

## High temperature stress in cotton *Gossypium hirsutum* L.

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**Abstract.** Heat stress is among one of the limiting and ever looming threats to cotton productivity in Pakistan. This factor inflicted huge losses in the recent years. In Pakistan genotypes developed for general cultivation face very high temperature of about 50 °C during the month of June which is about 20 °C more than the optimum temperature thus retard yield to greater extent. The plant parts like buds, flowers, fiber quality traits are greatly influenced due to high temperature. This mini review partially covers effect of heat stress on cotton fiber quality, plant parts, screening procedures, genetics and biotechnological aspects related to heat stress. All the information provided in the manuscript will help to better understand the phenomena of heat stress tolerance thus will ultimately aid in the development of heat tolerant cultivars in Pakistan.

**Key words:** Heat stress, cotton fiber, Pakistan, *G. hirsutum*, supra-optimal temperature.

**Introduction.** Cotton (*Gossypium hirsutum* L.) is an important cash and fiber crop. It is recognized as major industrial crop in both developed and developing countries (Imran et al 2011, 2012). Mainly cotton is the crop of warm areas (Riaz et al 2013). Cotton performs more efficiently at 23-32 °C at which metabolic activity is maximum (Burke et al 1988). Studies showed that the maximum number of bolls and square formation occurs at 30/22 °C day/night temperatures (Reddy et al 1992a). Different genotypes respond differentially so there is no clear cut evidence about optimum temperature which is required for normal growth (Burke & Wanjura 2009). Cotton plants having thermo-tolerance mechanisms can buffer the effects of short periods of high temperature stress. High temperature is often accompanied by water deficit, solar radiation and wind which enhance the damage to the crop plant (Paulsen 1994). Brown & Oosterhuis (2005) reported that present-day cotton cultivars have no much tolerance to heat stress.

Due to environmental stress cotton crop produces only 25% of the potential yield (Boyer 1982). In Pakistan cotton grown area falls into geographical zone of high temperature where temperature reaches near 50°C in summer due to which cotton have to face heat stress (Rahman 2006). Heat stress affects the yield and fiber quality as it influence growth of the seedlings, roots, vegetative parts and also influence flowering and fruit formation (Singh et al 2007). Enhancing tolerance to heat stress in crop plants is getting more important these days. Extensive research has been done to improve heat tolerance in Pima cotton (*Gossypium barbadense* L.) and other crops like Cowpea (*Vigna unguiculata* L.), wheat (*Triticum aestivum* L.) English ivy (*Hedera helix* L.) and Kentucky bluegrass (*Poa pratensis* L.) (Kittock et al 1988; Reynolds et al 2001; Ismail & Hall 1999; Yeh & Hsu 2004; Marcum 1998). This mini review will help to understand the effect of

heat stress and mechanism of heat stress tolerance in cotton *Gossypium hirsutum* L which ultimately aid to develop heat tolerant cultivars in cotton.

**Effects of Heat Stress on Different Parts of Cotton Plant.** Larger differences between potential and actual yield are merely due to the environmental stresses at stage of floral development (Boyer 1982). High temperature influences all stages of plant life from germination to the development of reproductive parts (Paulsen 1994). Seed germination and emergence is highly dependent on environmental temperature and also in further plant development and growth. Shoot growth and development needs higher temperature than roots and temperature of 30 °C is reported as optimum for cotton (Pearson et al 1970).

**Reproduction.** In cotton development of reproductive parts is highly subtle to heat stress, before and after anthesis (Oosterhuis 2002). With increase in temperature sequence of reproduction stage get hastened i.e., the time required for the first square to appear, for first flower and first mature boll to open declined as the mean temperature for each event increased (Reddy et al 1996). Zhao et al (2005) observed that there is a significant decrease in boll retention at high temperatures. Decline in boll retention at temperature higher than 30/22°C day/night has also been reported by Reddy et al (1992b) in both Pima and Upland cotton. Further studies by Reddy et al (1995a) revealed that boll retention at a temperature above 26.6 °C day time is highly sensitive and were influenced negatively. Cotton plants under day and night temperature of 36/28 °C show 70% less boll retention than the plants grown under a 30/22 °C day/night (Zhao et al 2005). According to Reddy et al (2005) upland cotton was less affected by heat stress than Pima cotton as when daily mean temperature of 36 °C was applied, Pima Cotton varieties did not produce fruiting branches and other reproductive parts. However, Upland cotton plants produce fruiting branches and also squares are formed under high temperature stress, but it did not develop bolls successfully (Reddy et al 1992a).

Before and during flowering reproductive processes are significantly influenced by high-temperature stress that leads to the decline in fruit set. Oosterhuis (1999) reported that heat stress approximately 17 days before flowering could cause a decline in pollen viability and fertilization. McDonald & Stith (1972), Meyer (1969), Powell (1969) also reported similar results previously. In temperature sensitive male sterile lines a temperature of 32 °C 15 to 17 days before anther dehiscence induce pollen sterility and when temperature exceeds 38 °C some of the fertile lines also start showing sterile anthers (Meyer 1969). Hedhly et al (2009) and Zinn et al (2010) reported that like most of the thermo-sensitive process, sexual reproduction in one of the tolerant processes, because all of the reproductive process must happen in extremely determined way. Heat stress depending on the duration, time and intensity can restrict the fertilization by hindering male and female gametophyte development, pollen germination (Burke et al 2004; Kakani et al 2005; Jain et al 2007) and pollen tube growth (Burke et al 2004; Hedhly et al 2004; Kakani et al 2005; Snider et al 2011a). Optimum temperature for pollen germination is 28 °C, and it is highly sensitive to high temperature (Burke et al 2004). Estimated cardinal temperature for pollen tube germination is 15.0, 31.8, and 43.3C (mini. T, opt. T, and max. T, respectively) and for pollen tube length is 11.9, 28.6, and 42.9 C, evaluated for 12 cultivars of cotton (Kakani et al 2005). There is strong correlation between maximum pollen tube growth and boll retention in *G. hirsutum* and optimum temperature for pollen tube growth reported by Liu et al (2006) is 27.8 °C. During the flowering period maximum daily temperature confronted by cotton plant frequently beat the optimum temperature for effective pollen tube growth and development, sometimes it exceeds 38 °C at afternoon (Snider et al 2009). Burke et al 2004; Kakani et al (2005) reported an optimal temperature range between 28 and 37°C for cotton pollen germination through *in Vitro* studies.

**Photosynthesis.** In tropical and subtropical areas, it is very common that heat stress inhibits photosynthesis and in temperate zones it happens occasionally (Larcher 1995). When temperatures are higher to some extent than optimum i.e. moderate heat stress, Inhibition of photosynthesis is revocable, but under severe heat stress, damage to the photosynthetic system is irreparable (Berry & Björkman 1980). It is shown by a

comprehensive review of literature that Photo-system II (PS-II) is frequently considered the most heat sensitive element of the photosynthetic system, however a number of previous studies (some dating back to 20 years) have reported that electron transport capacity (ETC) is unpretentious at the range of temperatures that inhibit Carbon dioxide (CO<sub>2</sub>) fixation (Salvucci & Crafts-Brandner 2004).

It has long been documented that CO<sub>2</sub> fixation and activation of ribulose-1, 5-bisphosphate carboxylase/oxygenase (Rubisco), dependent on light, are reversibly repressed by moderate heat stress (Kobza and Edwards 1987). According to Crafts-Brandner & Law (2000) and Crafts-Brandner & Salvucci (2000) it has been identified that the heat-labile component of Rubisco activation is Rubisco activase. There is not much known about how activase protein expression respond to the heat stress; however it has been documented that heat stress affects Rubisco activation and activase activity (Law et al 2001). Photosynthetic process is extremely sensitive to inhibition by moderately high temperature (i.e., <10°C above the optimum temperature), and this inhibition of photosynthesis has been interrelated to the thermal variability of Rubisco's molecular chaperone, Rubisco activase (Crafts-Brandner & Salvucci 2000; Salvucci & Crafts-Brandner 2004). Law & Crafts Brandner (1999) reported that in cotton CO<sub>2</sub> absorption and Rubisco activity are more tolerant to heat stress than wheat. In cotton the expression of an N-terminally integral isoform of Rubisco activase that functions in the acclimation of photosynthesis is induced by high temperature and may be the result of transcriptional regulation (Law et al 2001). Law et al (2001) proposed the possibility of the expression of a novel activase gene in cotton leaves that is induced by heat stress. Earlier in two monocotyledonous species, wheat (Law & Crafts-Brandner 2001) and maize (Sanchez de Jiménez et al 1995) the induction of a new supposed form of activase in response to heat stress was observed.

It was reported by Reddy et al (1995b) that net photosynthesis was low in cotton at both higher and lower temperatures than optimum (i.e., 28 °C). Photosynthesis decline but dark respiration rises exponentially with increasing temperatures (Bednarz & van Iersal 2001). CO<sub>2</sub> exchange rate (CER) is inhibited by high temperature by reducing mainly the activation state of Rubisco through inhibition of Rubisco activase (Law & Crafts-Brandner 1999).

**Effect of Heat Stress on Yield and Fiber Quality of Cotton.** Wanjura et al (1969) reported that heat stress highly effect the final yield in cotton and year to year variability in cotton yield has been linked with indeterminable seasonal temperature disparity (Oosterhuis 1999). Brown et al (2003) reported environmental stresses, predominantly drought and heat stress are accountable for year to year inconsistency in cotton yield (Lewis et al 2000).

Brown et al (2003) proposed that environmental stresses, particularly water deficit, and temperature stress were mainly responsible for year-to-year variability in cotton yield (Lewis et al 2000). Bibi et al (2008) reported that high temperature during day time rather than mean temperature decline photosynthesis and carbohydrates production and Loka & Oosterhuis (2010) found that respiration will increase at high night temperatures, further decreasing available amount of carbohydrates that results in boll size reduction, low seed set and reduced number of seeds per boll and number of fibers per seed (Arevalo et al 2008). For determining final yield number of seeds is an important yield component; for example Groves (2009) reported that 80% of total cotton yield variability is dependent on number of seeds per acre. Singh et al (2007) reported a strong negative correlation between high temperature and lint yield and quality.

Fiber micronaire value increases due to high night temperatures Krieg (2002) and shorter fibers are resulted at high temperature Meredith (2005). Gipson & Joham (1968) reported that decreased night temperatures, reduce the boll development and sum total of cellulose synthesis. Temperatures more than or equal to 21.1 °C at night time gives maximum degree of fiber elongation (Gipson & Joham 1969). Early phases of fiber elongation were highly sensitive to high night temperatures as compared to later phases (Gipson & Joham 1969). In upland cotton var. Dunn it was confirmed by Smutzer & Berlin (1975) and found that a temperature of 15 °C was optimum for fiber elongation.

Time required to reach the genetic potential for fiber length, increased at temperature below 22 °C at night time (Gipson & Joham 1969). Gipson & Joham (1968) observed that when temperatures were between 15 and 21 °C at night, maximum fiber length was attained and above or below this range of temperature there was a reduction in fiber length. The highest influence of increasing temperature was an increase in fiber strength, but with reducing ginning percentage, fluctuations in fiber length and less stable micronaire (Hesketh & Low 1968).

**Screening for Heat Tolerance.** It is very expensive and strenuous to screen out the germplasm under normal conditions for stress tolerance (Blum & Ebercon 1981). Sullivan (1972) suggested cell membrane thermostability (CMT) for measuring the quantity of electrolyte outflowed after giving heat treatment, from the leaf disc immersed in deionized water. He reported a close association between CMT and high temperature tolerance in sorghum (*Sorghum bicolor* L.). CMT was used in a number of crops like wheat (Blum et al 2001), tomato and potato (Chen et al 1982), soybean (Martineau et al 1979), rice and cotton as a mean of heat tolerance (Singh et al 2007). For a specific crop Tahir & Singh (1993) reported adjustments to this method. Bibi et al (2003) and Rahman et al (2004) reported positive correlation between CMT and yield in cotton.

CMT has the ability to differentiate the genotypes that are heat tolerant and heat sensitive, within a species, and is used as a suitable technique for screening and selection of cotton for heat tolerance (Rahman et al 2004; Azhar et al 2009). Heat stress weakens the hydrogen bonds and electrostatic interactions between polar proteins and modifies the structure of the cell membrane in aqueous phase of the membrane (Rahman et al 2004). Rahman et al (2004) also found that permeability of the membrane was changed due to the injury to the membrane and this result in the loss of electrolytes. Damage to cell membrane is reflected by the electrolytes leaked and most of the researchers are agreed on this finding (McDaniel 1982) and Anderson et al (1990) reported variable responses of the leaves at different stages of life. CMT and relative cell injury (RCI %) are inversely proportional as high CMT is reflected by Low RCI % and low CMT by high RCI% (Rahman et al 2004) and he concluded that CMT can be a good indicator for identifying heat prone and heat tolerant genotypes. Indicator for the CMT used is RCI % and is determined by the formula given below (Sullivan 1972):

$$RCI \% = \left[ 1 - \left\{ \frac{1 - \left( \frac{T_1}{T_2} \right)}{1 - \left( \frac{C_1}{C_2} \right)} \right\} \right] \times 100$$

Singh et al (2007) reported that for screening cotton genotypes tolerant to heat stress efficiently, use of biochemical techniques like chlorophyll a and b contents and a:b ratio; physiological practices such as CMT, carbon isotope discrimination, leaf electrolyte leakage (LEL); eco-physiologically centered remote sensing/infrared methods such as canopy temperature depression (CTD), leaf air vapor pressure deficit (VPD), chlorophyll fluorescence can also be used for screening purpose. But according to Singh et al (2007) infra-red technique is becoming more efficient technique for screening and is used by Wanjura et al (2004); Karademir et al (2012) for screening heat tolerant varieties of cotton and corn. For evaluating cotton genotypes against heat stress tolerance there is need to develop systematic methodologies (Burke et al 2004; Kakani et al 2005).

**Genetics of Heat Tolerance.** For the development of heat tolerant cotton cultivars with success there is a need to know about the morphological, physiological and genetic basis of the heat tolerance and genetic variation for heat tolerance within the species is needed for breeding (Azhar et al 2009). In cotton it has been studied comprehensively to explore the genetic basis of abiotic stress tolerance (Saranga et al 2009). Genetic variation within the species for heat tolerance is required for the development of the heat tolerant varieties through breeding. In literature there is sufficient data on variability for high temperature tolerance in crop species like, rice (Yoshida et al 1981), tomato (Abdul-Baki & Stommel 1995), cowpea (El-kholy et al 1997), wheat (Ibrahim & Quick 2001b) and cotton (Rahman et al 2004). CMT is used as a suitable technique for screening, selection and to differentiate cotton genotypes for heat tolerance (Rahman et al 2004; Azhar et al 2009). In wheat, for CMT Ibrahim & Quick (2001b) and Blum et al (2001) reported high

broad sense heritability and there is huge genetic variability linked with CMT in wheat (Ibrahim & Quick 2001a,b). It has been discovered by the genetic analysis that additive variance and both additive and dominance variances have principal part in the expression of CMT in wheat (Ibrahim & Quick 2001b; Xu et al 1998).

Percy et al (1996) proposed digenic epistasis as the basis of the heat tolerance inheritance in cotton but later it was reported that variability for heat tolerance in cotton is mainly due to non-additive genetic effect (Azhar 2003). Singh et al (2007) reported the improvement for heat tolerance in Pima cotton in USA using many indices like visual index, phenotypic index and fruit-height response index. In Pima cotton stomatal conductance inheritance varies from a simple additive-dominance model to digenic epistatic interaction (Percy et al 1996). QTLs causing physiological variations that are linked with tolerance to stress, like osmotic adjustment (Morgan & Tan 1996), carbon isotope ratio, stomatal conductance (Ulloa et al 2000), chlorophyll content, and canopy temperature (Saranga et al 2001) have been identified. Rahman et al (2004) and Azhar et al (2009) reported that variability exists in the characters that are linked with heat tolerance within the species.

**Breeding for Heat Tolerance.** To get high photosynthetic varieties and hybrids with a balanced ratio between sources and sink it will be better to select plants on the basis of physiology and genetics and it will give maximum expression of potential yield (Nasyrov 2004). For plant breeders rate of photosynthesis might be selection criteria, especially when lines with higher photosynthetic rates are identified and are breed with the lines having appropriate apportioning of photosynthates for reproductive and vegetative growth (Pettigrew et al 1993). Lu et al (1997) reported that, to achieve high heat tolerance and yield higher photosynthetic rate, stomatal conductance and smaller leaf area are functionally significant. The main property of the stomata is its response to temperature that can be changed means of selection for heat tolerance and higher yield (Lu & Zeiger 1994). Burke (2001) reported that there may be an improved, attained thermo-tolerance through chromosomal deletions in the A or D genome of cotton, proposed on the basis of research in wheat. It has been reported that there is indirect selection pressure on photosynthetic rate, stomatal conductance, and leaf areas due to exhaustive selection for high yield and heat resistance in Pima cotton (Lu et al 1997).

Morphological traits suitable for direct selection includes, okra leaf type (Pettigrew 2004), thicker leaves (Hall 2001), earliness (Ahmed et al 1993) and pollen selection (Hall 1992). Physiological traits includes: cell membrane thermostability (Blum & Ebercon 1981; Bibi et al 2003; Rahman et al 2004), carbon isotopes discrimination difference (Lu et al 1996), chlorophyll contents and chlorophyll a:b ratio (Saranga et al 2004). It has been reported effective, breeding during the reproductive development in cowpea and cotton (Hall 1992; Rodriguez-Garay & Barrow 1988; Burke 2001); suggested that improved vegetative heat tolerance may also give enhanced heat tolerance during pollen development. Genotypes with increased evaporative cooling and lower leaf temperature facilitated by higher stomatal conductance may be selected for heat resistance.

Wells et al (1986) reported that there is 7% more leaf canopy photosynthesis in sub-okra type than normal near iso-lines and is one of the reason for enhanced yield. Similarly, okra leaf type showed greater canopy photosynthesis per unit leaf area than normal leaf type (Peng & Krieg 1991), but it has also been reported no difference in the leaf CO<sub>2</sub> exchange rate (CER) amongst super-okra leaf type and normal iso-lines (Elmore et al 1967). There is a difference in the competition for solar radiation with less mutual shading between leaf types in super-okra leaf type populations than populations of other leaf types. Kerby & Buxton (1978) reported that leaf type and total leaf area development have effects on the solar radiation interception and dry matter production. Pettigrew (2004) reported 14% more efficiency of light adapted PS-II and also 14% more transport rate of photosynthetic electrons in lines with okra leaf type than lines with normal leaf type. Mahan et al (1995) proposed that the ability of a plant to resist high temperature can be developed by changing the canopy architecture of the plant that assist to decrease heat stress with marginal effects on the consumption of water. It has also been suggested that for increasing photosynthetic efficiency and reducing

photorespiration, genetic alteration may be of more importance (Perry et al 1983). Evans (1983) suggested that breeding for increase in reproductive sink and the photosynthetic source could result in increased cotton yield. Wang et al (2003) proposed that there should be the following steps included in an inclusive breeding approach for abiotic stress tolerance:

- 1) Germplasm selection (especially wild relatives) and conventional breeding.
- 2) Exposition of the precise molecular control mechanisms in genotypes that are tolerant and sensitive.
- 3) Improvement through biotechnological tools for selection and breeding using molecular probes and markers for selection (among natural and bred populations), functional genomic analysis, and with genetic transformation.
- 4) Revision and upgrading present-day agricultural practices.

**Use of Biotechnology for the Improvement of Stress Tolerance.** In ideotype breeding genetic engineering is valuable in isogenic pair of lines that can be developed in one step and relatively in short time by the transformation of a single gene without any change in the genetic background of a cultivar (Hall 2001). Singh et al (2007) suggested that transformed plants must be assessed because method of gene transfer and placement of the inserted gene can impact its expression. DNA markers can be utilized for the selection of genetic background of the recurrent parent in a backcross procedure for developing near isogenic lines rather it will take several generations and years normally and for the development of indirect selection methods, DNA markers can also be used (Singh et al 2007). For this purpose markers that are closely linked to the trait of interest are needed (Singh et al 2007) and according to this we can save time by using biotechnological tools for plant improvement. Singh et al (2007) suggested that this indirect selection method is an influential tool and can be used where non-destructive screening of single plants in the first segregating generation is needed. But the role of crop breeders can never be substituted by molecular biological tools for improving agronomic traits, however these can help to be responsive for time extent of the traits that are sensitive to environment (Cheikh et al 2000). A combination of the conventional breeding and biotechnological tools may enable us to allow for adequate food production and to take care of the environment for better production and also to provide food security for the increasing population of the world (Singh et al 2007).

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