±	0.0150	0.0589	0.0243
CD at 5%	-	-	-

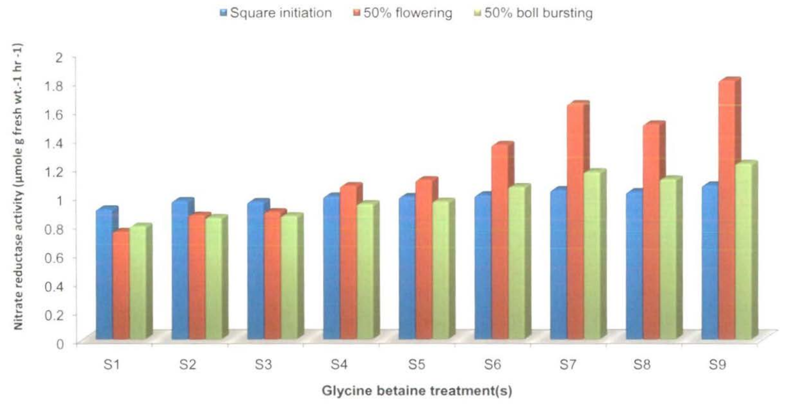


Fig. 9: Effect of exogenous application of Glycine betaine on nitrate reductase activity of cotton under moisture stress

At square initiation stage, in case of factor A, V_2 (1.0077 µmoles/g) found statistically superior over V_1 (0.9831 µmoles/g/hr). While in case of factor B, all treatments differed significantly. The treatment S_9 (1.0737 µmoles/g/hr) recorded significantly highest NR activity among all the treatments followed by S_7 (1.0403 µmoles/g/hr), whereas, lowest NR activity was observed in S_1 (0.9058 µmoles/g/hr). The treatments [S_6, S_5 and S_4] [S_3 and S_2] were recorded at par values of NR activity. However, the interaction effect between varieties and treatments found non significant for this trait.

At 50% flowering stage, NR activity found at its peak compared with other observational growth stages in cotton under moisture stress. In case of factor A, V_2 (1.2495 µmoles/g/hr) found statistically superior over V_1 (1.1904 µmoles/g/hr).

While in case of factor B, all treatments differed significantly. The treatment S_9 (1.8061 µmoles/g/hr) recorded significantly highest NR activity among all the treatments followed by S_7 (1.6410 µmoles/g/hr), whereas, lowest NR activity was observed in S_1 (0.7501 µmoles/g/hr) the control, which found statistically at par with S_2 (0.8629 µmoles/g/hr). The treatments [S_5 and S_4], [S_3 and S_2], [S_2 and S_1], were recorded at par values for NR activity. However, the interaction effect between varieties and treatments found non significant for this trait.

At 50% boll bursting stage, in case of factor A, V_2 (1.0081 µmoles/g/hr) found statistically superior over V_1 (0.9841 µmoles/g/hr). While in case of factor B, all treatments differed significantly. The treatment S_9 (1.2253 µmoles/g/hr) recorded significantly highest NR activity among all the treatments followed by S_7 (1.1663 µmoles/g/hr). Whereas lowest NR activity was observed in S_1 (0.7874 µmoles/g/hr) the control. The treatments [S_5 and S_4], [S_3 and S_2] were recorded at par values for NR activity. However, the interaction effect between varieties and treatments found non significant for this trait.

Reduction in NR activity due to water stress in various crops is well established (Sinha and Nicholas, 1981). Increase in NR activity as compared to control in moisture stressed cotton plant due to exogenous application of Glycine betaine might be due to fact that it helps for maintenance of the structures and activities of enzymes and protein complexes. However, the data recorded implies to only one season crop growth, which needs further confirmation on multilocation basis.

4.3.6 Photosynthetic efficiency (Fv/Fm)

The data recorded for photosynthetic efficiency at different growth stages of cotton under moisture stress is presented in Table 11 and depicted in Fig. 10.

At square initiation stage, in case of factor A which comprises of varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), the statistical difference for photosynthetic efficiency found significant. V_2 (0.807 Fv/Fm) found superior over V_1 (0.795 Fv/Fm). While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (0.860 Fv/Fm) recorded significantly highest photosynthetic efficiency among all the treatments whereas, lowest photosynthetic efficiency was observed in S_1 (0.752 Fv/Fm) the control. The treatments [S_4 and S_5], [S_2 and S_3] remained at par among themselves. However, the interaction effect between varieties and treatments found non significant.

At 50% flowering stage, in case of factor A, the statistical difference for photosynthetic efficiency found significant. the variety V_2 (0.753 Fv/Fm) found superior over the variety V_1 (0.743 Fv/Fm). While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (0.780 Fv/Fm) recorded significantly highest photosynthetic efficiency among all the treatments followed by S_7 (0.773 Fv/Fm), whereas, lowest photosynthetic efficiency was observed in S_1 (0.718 Fv/Fm). The treatments [S_4 and S_5], [S_2 and S_3] were recorded at par among themselves. However, the interaction effect between varieties and treatments found non significant.

Table 11. Effect of exogenous application of Glycine betaine on photosynthetic efficiency of cotton under moisture stress.

Treatment(s)	Photosynthetic efficiency (Fv/Fm)		
	Square initiation	50% flowering	50% boll bursting
FACTOR "A" (VARIETY)			
V_1 - Bramha BG (I)	0.795	0.743	0.646
V_2 - Mallika BG (I)	0.807	0.753	0.652
'F' test	Sig	Sig	Sig
SE(m)±	0.002	0.001	0.001
CD at 5%	0.005	0.002	0.003
FACTOR "B" Glycine betaine Treatment(s)			
S_1 – Control	0.752	0.718	0.612
S_2 -2.5% (w/w) Glycine betaine seed treatment.	0.764	0.726	0.625
S_3 - 5% (w/w) Glycine betaine seed treatment.	0.771	0.729	0.628
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.789	0.740	0.642
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.794	0.744	0.646
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.812	0.759	0.659
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.841	0.773	0.677
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.824	0.764	0.664
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.860	0.780	0.688
'F' test	Sig	Sig	Sig
SE(m)±	0.004	0.001	0.002
CD at 5%	0.011	0.004	0.007
INTERACTION "AXB"			
'F' test	NS	NS	NS
SE(m)±	0.005	0.002	0.003
CD at 5%	-	-	-

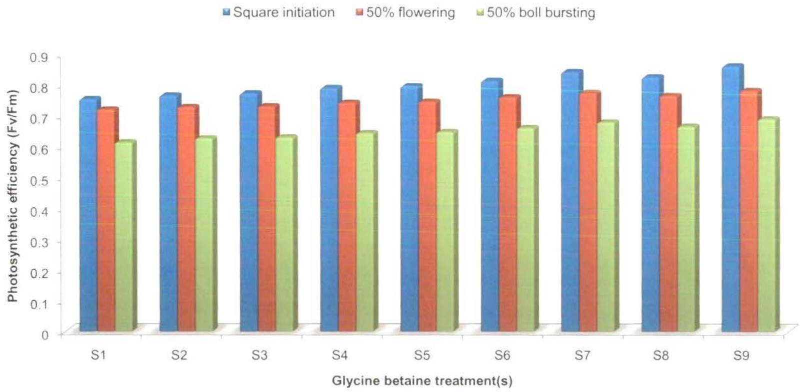


Fig. 10: Effect of exogenous application of Glycine betaine on photosynthetic efficiency of cotton under moisture stress

At 50% boll bursting stage, in case of factor A the variety V_2 (0.652 Fv/Fm) found superior over the variety V_1 (0.645 Fv/Fm). While in case of factor B, all treatments differed significantly at this stage. The treatment S_9 (0.688 Fv/Fm) recorded significantly highest photosynthetic efficiency among all the, whereas, lowest photosynthetic efficiency was observed in S_1 (0.612 Fv/Fm) the control. The treatments [S_8 and S_6], [S_4 and S_5], [S_2 and S_3] were recorded at par among themselves. However, the interaction effect between varieties and treatments found non significant.

Water stress reduces the photosynthetic efficiency of plants (Hsiao, 1973). Exogenous application of Glycine betaine under moisture stress might be result of its accumulation in the chloroplast where it plays a vital role in adjustment and protection of thalakoid membrane under stress there by maintaining photosynthetic efficiency (Robinson and Jones, 1986; Genard *et al.*, 1991). The present results are in accordance with the results of Koti (1997), Xing and Rajashekar (1999) also reported reduced effect of water deficit on chlorophyll fluorescence by exogenous application of Glycine betaine.

4.4 Yield and yield attributing characters

4.4.1 Number of monopodia

The data on, number of monopodia under moisture stress is presented in Table 12 and depicted in Fig. 11A. The difference for number of monopodia In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), found statistically significant. V_2 (1.09) found to be superior over V_1 (0.89). In case of factor B (Glycine betaine treatment) treatment S_9 (1.67) recorded significantly more number of monopodia, whereas treatment S_1 (0.40) the control recorded least number of monopodia amongst all the treatments. The treatments [S_9 and S_7], [S_7, S_8 and S_6], [S_5, S_4 and S_3], [S_4, S_3 and S_2], [S_2 and S_1] were remained at par amongst themselves. However, the interaction effect between varieties and treatments found non significant.

Table 12. Effect of exogenous application of glycine betaine on Yield and yield attributing characters of cotton under moisture stress

Treatment(s)	Yield and Yield attributing characters							
	No. of Monopodia	No. of Sympodia	No. of bolls per plant	Single boll weight (g)	Boll weight per plant (g)	Test weight (g)	Seed cotton yield (g/ plant)	Harvest Index (%)
FACTOR "A" (VARIETY)								
V_1 - Bramha BG (I)	0.89	12.27	8.37	2.15	21.43	6.64	18.56	27.82
V_2 - Mallika BG (I)	1.09	12.68	8.58	2.22	22.35	6.81	19.48	29.04
'F' test	Sig	Sig	NS	Sig	SIG	Sig	Sig	Sig
SE±	0.03	0.06	0.08	0.02	0.11	0.03	0.11	0.16
CD±	0.09	0.18	-	0.05	0.31	0.10	0.31	0.46
FACTOR "B" Glycine betaine Treatment(s)								
S_1 – Control	0.40	9.30	5.43	1.73	11.27	6.12	9.39	19.43
S_2 - 2.5% (w/w) Glycine betaine seed treatment.	0.57	10.17	6.66	1.86	14.40	6.51	12.37	21.97
S_3 - 5% (w/w) Glycine betaine seed treatment.	0.63	10.60	7.36	1.88	16.13	6.47	13.87	23.17
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.67	11.75	8.12	2.07	19.36	6.72	16.76	26.41
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	0.72	12.26	8.71	2.11	21.08	6.70	18.33	27.85
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	1.33	13.35	9.15	2.33	24.52	6.78	21.31	30.91
S_7- 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	1.53	14.84	10.18	2.56	29.66	7.03	25.96	35.09
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	1.40	14.22	9.93	2.45	27.84	6.94	24.32	33.37
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	1.67	15.77	10.73	2.69	32.76	7.25	28.86	37.70
'F' test	Sig	Sig	Sig	Sig	Sig	Sig	Sig	Sig
SE±	0.07	0.13	0.16	0.04	0.23	0.07	0.23	0.34
CD±	0.20	0.38	0.46	0.11	0.66	0.21	0.66	0.98
INTERACTION "AXB"								
'F' test	NS	NS	NS	NS	NS	NS	NS	NS
SE±	0.10	0.19	0.23	0.05	0.32	0.10	0.32	0.48
CD±	-	-	-	-	-	-	-	-

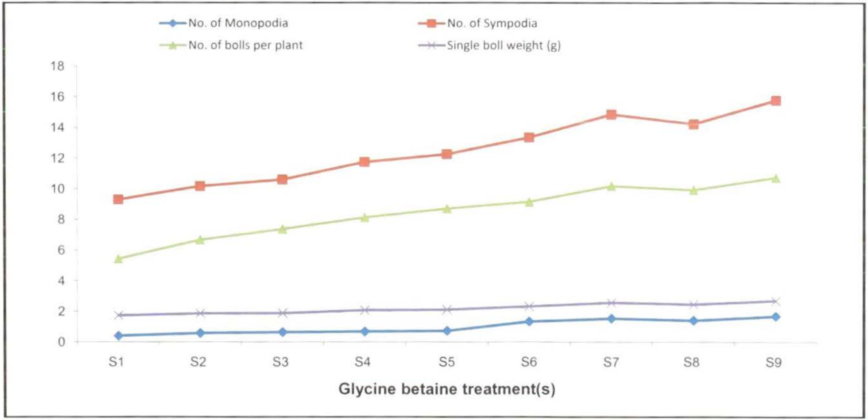


Fig. 11A: Effect of exogenous application of Glycine betaine on No. of monopodia, No. of sympodia, No. of bolls per plant and Single boll weight of cotton under moisture stress

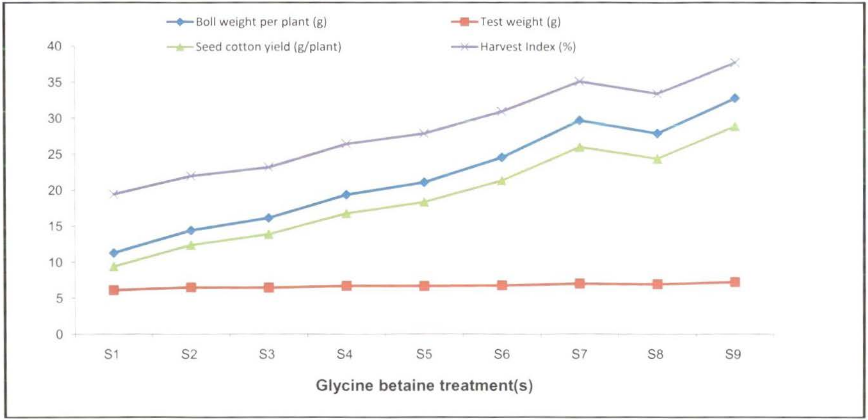


Fig. 11B: Effect of exogenous application of Glycine betaine on Boll weight per plant (g), Test weight (g), Seed cotton yield (g/plant) and Harvest index (%) of cotton under moisture stress

The number of nodes from the base of the main stalk to the first fruiting branch (monopodia) varies considerably among cotton varieties and is affected by moisture availability, abortion of terminal bud of plant. The increased plant height due to exogenous application of glycine betaine (Table 1) might have helped to increase number of nodes from where monopodia emerge. Similar results were observed by Patil *et al.* (2009). He also reported varietal difference in number of monopodia under rainfed condition.

4.4.2 Number of sympodia

The data on, number of sympodia under moisture stress is presented in Table 12 and depicted in Fig. 11A. The difference for number of sympodia In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), found statistically significant. V_2 (12.68) found to be superior over V_1 (12.27).

The difference for number of sympodia In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), found statistically significant. V_2 (12.68) found to be superior over V_1 (12.27). In case of factor B (Glycine betaine treatment) treatment S_9 (15.77) recorded significantly more number of sympodia, followed by S_7 (14.84) S_8 (14.22) S_6 (13.35), whereas treatment S_1 (9.30) the control recorded least number of sympodia amongst all the treatments. However, the interaction effect between varieties and treatments found non significant.

The increase in number of monopodia and sympodia in cotton under moisture stress is recorded when Glycine betaine is exogenously applied. It might be attributed to increased plant height which provides site for nodes and internodes from where monopodial and sympodial branches emerges (Eaton, 1955). Morphological characters like number of monopodial and sympodial branches are associated with increase in seed cotton yield (Basu and Bhatt, 1987) Similar results were observed by Patil *et al.* (2009). He also reported varital difference in number of Sympodia under rainfed condition.

4.4.3 Number of bolls per plant

The data (Table 12) on, number of bolls per plant and depicted in Fig. 11A. The difference between number of bolls per plant In case of factor A, found statistically non significant. In case of factor B (Glycine betaine treatment) treatment S_9 (10.73) recorded numerically more number of bolls per plant, followed by S_7 (10.18) S_8 (9.93) S_6 (9.15), whereas treatment S_1 (5.43) the control recorded least number of bolls per plant amongst all the treatments. However, the interaction effect between varieties and treatments found non significant.

Water stress prior to flowering reduced number of fruiting sites due to inhibition of site initiation which then affects the boll number per plant and boll size as they have very strong relation with water supply (Thind, 2007). The exogenously applied Glycine betaine might have contributed to increase in fruiting sites by increasing number of sympodia results in to keeping better number of bolls per plant of cotton under moisture stress. The present findings are corroborated with Baraiya *et al.* (2009); Patil *et al.* (2009).

4.4.4 Single boll weight (g)

The data on, single boll weight under moisture stress is presented in Table 12 and depicted in Fig. 11A. plate 2 shows the effect of exogenous application of Glycine betaine on the boll size of cotton under moisture stress. In case of factor A, found statistically significant. V_2 (2.22 g) found statistically superior over the Bt hybrid V_1 (2.15 g). In case of factor B (Glycine betaine treatment) treatment S_9 (2.69 g) recorded significantly higher single boll weight, followed by S_7 (2.56 g), S_8 (2.45 g), S_6 (2.33 g), whereas treatment S_1 (1.73 g) the control recorded least single boll weight amongst all the treatments. The interaction effect between varieties and treatments found non significant.

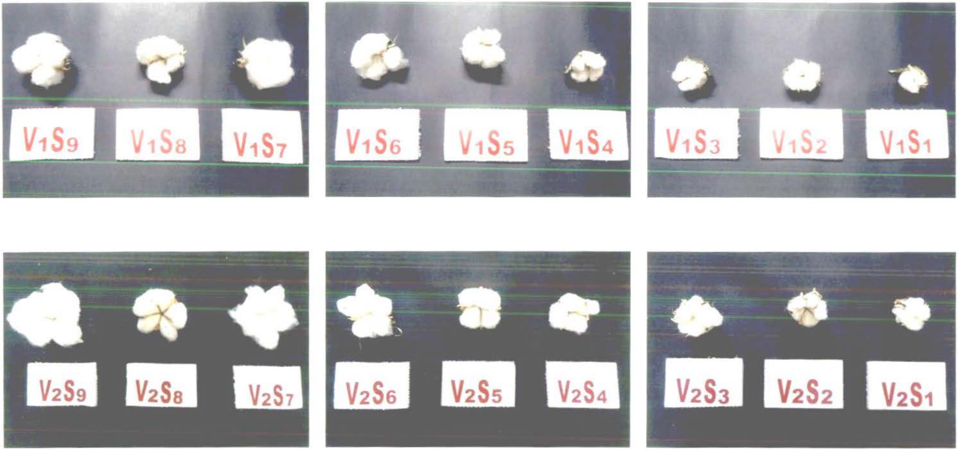


Plate 2. Effect of Glycine Betaine on boll size of cotton under moisture stress

The increase in boll size due to exogenous application of glycine betaine under moisture stress is might be attributed to the increased leaf area (Table 2) and improved photosynthetic efficiency (Table 11) which results in to better carbohydrate accumulation. It then might have helped to reduce inter-boll competition which results in to increased individual boll weight. Agboma et al. (1997) also found similar results of increase in seed number and seed size (larger seeds) with exogenous application of Glycine betaine.

4.4.5 Boll weight per plant (g)

The data on, boll weight per plant under moisture stress is presented in Table 12 and depicted in Fig. 11B. The difference between boll weight per plant. In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), found statistically significant. V_2 (22.35 g) found to be superior over V_1 (21.43 g).

In case of factor B (Glycine betaine treatment) treatment S_9 (32.76 g) recorded significantly higherboll weight per plant, followed by S_7 (29.66 g), S_8 (27.84 g), S_6 (24.52g), whereas treatment S_1 (11.27 g) the control recorded least single boll weight amongst all the treatments. However, the interaction effect between varieties and treatments found non significant.

The increase recorded in boll weight per plant might be attributed to higher levels of the major yield attributing factors like single boll weight, number of bolls per plant, morphological characters like number of monopodial and sympodial branches, (Faqir *et al.*, 1984 and Basu and Bhatt, 1987).

4.4.6 Seed cotton yield (SCY) per plant (g)

The data on, SCY per plant (g) under moisture stress is presented in Table 12 and depicted in Fig. 11B. The difference between SCY per plant found statistically significant. In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), the differences found statistically significant. V_2 (19.48 g) found statistically superior over V_1

(18.56 g). In case of factor B (Glycine betaine treatment) treatment S_9 (28.86 g) recorded significantly higher SCY per plant, followed by S_7 (25.96 g), S_8 (24.32 g), S_6 (21.31 g), and the treatment S_1 (9.39 g) recorded least SCY per plant amongst all the treatments studied. However, the interaction effect between varieties and treatments found non significant.

The major factors attributed for the differences in the yield of seed cotton are the yield attributes viz., boll weight (g), number of bolls per plant, number of seeds per plant and seed weight per plant, morphological characters like number of monopodial branches, number of sympodial branches and plant height (Reddy and Rao, 1981). Higher SCY per plant due to exogenous application of Glycine betaine might have been attributed to increase in these aspects which contributes yield in cotton. Similar results of increase in yield were recorded by Naidu *et al.* (1995), Gorham *et al.* (1998_a) Patil *et al.* (2009) in cotton.

4.4.7 Test weight (g)

The data on, test weight under moisture stress is presented in Table 12 and depicted in Fig. 11B. The difference between test weight.

In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), found statistically significant. V_2 (6.81 g) found to be superior over V_1 (6.64 g). In case of factor B (Glycine betaine treatment) treatment S_9 (7.25 g) recorded significantly higher test weight, whereas treatment S_1 (6.12 g) recorded least test weight amongst all the treatment. The treatments [S_7 and S_8], [S_8 and S_6], [S_5, S_4 and S_2], [S_4 and S_2], [S_2 and S_3] were remained at par amongst themselves for test weight. The interaction effect between varieties and treatments found non significant.

Seed weight and oil percentage are governed by moisture in cotton (Thind, 2007). An increase recorded in test weight might be due to the fact that exogenously applied Glycine betaine might have maintained photosynthetic activity by means of osmotic adjustment, the organells and cytoplasmic activities takes place at about normal pace and help plant to perform better in terms of growth, photosynthesis and assimilate partitioning to seed filling (Ludlow and Muchow, 1990; Subbarao *et al.*, 2000 and Hussain *et al.*, 2009). Shakoor (2010) also reported positive relation between higher RWC in cotton with 100 seed weight . The present results are in conformity with Hussain *et al.* (2008; 2010), Mahmood *et al.* (2009) in sunflower.

4.4.8 Harvest index (%)

The data (Table 12) revealed that, The difference between harvest index In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), found statistically significant. The variety V_2 (29.04 %) found statistically superior over V_1 (27.82 %). In case of Glycine betaine treatment, S_9 (37.70 %) recorded significantly higher harvest index, followed by S_7 (35.09 %), S_8 (33.37 %), S_6 (30.91), whereas treatment S_1 (19.43) the control recorded least harvest index amongst all the treatment. However, the interaction effect between varieties and treatments found non significant.

4.5 Fibre properties

4.5.1 2.5% span length (mm)

The data recorded for 2.5% span length in cotton under moisture stress is presented in Table 13. The data indicated that factor A (varieties, V_1 and V_2) factor B (Glycine betaine treatment) and interaction effect found statistically non significant in cotton under moisture stress. The present data revealed that there is no significant effect of glycine betaine and span length of cotton. However, the data recorded implies to only one season crop growth, which needs further confirmation on multilocation basis.

4.5.2 Uniformity ratio

The data recorded for uniformity ratio in cotton under moisture stress is presented in table 13. The data indicated that the differences for factor A (varieties, V_1 and V_2) factor B (Glycine betaine treatment) and interaction effect found statistically non significant in cotton under moisture stress. The data presented implies to only one season crop growth.

Table 13. Effect of exogenous application of Glycine betaine on fibre properties of cotton under moisture stress.

Treatment(s)	Fibre properties				
	2.5% span length (mm)	Uniformity ratio	Fibre strength (G/ tex)	Elongation (%)	Fibre fineness
FACTOR "A" (VARIETY)					
Bramha BG (I)	25.66	55.41	17.70	5.69	3.87
Mallika BG (I)	25.67	55.41	17.75	5.71	3.87
'F' test	NS	NS	NS	NS	NS
SE±	0.06	0.21	0.07	0.03	0.01
CD±	-	-	-	-	-
FACTOR "B" Glycine betaine Treatment(s)					
S_1 - Control	25.43	55.00	16.87	5.62	3.685
S_2 -2.5% (w/w) Glycine betaine seed treatment.	25.53	55.50	17.07	5.68	3.725
S_3 - 5% (w/w) Glycine betaine seed treatment.	25.68	55.33	16.97	5.70	3.749
S_4 - 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	25.48	55.33	16.97	5.78	3.720
S_5 - 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	25.60	55.67	17.07	5.71	3.737
S_6- 2.5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	25.65	55.50	18.50	5.63	4.027
S_7 - 2.5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	25.93	55.17	18.57	5.70	4.071
S_8 - 5% (w/w) Glycine betaine seed treatment + 1% (w/v) Glycine betaine foliar spray at 15 days after germination.	25.70	55.67	18.45	5.76	4.035
S_9 - 5% (w/w) Glycine betaine seed treatment + 2% (w/v) Glycine betaine foliar spray at 15 days after germination.	25.95	55.50	18.62	5.74	4.074
'F' test	NS	NS	Sig	NS	Sig
SE±	0.13	0.45	0.13	0.06	0.019
CD±	-	-	0.39	-	0.055
INTERACTION "AXB"					
'F' test	NS	NS	NS	NS	NS
SE±	0.18	0.63	0.19	0.09	0.027
CD±	-	-	-	-	-

4.5.3 Fibre strength (g/tex)

The data on, fibre strength (Table 13) under moisture stress revealed that, the difference between fibre strength In case of factor A, i.e. plant varieties V_1 (Bramha BG I) and V_2 (Mallika BG I), found statistically non significant. In case of factor B (Glycine betaine treatment) treatment S_9 (18.62 g/tex) recorded longer fibre strength, whereas treatment S_1 (19.43) recorded minimum fibre strength amongst all the treatment. The treatments, [S_9, S_7, S_6 and S_8], [S_5, S_2, S_4, S_3 and S_1] recorded at par fibre strength. The interaction effect between varieties and treatments also found non significant. The data recorded implies to only one season crop growth, which requires further confirmation on multilocation basis.

4.5.4 Elongation (%)

The data recorded for 2.5% span length in cotton under moisture stress is presented in table 13. The data indicated that factor A (varieties, V_1 and V_2), factor B (Glycine betaine treatment) and interaction effect found statistically non significant.

4.5.5 Fibre fineness

The data on, fibre fineness under moisture stress is presented in Table 13. The difference between fibre fineness In case of factor A, found statistically non significant. In case of factor B (Glycine betaine treatment) treatment S_9 (4.074) recorded longer fibre fineness, whereas treatment S_1 (3.685) recorded least fibre fineness amongst all the treatment. The treatments, [S_9, S_7, S_6 and S_8], [S_5, S_2, S_4, S_3 and S_1] remained at par amongst themselves for fibre fineness. However, the interaction effect between varieties and treatments found non significant. The data recorded implies to only one season crop growth. The present findings needs further confirmation.

CHAPTER V

SUMMARY AND CONCLUSIONS

An experiment was conducted to investigate "Effect of exogenous application of Glycine betaine on cotton (*Gossypium hirsutum L.*) under moisture stress" on growth and yield and to asses physio-chemical parameters indicating moisture stress tolerance in cotton. Experiment was laid out on the experimental field of Cotton Research Unit, Post Graduate Institute, Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola situated at an elevation of 307.4 meter above the mean sea level at $22^0 42'$ north latitude and $77^0 02'$ East longitude and has subtropical climate, during the *Kharif* season of 2011-12. The experiment was laid out in Factorial Completely Randomised design, with 18 treatment combinations comprising different levels of two factors. Factor A consists two commercial Bt-cotton cultivars (V_1) Bramha BG I and (V_2) Mallika BG I and nine different levels of Factor B, which comprises $\mathbf{S_1}$ (control) and eight treatments of Glycine betaine $\mathbf{S_2}$ [2.5% (w/w) GB seed treatment], $\mathbf{S_3}$ [5% (w/w) GB seed treatment], $\mathbf{S_4}$ [1% (w/v) GB foliar spray at 15 DAG], $\mathbf{S_5}$ [2% (w/v) GB foliar spray at 15 DAG], $\mathbf{S_6}$ [2.5% (w/w) GB seed treatment + 1% (w/v) GB foliar spray at 15 DAG], $\mathbf{S_7}$ [2.5% (w/w) GB seed treatment + 2 % (w/v) GB foliar spray at 15 DAG], $\mathbf{S_8}$ [5% (w/w) GB seed treatment + 1% (w/v) GB foliar spray at 15 DAG], $\mathbf{S_9}$ [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG].

The experiment was conducted under open shade conditions. The pot (polythene bags) culture was used. Each pot (polythene bag size of 90cm x 60cm x 60cm) was filled by 20 kg pot mixture i.e. Soil + FYM + Sand (4:2:1). Two identical sets (one for recording morphological and biochemical observations and another for phonological parameters, yield and its desirable traits) was arranged, comprising of all treatments and three replications each. For first 15 days after germination pots (polythene bags) were irrigated daily and the irrigation level was maintained at field capacity. After 15 DAG, moisture stress was imposed by providing irrigation to the pots (polythene bags) by the interval of 21 days. At the time of irrigation the pots (polythene bags) were irrigated up to the field capacity. However, the polythene bags were punctured at lower periphery to improve the drainage.

The observations were recorded on various morphological parameters (Plant height, Leaf area, SLW and dry matter production per plant) at Square initiation, 50% flowering, 50% boll bursting and maturity. Phenological parameters such as Days to 50% flowering, 50% boll bursting and maturity. Biochemical parameters (CSI, Stomatal frequency, RWC, Proline content, NR activity and Photosynthetic efficiency) were recorded at squaring, 50% flowering, 50% boll bursting. Whereas, yield and yield attributing characters were recorded at maturity of the crop and then after fibre properties. The results are summarized as under.

5.1 Morphological parameters

The morphological parameters like plant height, leaf area, SLW and dry matter production per plant differed significantly at all observational growth stages of plant. For factor B, the treatment S_9 [*5% (w/w) glycine betaine seed treatment + 2% (w/v) glycine betaine foliar spray at 15 days after germination*] recorded maximum plant height (100.47 cm) at maturity whereas, treatment S_1 (control) recorded minimum (70.32 cm) amongst all treatments. However, the varietal difference for plant height found nonsignificant.

In respect of leaf area per plant for factor B, S_9 [*5% (w/w) glycine betaine seed treatment + 2% (w/v) glycine betaine foliar spray at 15 days after germination*] recorded maximum leaf area (1334.12 cm^2) at 50% boll bursting stage. Whereas, S_1 (control) recorded minimum (965.32 cm^2) leaf area amongst all treatments. In concerned with factor A, the variety V_2 (Mallika BG I) found superior for the trait of leaf area per plant, it recorded (1196.49 cm^2) over V_1 (Bramha BG I) (1191.41 cm^2)

The highest SLW was recorded at 50% flowering stage, for factor B, treatment S_9 [*5% (w/w) glycine betaine seed treatment + 2% (w/v) glycine betaine foliar spray at 15 days after germination*] recorded maximum SLW (3.536 mg/cm^2) however, S_1 (control) recorded minimum SLW (3.134 mg/cm^2) amongst all treatment. In factor A, the maximum SLW was found in V_2 (Mallika BG I) (3.363 mg/ cm^2) over V_1 (Bramha BG I) (3.347 mg/ cm^2).

At maturity maximum dry matter production was recorded by treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] accumulated maximum dry matter (53.571 g) and S_1 (control) recorded minimum (33.597 g) amongst all treatments, whereas, variety V_2 (Mallika BG I) (45.691 g) recorded highest dry weight and minimum was recorded in V_1 (Bramha BG I) (45.429 g).

5.2 Phenological parameters

It was observed that, the Phenological parameters (Days to 50% flowering, 50% boll bursting and maturity) recorded non significant difference in terms of factor A.

In terms of factor B the significant difference were recorded for all phenological parameters. For days to 50% flowering S_7 [*2.5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG*] recorded minimum (64.67) DAS and was found statistically at par with S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] which recorded (65.17 DAS) whereas, S_1 (control) took highest number of DAS (70.67) for 50% flowering.

For days to 50% boll bursting S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] took minimum (126.33) DAS whereas, S_1 (control) took highest number of DAS (130.44) for 50% boll bursting.

For days to maturity S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] took minimum (152.17) DAS whereas, S_1 (control) took highest number of DAS (159.17) for attaining maturity.

5.3 Biochemical parameters

In respect of biochemical parameters, the parameters CSI, RWC, proline content, NR activity and photosynthetic efficiency differed significantly at all three observational growth stages.

In case of CSI, S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] recorded maximum CSI (68.437) at 50% flowering stage, whereas, S_1 (control) recorded minimum (46.082) CSI amongst all treatments. However, V_2 (Mallika BG I) (57.945) found significantly superior over V_1 (Bramha BG I) (57.383)

In case of RWC, S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] recorded maximum RWC (83.13 %) at square initiation stage, whereas, S_1 (control) recorded minimum (66.83 %) RWC amongst all treatments. However, V_2 (Mallika BG I) (76.20 %) found significantly superior over V_1 (Bramha BG I) (75.80 %).

In case of proline content, S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] recorded maximum proline content (54.78 µg/g) at square initiation stage, whereas, S_1 (control) recorded minimum (34.36 µg/g) proline content amongst all treatments. However, V_2 (Mallika BG I) (46.72 µg/g) found significantly superior over V_1 (Bramha BG I) (46.46 µg/g).

In case of NR activity S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] recorded maximum NR activity (1.8061 µmoles/g/hr) at 50% flowering stage, whereas, S_1 (control) recorded minimum (0.7501 µmoles/g/hr) NR activity amongst all treatments. However, V_2 (Mallika BG I) (1.2495 µmoles/g/hr) found significantly superior over V_1 (Bramha BG I) (1.1904 µmoles/g/hr).

In case of photosynthetic efficiency S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] recorded maximum photosynthetic efficiency (0.860 Fv/Fm) at square initiation stage, whereas, S_1 (control) recorded minimum (0.752 Fv/Fm) photosynthetic efficiency amongst all treatments. However, V_2 (Mallika BG I) (0.807 Fv/Fm) found significantly superior over V_1 (Bramha BG I) (0.795 Fv/Fm).

Regarding stomatal frequency the non significant difference for both Factors A and B is recorded at all subsequent crop growth stages.

5.4 Yield and yield attributes

In case of number of monopodia, owing to factor B, treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] (1.67) recorded maximum number of monopodia. Whereas, control S_1 (0.40), recorded minimum number of monopodia amongst all treatments. In concerned with factor A, V_2 (Mallika BG I) (1.09) found significantly superior over V_1 (Bramha BG I) (0.89).

In case of number of sympodia, owing to factor B, treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] (15.77) recorded maximum number of sympodia, whereas, control S_1 (9.30), recorded minimum number of sympodia amongst all treatments. In concerned with factor A, V_2 (Mallika BG I) (12.68) found significantly superior over V_1 (Bramha BG I) (12.27).

In case of number of bolls per plant, owing to factor B, treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] (10.73) recorded maximum number of bolls per plant, whereas, control S_1 (5.43), recorded minimum number of bolls per plant amongst all treatments. However, the varietal difference for plant height found non significant.

In case of single boll weight, owing to factor B, treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] (2.69 g) recorded higher single boll weight, whereas, control S_1 (1.73 g), recorded minimum single boll weight amongst all treatments. In concerned with factor A, V_2 (Mallika BG I) (2.22 g) found significantly superior over V_1 (Bramha BG I) (2.15 g).

In case of boll weight per plant, owing to factor B, treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] (32.76 g) recorded higher boll weight per plant, whereas, control S_1 (11.27 g), recorded minimum boll weight per plant amongst all treatments. In concerned with factor A, V_2 (Mallika BG I) (22.35 g) found significantly superior over V_1 (Bramha BG I) (21.43 g).

In case of seed cotton yield per plant, owing to factor B, treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] (28.86 g/plant) recorded higher seed cotton yield per plant, whereas, control S_1 (9.39 g/plant), recorded minimum seed cotton yield per plant amongst all treatments. In concerned with factor A, V_2 (Mallika BG I) (19.48 g/plant) found significantly superior over V_1 (Bramha BG I) (18.56 g/plant).

In case of test weight, owing to factor B, treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] (7.25 g) recorded higher test weight, whereas, control S_1 (6.12 g), recorded minimum test weight amongst all treatments. In concerned with factor A, V_2 (Mallika BG I) (6.81 g) found significantly superior over V_1 (Bramha BG I) (6.64 g).

In case of harvest index, owing to factor B, treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] (37.70) recorded higher harvest index, whereas, control S_1 (19.43), recorded minimum harvest index amongst all treatments. In concerned with factor A, V_2 (Mallika BG I) (29.04) found significantly superior over V_1 (Bramha BG I) (27.82).

5.5 Fibre properties

The effect of exogenous application of Glycine betaine over the fibre properties like fibre strength and fibre fineness found significant. The fibre strength and fibre fineness increased with increase in concentration of Glycine betaine S_9 recorded maximum. Whereas, S_1 recorded minimum values. The other fibre properties (2.5% span length, Uniformity ratio and Elongation) were not affected by exogenous application of Glycine betaine. However, in terms of factor A statistical difference between both the varieties V_1 (Bramha BG I) and V_2 (Mallika BG I) for above recorded fibre properties found non significant.

Conclusions

The investigation "Effect of exogenous application of Glycine betaine on cotton (*Gossypium hirsutum L.*) under moisture stress" concluded that the parameters like leaf area, dry matter accumulation, SLW, CSI, RWC, Proline content, NR activity and photosynthetic efficiency differed significantly with respect of both factors A and B, they might be associated with the moisture stress tolerance in cotton

The recorded data shows that exogenous application of Glycine betaine successfully averted the negative effects of moisture stress on growth and yield of cotton. The Glycine betaine treatment S_9 [5% (w/w) GB seed treatment + 2% (w/v) GB foliar spray at 15 DAG] found superior amongst all the treatments, which recorded high harvest index (37.70 %) and might have helped for elevating the negative effects of moisture stress in cotton as compared to control (19.43 %). However, the Variety V_2 (Mallika BG I) found promising than V_1 (Bramha BG I)

There is a need to standardize the dosage and concentration of Glycine betaine along with the stage of spray and number of sprays required for elevating moisture stress in cotton. However, the data recorded implies only one season crop growth which needs further confirmation on multilocation basis.

CHAPTER VI

LITERATURE CITED

Acevedo, E., E. Fereres, T.C. Hsiao, and D.W. Henderson. 1979. Diurnal growth trends, water potential, and osmotic adjustment of maize and sorghum leaves in the field. Plant Physiol.64:476-480

Adarsha, T.S., B.C. Patil and V.H. Ashvathama, 2003. Studies on growth, phenology and productivity of different cotton genotypes. Indian J. Plant Phisiol. 8(1): 79-81

Agboma, P., Sinclair, T., Jokinen, K., Peltonen-Sainio, P. and Pehu, E., 1997. An evaluation of the effect of exogenous glycine betaine on the growth and yieldof soybean. Field Crops Res. 54: 51–64

Anonymous, 2012. AICCIP annual report 2011-12.

Arora, D.K. and Seema Gupta. 2003. *In* physiological processes in plants. Eds. Anmol publication Pvt. Ltd. pp 92-93

Ashraf, M. and M. R. Foolad, 2007. Roles of glycine betaine and proline in improving plant abiotic stress resistance. Environmental and Experimental Botany. 59: 206-216

Baraiya, B. R., S. K. Barde, H.C. Singhal and Lekharam. 2009. Genetic evaluation of *Gossypium hirsutum* genotypes for yield, drought parameters and fibre quality. Annals of Plant Physiology, 23(2): 144-148

Barketali, S., Channa, M. H. And Ahamed, M., 1982, Correlation studies in *G. hirsutum* cotton. *Pakistan cotton*, 26:39-51.

Barrs, M. D. and P. E. Weathery, 1962, A re-examination of relative turgidity for estimating water deficit in leaves. *Australian Journal of Biological Sciences*, 15: 413-428.

Basal, H., C.W. Smith, P.S. Thaxton and J.K. Hemphill, 2005. Seedling and drought tolerance in upland cotton. Crop Sci. 45: 766 - 771.

Basu, A. K. and M. G. Bhat, 1987, Variability for harvest index in upland cotton. *Indian Journal of Agriculture Sciences*, 57:219-288.

Bates, L. S., R. P. Waldren, and I. D., Terre, 1973, Rapid determination of free proline in water stress studies. *Plant and Soil*, 39 : 205-208.

Beckar, T. and H. Fock. 1986. The activity of nitrate reductase and the pool size of some amino acids and some sugers in water stressed maize leaves. Photosynthesis Res. 8(3): 267-274

Bharadwaj, S. N. and K. P. Singh, 1988, Relation between specific leaf weight, leaf conductance, carbon exchange rate and chlorophyll content in genotypes of upland cotton (*G. hirsutum* L.). *Indian Journal of Plant Physiology*, 31 : 102-104.

Blum, A. And Ebercon, A., 1976, Genotypic responses in sorghum to drought stress III. Free proline accumulation and drought resistance. *Crop Sciences*, 16 : 425-431.

Boyer, J. S., 1982: Plant productivity and environment. Science 218, 443–448.

Cassandra, R. And Oosterhius, 1999, Effect of glycine betaine and water regime on diverse cotton cultivars. *Proceedings of the 2000 Cotton Research Meeting and University of Arkansas Agricultural Experiment Station Special Report*, pp. 198-199.

Channaveeraiah, D. M., 1983, Influence of morhpo-physiological characters on productivity of rainfed cotton (*G. hirsutum L.*) *M. Sc. (Agri.)* Thesis, University of Agricultural Sciences, Bangalore.

Chaudhary, S.D. and M. Syed. 1978. Effect of moisture stress on growth , grain yield and germination of seed of sorghum. Sorghum News Letter 21: 50-57

Chetti, M. B., S.M. Hiremath and M. Kalpana. 2002,. Physiological approaches for enhancing productivity potential under drought condition Eds. Saraswathi Printers, Dharwad. pp: 99-101

Dhopte A.M. and M. Livera. 1989. Estimation of Stomatal index and stomatal frequency with replica method . Useful Techniques for plant scientists Eds., pp: 278-279

Donald, C.M., 1962, In search of yield. *Journal of Australian International Agricultural Sciences*, 28: 171-178.

Eaton, F. M., 1955. Physiology of cotton plant. Annual reviews of plant physiology, 6: 299-328.

Faqir M. A., Abdur, R. A. And Khan, M. A., 1984, Association of yield with various economic characters in *G. hirsutum L. Pakistan Cottons*, 28: 127-134.

Farooq, M., S. M. A. Basra, A. Wahid, Z. A. Cheema, M. A. Cheema, and A. Khaliq, 2008. Physiological role of exogenously applied glycinebetaine in improving drought tolerance of fine grain aromatic rice (Oryza sativa L.). J. Agron. Crop Sci. 194, 325–333.

Genard, H., Le Saos, J., Hillard, J., Tremolieres, A., Boucaud, J., 1991. Effect of salinity on lipid composition, glycine betaine content and photosynthetic activity in chloroplasts of Suaeda maritime. Plant Physiol. Biochem. 29, 421–427.

Giri, A.N. and U.C. Upadhyay, 1980, Correlation and regression studies ion H-4 upland cotton under different plaguing pattern and inter-cropping system. *Indian Journalof Agricultural Sciences*, 50: 907-910.

Gorham, J., K. Jokinen, M. N. Malik, and I. A. Khan, 1998a, Glycine betaine treatment improves cotton yield in field trails. In : *Proceedings of World Cotton Research Conference-2*, Athens, Greece, September, 6-12, pp. 621-627

Gorham, J., J. Bridges, K. Jokinen, and K. Tiihonen, 1998b, Exogenously applied glycine betaine is not rapidly translocated in cotton. In : *Proceedings of World Cotton Research Conference-2*, Athens, Greece, September, 6-12, pp. 628-631.

Hanson, A.D., and N.A., Scott, 1980. Betaine synthesis from radioactive precursors in attached, water-stressed barley leaves. Plant Physiol. 66, 342–348.

Hatfield, J.L.,; J.E. Quisenberry, and R.E. Dilbeck. 1987. Use of canopy temperature to identify water conservation in cotton germplasm. Crop Sci. 26:269-273

Hsiao, T.C. 1973. Plant responses to water stress. Annu. Rev. Plant Physiol. 24: 519-570.

Hussain, M., M. A. Malik, M. Farooq, M. Y. Ashraf, and M. A. Cheema, 2008: Improving drought tolerance by exogenous application of glycinebetaine and salicylic acid in sunflower. J. Agron. Crop Sci. 194, 193–199.

Hussain, M., M. A. Malik, M. Farooq, M. B. Khan, M. Akram, and M. F. Saleem, 2009: Exogenous glycinebetaine and salicylic acid application improves water relations, allometry and quality of hybrid sunflower under water deficit conditions. J. Agron. Crop Sci. 195, 98–109.

Hussain, M., M. Farooq, K. Jabran and A. Wahid. 2010. Foliar application of glycine betaine and salicyclic acid improves growth, yield and water productivity of hybrid sunflower planted by different sowing methods. J. Agronomy and crop science. 196: 136-145

Jones, M.M., and N.C. Turner. 1978. Osmotic adjustment in leaves of sorghum in response to water deficits. Plant Physiol. 61:122-126.

Kale R. V. 2009. Physiological studies on drought tolerance in promising genotypes of pigeon pea (*Cajanus cajan* L.). M.Sc. Thesis, Dr. PDKV, Akola (M.S.)

Kar, M., B.B. Patro, C.R. Sahoo and B. Hota. 2005. Traits related to drought resistance in cotton hybrids. Ind. J. Pl. Physiol. 10(4): 377-380.

Karademir, C., E. Karademir, R. Ekinci and K. Berekatoglu, 2011. Yield and fibre quality properties of cotton (*Gossypium hirsutum* L.) under water stress and non stress conditions. African J. of Biotechnology 10(59): 12575-12583

Kavikishor PB, Hong Z, Miao G, Hu C, Verma DPS (1995) Overexpression of D1-pyrroline-5-carboxylate synthetase increases proline overproduction and confers osmotolerance in transgenic plants. Plant Physiol 108: 1387–1394

Khorgade, P.W. and A. P., Ekbote, 1980, Path coefficient analysis in upland cotton. *Indian Journal of Agricultural Sciences*, 50: 6-8.

Koti Shailaja, 1997. Effect of glycine betaine in the alleviation of Abiotic stresses in groundnut genotypes. Ph.D. thesis, ANGRAU, Hyderabad (A.P.)

Kumar, R and R. Kurju, 2003. Role of secondary traits in improving drought tolerance during flowering stage in rice. Indian J. Plant Physiol. 8(3): 236-240

Kush, G. S., 1998, Strategies For increasing crop productivity. Proceedings Second International *Crop Science* Congress : *Crop Productivity and Sustainability- Shaping the Future*, Oxford and IBM Publishing Company Private Limited, New Delhi, pp. 19-43.

Landiver, J.A., D. N. Baker, And J. N. Jenkins, 1983, Application of Gossym to genetic feasibility studies II Analysis of increasing photosynthesis, specific leaf weight and longevity of leaf in cotton. *Crop Science*, 23:504-510.

Laude, H.M., 1971. Drought influence on physiological process and subsequent growth. *In* Larson L.K. and J.D. Eastin (*Eds)* Drought injury and resistance in crops. CSSA Wisconsin, USA. pp. 45-56

Ludlow, M. M., and R. C. Muchow, 1990: A critical evaluation of traits for improving crop yield in water limited environments. Adv. Agron. 43, 107–153.

Ma, X. L., Y. J. Wang, S. L. Xie, C. Wang, and W. Wang, 2007: Glycinebetaine application ameliorates negative effects of drought stress in tobacco. Russian J. Plant Physiol. 54: 472–479.

Mahmood, Tahir, Muhammad Ashraf and Muhammad Shahbaz. 2009. Does exogenous application of glycine betaine as pre-sowing seed treatment improve growth and regulate some key physiological attributes in wheat plants grown under water deficit conditions? Pak. J. Bot. 41(3): 1291-1302

Marani, A. 1973. Effects of soil moisture stress on two varieties of upland cotton in Israel. Exp. Agri. 9 (1): 121-128.

May, L.H. and E.L. Milthorpe. 1962. Drought resistance of crop plants. Field Crop Abstracts, 15(3): 171-179

Mottaram M., 1999. Betaine osmolyte regulator of cotton. United States Patent no. 5,952,267. Date: 14 Sep. 1999.

Naidu, P., Cameron, D. F. and S. V. Konduri, 1995, Improving drought tolerance of cotton by glycine betaine applicaton and selection. *Proceedings of the* 9^{th} *Australian Agronomy Conference*, pp. 1-5.

Oosterhuis, D.M., S.D., Wullschleger, and J.M. Stewart, 1987. Osmotic adjustment in commercialcultivars and wild types of cotton. Agronomy Abstracts. pp: 97

Panse, V.G. And P.V. Sukhatme, 1967, *Statistical methods for agricultural workers*. ICAR publication, New Delhi, pp. 167-174.

Patil, M. D. 2005. Drought mangement and molecular characterization of cotton genotypes. M.Sc. (Agri) Thesis, UAS, Dharwad (Karnataka.)

Patil. M.D., D.P. Biradar and V.C. Patil. 2009. Agronomic response of cotton as influenced by drought management practices. Karnataka j. Agric. Sci. , 22(4): 755-758

Patil, M.D., D.P. Biradar, V.C. Patil and B.S. Janagoudar, 2011. Response of cotton genotypes to drought mitigation practices. American-Eurasian J. Agric. & Environ. Sci., 11(3): 360-364

Patterson, D. T., 1989, Growth and water relations of cotton (*G. hirsutum* L.) spurred anola (*Anoda cristata*) and velvet leaf (*Abutilou theophrati*) during stimulated drought and recovery. *Field Crops Abstract*, 41 : 132.

Pawar, H.C., R. M. Naik, R. D. Satbhai and S.S. Mehetre. 2010. Proline, P5CS activity and glycine betaine content in intra-hirsutum (HxH), Inter-specific (HXB) and *G. arborium* cultivars under water stress. Indian J. Plant Physol. 15(2): 125-130

Pettigrew, W. T., 2004b. Physiological consequences of moisture deficit stress in cotton. Crop Science. 44: 1265-1272

Pettigrew, W. T., 2004a, moisture deficit effects on cotton lint yield, yield componants and boll distribution, Agron. J., 96: 377-383.

Quisenberry, J.E. and B. Roask, 1976. Influence of indeterminate growth habit on yield and irrigation water use efficiency in upland cotton. Crop Science, 16: 762-765

Quisenberry, J. E., C.E. Wendt, J.D. Berun and B.L. McMichal, 2011. Potential for using leaf turgidity to select drought tolerance in cotton. Crop Sci, 52: 294-298.

Rajeshwari, V.R. 1995. Evaluation of cotton genotypes for drought tolerance under rainfed condition. Ann. Pl. Physiol. 2: 109-112.

Rasulou, B. K. M. and K. A. Asrorov, 1991, Dependence of intensity of photosynthesis on specific leaf weight in different species of cotton. *Physiologia Photosynthetica*, pp. 270-283.

Reddy, A. S. and T. A. Rao, 1981, Effect of insecticides and plant hormones on growth and yield of cotton. Pesticides, 15 : 27-28.

Rhodes, D. and A.D. Hanson, 1993. Quaternary ammonium and tertiary sulfonium compounds in higher-plants. Annu. Rev. Plant Physiol. Plant Mol. Biol. 44:357–384.

Robinson, S.P and G.P. Jones, 1986. Accumulation of glycine betaine in chloroplasts provides osmotic adjustment during salt stress. Aust. J. Plant Physiol.13, 659–668.

Roy, B. and A.K. Basu, 2009. Eds, Abiotic Stress Tolerance In Crop Plants. New India Publishing Agency, New Delhi pp 13-255

Sarwar, M. K. Shahazad, Ihsan Ullah, Meheboob-Ur-Raheman, M.Yasin Ashraf and Yusuf Zafar. 2006. Glycine betaine accumulation and its relation to yield and yield components in cotton genotypes grown under water deficit condition. Pak. J. Bot. 38(5): 1449-1456

Serraj, R. and T.R. Sinclair, 2002. Osmolyte accumulation : Can it really help increase crop yield under drought conditions? Plant Cell Environment. 25:333-341.

Shakoor Muhammad S. 2010 Genetics of physio-morphological traits in cotton (*Gossypium hirsutum*) under drought and normal conditions. Ph. D. Thesis, University of Agriculture Faisalabad Pakistan.

Shewale J. W. 2007. Studies on characteristics for drought tolerance in upland cotton (*Gossypium hirsutum* L.). M.Sc. Thesis, Dr. PDKV, Akola (M.S.)

Shimshi, D. and A. Marani. 1971. Effects of soil moisture stress on two varieties of upland cotton in Israel. Exp. Agri. 7 (1): 225-239.

Simpson, G. M., 1981, *Water Stress on Plants,* New York, USA, pp. 1-33.

Singh, D. 1992. Nitrate reductase activity in relation to drought tolerance in cotton. AICCIP Silver jubilee symposium. A-96

Singh, S.B., D. Singh and S.S. Narayanan. 1996. Variation in physio-morphological characters related to drought tolerance in cotton (*Gossypium* spp.). Ind. J. Agric. Sci. 66 (6): 357-359.

Sinha, S. K. and D. J. Nicholas. 1981. Nitrate reductase *In* . L.G. Paleg and D.A. Aspinall Eds. physiology and biochemistry of drought resistance in plant, Academic press, London: 145-169.

Srivastava, H.S., 1974. *In vivo* activity of nitrate reductase in maize seedlings , Indian J. Biochem. Biophysics., 11: 230-232.

Subbarao, G. V., N. H. Nam, Y. S. Chauhan, and C. Johansen, 2000: Osmotic adjustment, water relations and carbohydrate remobilization in pigeonpea under water deficits. J. Plant Physiol. 157, 651–659.

Sung, F.J.M., 1981. The effect of leaf water ststus on stomatal activity, transpiration and nitrate reductase activity, of sweet potato. Agricultural water management. 4: 465-470.

Taiz, L., and E. Zeiger, 2005. Plant Physiology. (4th), Sinauer Associates, Inc.; ISBN 978- 0878938568, Sunderland, Massachusetts. pp: 593.

Thind, S. K., 2007. Advances in cotton physiology. Satish serial publishing house, New Delhi, India. 1-153.

Weising, K., M. Nybon, K. Wolf and W. Meyer, 1995. DNA finger printing plants of fungi. Plant Genetic Resources. Newsletter. 97:3-39.

Weretilnyk, E.A., Bednarek, S., McCue, K.F., Rhodes, D., Hanson, A.D., 1989. Comparative biochemical and immunological studies of the glycine betaine synthesis pathway in diverse families of dicotyledons. Planta 178, 342–352.

Wyn Jones, R.G., Gorham, J., McDonnell, E., 1984. Organic and inorganic solute contents as selection criteria for salt tolerance in the Triticeae. In: Staples, R., Toennissen, G.H. (Eds.), Salinity Tolerance in Plants: Strategies for Crop Improvement. Wiley and Sons, New York, pp. 189–203.

Xing WeiBing, and Rajashekar, C.B., 1999. Alleviation of water stress in beans by exogenous glycine betaine. Plant Sci. 148, 185–192

Xu, Zhenzhu and Guangsheng Zhou, 2008. Responses of leaf stomatal density to water status and its relationship with photosynthesis in grass. Journal of Experimental Botany, 59(2): 3317-3325.

VITA

1. Name of student : **Pande Harshal Sanjay**
2. Date of Birth : 13th March 1988
3. Name of college : Department of Agricultural Botany,
   Discipline – Plant Physiology,
   Post Graduate Institute,
   Dr. PDKV, Akola
4. Residential Address : Near Chawadi
   Old Town, Buldhana (M.S.)
   PIN-443001
   Mobile No. – 9822459882
   harshal.pande@gmail.com
5. Academic Qualification

Sr. No.	Name of degree awarded	Year in which obtained	Division / Class	Name of awarding university	Subjects
1	B.Sc. (Hort.)	2009	First	Dr. PDKV, Akola	Horticulture and Allied subjects

6. Research paper published : Nil
7. Field of interest : Research and Development

Place : Akola

Date : 14/06/2012

Signature of student

APPENDIX- I

Weekly weather data from July 2011 to January 2012 recorded at Meteorological Observatory Department of Agronomy Dr. PDKV., Akola Actual: 2011-12 Normal: 1971-2000

weeks	Dates	T MAX (°C)		T MIN (°C)		BSH (hrs)		WS (km/hr)		RH I (%)		RH II (%)		Evap (mm)		RF (mm)		CRF (mm)	Rainy Days	
		N	A	N	A	N	A	N	A	N	A	N	A	N	A	N	A		N	A
	2011																			
23	4-10	40.2	37.3	26.9	25.1	8.4	5.9	15.2	3.7	62	75	30	40	14.0	8.1	16.8	31.9	92.0	1.0	2.0
24	11-17	38.0	38.0	25.7	25.4	7.1	7.7	13.4	11.3	69	66	40	33	11.1	11.5	43.6	23.5	115.5	1.7	2.0
25	18-24	35.5	35.7	25.0	27.1	5.8	1.7	14.2	18.8	74	66	48	42	9.2	12.1	43.5	0.0	115.5	2.0	0.0
26	25-1Jul	33.8	33.1	24.3	24.8	4.8	0.7	12.8	11.9	80	77	55	45	7.4	7.2	43.4	17.2	132.7	2.2	1.0
27	2-8	33.2	34.6	24.0	25.0	4.8	3.0	12.0	8.2	81	79	58	54	6.5	7.3	39.4	43.7	176.4	2.2	3.0
28	9-15	32.3	31.4	23.8	23.8	3.8	2.4	11.2	11.3	83	88	60	59	5.5	5.1	42.8	26.4	202.8	2.5	3.0
29	16-22	31.9	30.4	23.6	24.0	4.0	0.8	10.4	7.8	84	91	63	69	5.2	4.4	52.8	58.1	260.9	2.4	2.0
30	23-29	31.3	29.8	23.3	23.8	4.0	2.8	10.8	4.8	86	89	64	67	4.8	3.6	43.4	26.0	286.9	2.6	3.0
31	30-5 Aug	30.9	30.9	23.3	24.1	3.5	3.8	10.6	8.0	86	88	67	65	4.6	4.9	49.6	17.5	304.4	2.4	2.0
32	6-12	29.9	30.5	23.0	23.7	3.2	1.0	10.9	6.1	88	87	70	64	4.1	3.6	61.0	8.5	312.9	2.8	1.0
33	13-19	30.4	30.1	23.0	23.3	4.0	3.0	12.4	3.6	87	89	67	65	4.5	4.1	35.9	47.6	360.5	2.0	3.0
34	20-26	30.4	31.3	22.8	23.4	4.1	4.0	11.9	3.6	87	94	67	65	4.3	4.3	42.5	18.9	379.4	1.9	1.0
35	27-2 Sep	30.5	28.5	22.7	23.2	4.2	0.8	9.3	4.7	87	95	66	82	4.6	3.3	42.4	46.6	426.0	2.1	4.0
36	3-9	31.0	29.8	22.5	23.2	5.3	3.1	8.6	3.3	87	92	62	68	5.3	3.3	33.6	63.1	489.1	1.5	3.0
37	10-16	32.1	29.9	22.4	23.4	6.6	5.0	8.0	2.6	85	90	57	70	5.1	4.3	22.0	21.5	510.6	1.1	2.0
38	17-23	32.9	30.9	22.4	22.5	6.8	6.1	6.4	5.0	84	89	55	57	5.2	4.7	23.7	3.5	514.1	1.4	1.0
39	24-30	33.5	32.7	22.1	22.5	7.3	5.7	5.1	3.8	84	85	50	44	5.0	5.0	24.4	0.0	514.1	1.4	0.0
40	1-7 Oct	33.7	34.9	21.2	21.3	7.6	7.7	4.8	1.4	82	79	47	35	5.4	6.1	23.4	0.0	514.1	1.1	0.0
41	8-14	34.0	35.5	19.8	21.0	8.1	5.8	4.5	1.5	78	80	40	35	5.3	5.6	13.1	0.8	514.9	0.7	0.0
42	15-21	33.7	35.7	18.3	20.1	8.2	5.6	4.6	1.0	76	80	37	29	5.3	5.4	6.1	0.9	515.8	0.4	0.0
43	22-28	33.1	34.6	16.8	15.9	8.3	7.4	4.4	1.7	74	70	34	19	5.3	6.0	7.6	0.0	515.8	0.4	0.0
44	29-4Nov	32.7	32.9	16.0	15.3	8.4	7.3	4.1	2.1	73	65	32	24	5.3	6.6	2.3	0.0	515.8	0.2	0.0
45	5-11	32.3	33.8	15.2	14.7	8.4	8.2	3.9	1.1	71	63	32	21	5.1	5.5	3.0	0.0	515.8	0.2	0.0
46	12-18	31.6	33.5	14.6	14.6	8.3	7.8	3.9	0.8	73	63	32	18	4.8	5.4	5.3	0.0	515.8	0.2	0.0
47	19-25	31.0	32.0	13.3	12.9	8.4	7.9	3.7	1.5	72	69	30	23	4.6	5.8	7.7	0.0	515.8	0.3	0.0
48	26-2 Dec	30.5	31.4	12.8	15.4	8.4	6.5	3.6	1.1	71	75	32	31	4.4	5.1	5.5	0.0	515.8	0.3	0.0
49	3-9	30.0	31.6	11.9	13.9	8.4	7.7	3.8	0.8	71	72	30	25	4.3	4.6	1.0	0.0	515.8	0.1	0.0
50	10-16	29.6	29.9	10.9	11.4	8.4	7.9	3.6	0.8	71	72	28	25	4.2	5.0	0.8	0.0	515.8	0.1	0.0
51	17-23	29.5	29.6	10.8	11.3	8.5	7.4	3.8	1.2	70	65	29	23	4.1	4.7	0.9	0.0	515.8	0.1	0.0
52	24-31	29.1	29.5	11.1	11.4	8.3	6.5	4.5	0.9	71	69	30	23	4.2	4.5	2.6	0.0	515.8	0.2	0.0
	2012																			
1	1-7 Jan	28.8	30.3	11.0	16.3	8.2	4.1	4.4	1.8	71.4	78	30.7	36	4.2	4.2	2.8	0.0	0.0	0.2	0.00
2	8-14	29.3	26.7	11.7	8.3	8.3	8.1	4.4	1.0	71.2	67	30.3	18	4.4	4.3	3.3	0.0	0.0	0.2	0.00
3	15-21	30.0	28.0	12.0	10.3	8.6	8.3	4.5	1.7	67.8	55	27.6	18	4.9	5.0	0.7	0.0	0.0	0.1	0.00
4	22-28	30.6	29.4	12.0	15.1	8.8	5.8	4.6	1.8	64.8	61	25.9	30	5.2	5.0	0.9	0.0	0.0	0.1	0.00