



RESEARCH ARTICLE

Effects of Foliar Spray on Morpho-Physiological Parameters under High Temperature Stress in Cotton (*Gossypium hirsutum* L.)

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ABSTRACT

High temperature is a critical barrier in most cotton growing areas, limiting cotton growth and development. The present study aimed to evaluate the effects of foliar spray on KC 3 cotton variety grown under ambient (32.66 °C) and high temperature (37.21 °C) stress in open-top chamber (OTC) with a temperature of 5 °C from the ambient temperature for 10d from flowering to boll development stage. Foliar spray of kaolin @ 3% and calcium carbonate @ 5% were sprayed separately to the set of pots both in ambient and elevated temperature on 70th day of flowering. Observations on morphological and physiological parameters were recorded on viz., plant height (cm plant⁻¹), leaf area (cm² plant⁻¹), relative water content (%), canopy temperature (°C), SPAD, chlorophyll fluorescence (Fv/Fm ratio). Kaolin @ 3% foliar spray significantly increased the plant height, leaf area, relative water content, chlorophyll content and reduced the canopy temperature both in high temperature and ambient temperature conditions. Among these treatments, T₂ - kaolin 3% (Ambient) followed by T₅ - Kaolin 3% (elevated temperature of 5 °C) recorded higher values as compared to calcium carbonate treatment both in ambient temperature and high temperature condition.

Keywords: Cotton; High temperature; Foliar application; Morpho-Physiological parameters

INTRODUCTION

Cotton is one of the world's most important cash crops, which is highly essential for the textile industry and also useful for oils, livestock feed. Globally, climatological extremes including high temperature were forecast to have a deleterious effect on plant growth and development cause catastrophic losses in crop productivity and quality (Yuan *et al.*, 2017). Mainly, cotton is a multipurpose crop that is highly sensitive to abiotic stresses. Among these, high temperature is the crucial one that threatens global food security. Raising one-degree Celsius (°C) in temperature level highly influences crop productivity and causes losses up to 17% in the growing season of all commercial crops (Lobell and Asner, 2003). It is predicted that the rise in temperature till 2100 is 5.8 °C and 2.6 °C up to 2050 mainly due to global warming (Sarwar *et al.*, 2019). Through the cotton crop responds various stresses at different growth stages which depends upon the stress severity, high-temperature stress causes drastic changes at the cellular level.

The high temperature affects plant growth by altering physiological and metabolic activities

and cause considerable yield variability (Hedhly *et al.*, 2009). Recent studies have shown that the reproductive phase is extremely sensitive to environmental stresses, especially high-temperatures that critically reduce the yield of commercial crops (Fraser *et al.*, 2020). The use of foliar applications of kaolin and calcium carbonate will help to alleviate the effects of high-temperature stress on plants. These foliar applications promote plant growth and development through various mechanisms (Conde *et al.*, 2016). Plants react in various morphological and physiological ways to survive under stress (Janská *et al.*, 2010). High-temperature stress has its impact, which reduces leaf water potential (Wahid and Close 2007), increases membrane lipid fluidity (Xu *et al.*, 2006), and increases the generation of reactive oxygen species (ROS) (Wahid *et al.*, 2007). High-temperature stress causes morphological changes viz., reduced water loss, stomata closing mechanism (Hasanuzzaman *et al.*, 2013)

Kaolin is an aluminosilicate (Al₄Si₄O₁₀(OH)₈) non-toxic clay mineral. Kaolin spray lowers leaf temperature by raising leaf reflectance, which

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lowers transpiration rate more than photosynthesis in plants exposed to high levels of solar radiation (Nakano and Uehara 1996). Cantore *et al.* (2009) found that foliar application of kaolin suspension reduces plant stress, which is essential for optimal plant growth, yield, and quality on tomato and potato. Calcium carbonate is a non-toxic, edible material that can help to reduce soil pollution (Wang *et al.*, 2015). Calcium carbonate can be made from renewable sources such as eggshell residues (Saeb *et al.*, 2013) and marine calcareous shells (Lu *et al.*, 2015) in addition to its conventional sources of rock and minerals. As demonstrated in mango, coffee, and Persian walnut (Gharaghani *et al.*, 2018), the application of particle films on the adaxial leaf surface provided photo safety and significantly increased the rate of net photosynthesis stomatal conductance, and transpiration, and reduced leaf temperature. In this study, we postulated whether the foliar spray of kaolin and calcium carbonate could mitigate the high-temperature stress in cotton.

MATERIAL AND METHODS

Plant materials

The experimental materials consisted of cotton variety of KC 3 an indeterminant type, the seed materials obtained from Agricultural Research Station, Kovilpatti. KC 3 cotton variety (TKH 97×KC 1).

Treatment details

The present work was conducted at the Department of Crop Physiology, Tamil Nadu Agricultural University, Coimbatore, during 2020-2021. Pot culture experiments were carried out under an open-top chamber (OTC).

Plants were grown up to flowering to boll development stage and the high-temperature stress was imposed at 60 DAS at flowering to boll development stage under pot culture experiment. The potted plants were kept under the open-top chamber (OTC) with the size of 3m×3m. Nine pots with three replications were maintained and the treatments were arranged in a completely randomized design (CRD) under ambient temperature conditions, the treatments are T₁ - control, T₂ - kaolin 3%, T₃ - calcium carbonate 5%. Another set of nine pots were kept under the OTC. Temperature in control T₄ - with elevated temperature of 5°C from ambient, T₅ - kaolin 3% and T₆ - calcium carbonate 5% under high-temperature conditions. High-temperature stress was imposed for 10 days (60 DAS). After 10 days, the foliar spray was done on 70th day, both in ambient and high-temperature conditions. After 5 days (75 DAS), the exposed plants were taken for observation and analyzed for morpho-physiological characters on plant height (cm plant⁻¹), leaf area

(cm² plant⁻¹), relative water content (%), canopy temperature (°C), SPAD value and chlorophyll fluorescence (Fv/Fm ratio).

Morpho-Physiological characters

The morpho-physiological parameters such as plant height, leaf area, canopy temperature, relative water content, SPAD value and chlorophyll fluorescence were observed at 75 DAS of flowering to boll development stage.

Plant height (cm plant⁻¹)

Plant height was measured from the ground level to the tip of the growing point after imposing treatments and expressed as cm plant⁻¹.

Leaf area (cm² plant⁻¹)

Leaf area per plant was measured using a leaf area meter (LICOR, Model LI 3000) and expressed as cm² plant⁻¹.

Canopy temperature (°C)

Canopy temperature measured at each replication in all treatments. It is denoted at degree Celsius (°C).

Relative water content (%)

Relative water content was calculated according to the formula given by Barrs and Weatherley (1962) and expressed in per cent.

$$\text{RWC (\%)} = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Turgid weight} - \text{Dry weight}} \times 100$$

Chlorophyll index

A portable chlorophyll metre (Minolta SPAD 502) was used to record the chlorophyll index given by Minolta (1989) method. The Minolta SPAD-502 measures the relative chlorophyll content as a ratio of light transmittance at 650 nm and 940 nm wavelengths.

Chlorophyll fluorescence (Fv/Fm ratio)

Chlorophyll fluorescence in dark adapted leaves was measured with a portable OS1p, a modulated Fluorometer (Model - OS1p040111 Advanced, Opti-Sciences, USA) (McArtney, 2012).

Statistical analysis

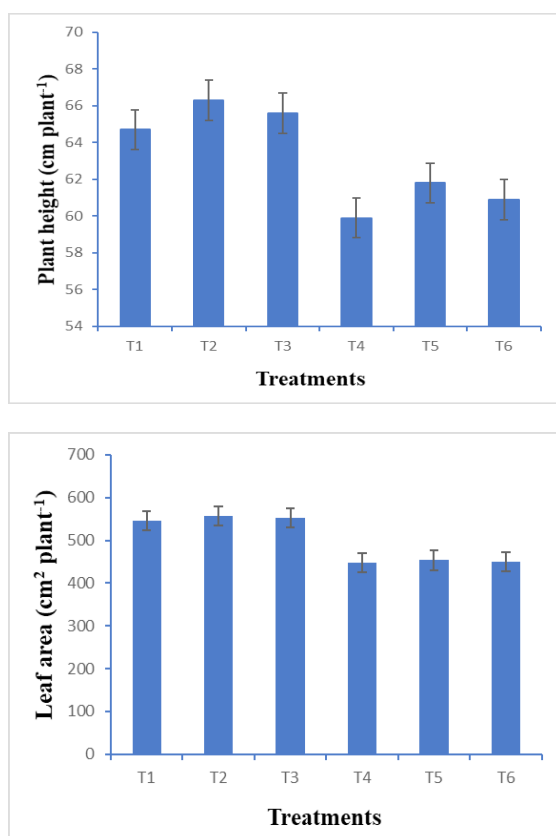
All the treatments were maintained with three replications and the data were analyzed under a completely randomized design (CRD) by using SPSS proposed by Gomez and Gomez (1984). Treatment differences were observed and critical differences were calculated at 5% level. The results were presented in the tables.

RESULTS AND DISCUSSION

Plant height (cm plant⁻¹)

The plant height was measured from flowering to boll formation (75th day) (Fig 1). In general, the plants were grown under ambient conditions (64.7 cm) recorded higher value than stressed plants due to high temperature (59.9 cm) cause the reduction in plant growth and development. The same trend was observed by foliar spray of kaolin @ 3% in ambient and high temperature recorded higher plant height (66.3 cm, 61.8 cm) than calcium carbonate @ 5% (65.6 cm, 60.9 cm) respectively. According to Khalil (2006), the results were showed in plant height and dry weight of *Physalis peruviana* L. seedlings were increased by foliar spray of kaolin.

Fig 1. Effects of high-temperature stress and foliar spray on plant height (cm plant⁻¹) and leaf area (cm² plant⁻¹)



Leaf area (cm² plant⁻¹)

Leaf area is the important parameter that determines the photosynthetic rate indicate the growth (Fig 1). The plants were grown under ambient condition and recorded the maximum leaf area (545.5 cm² plant⁻¹) compared to high temperature exposed plants (447.7 cm² plant⁻¹). The foliar spray of kaolin @ 3% sprayed both in ambient and high temperature recorded higher leaf area (556.3 cm² plant⁻¹, 453.5 cm² plant⁻¹) as compared calcium carbonate @ 5% (552.6 cm² plant⁻¹, 450.1 cm²

plant⁻¹) respectively. The present findings were consistent with observation of Saour, (2005), Bedrech and Farag (2015), ELSaid, (2015), and Omran (2013), who found that kaolin followed by calcium carbonate treatments increased the leaf area significantly when compared to untreated trees.

Canopy temperature (°C)

The canopy temperature is the most critical parameter which indicates the buffering body temperature of plants. The plants under high temperature recorded, the higher canopy temperature (37.1°C) than the ambient temperature (33.2°C). The kaolin @ 3% sprayed both in ambient and high temperature recorded lowest canopy temperature (29.9°C, 33.4°C) as compared to calcium carbonate @ 5% (30.8°C, 34.2°C) respectively. Spraying of kaolin on the leaves of temperate deciduous fruit trees decreased the canopy temperatures without affecting CO₂ assimilation (Glenn *et al.*, 1999, Tworowski *et al.*, 2002). The temperature drop was likely increased by the reflection of incident short and long-wave radiation from the white-colored kaolin-sprayed leaves (Glenn *et al.*, 2009). Glenn *et al.* (1999) found a 3 to 4 folds increase in spectral radiance in the visible spectrum reflected from tree canopies sprayed with kaolin at 30g.m⁻² leaf surface, reduced the leaf temperature up to 3 °C.

Table 1. Effects of high-temperature stress and foliar spray on SPAD value and chlorophyll fluorescence (Fv/Fm ratio).

Treatments	SPAD	Chlorophyll fluorescence (Fv/Fm)
T ₁ Unstressed - Control (Ambient)	37.3	0.69
T ₂ Kaolin 3% (Ambient)	41.0	0.74
T ₃ Calcium carbonate 5% (Ambient)	40.4	0.70
T ₄ Stressed – Control (+5 °C)	34.6	0.64
T ₅ Kaolin 3% (+5 °C)	37.1	0.66
T ₆ Calcium carbonate 5% (+5 °C)	36.4	0.65
Mean	37.6	0.68
SED	0.85	0.01
CD (P=0.05)	1.89	0.03

Relative Water Content (%)

Relative water content measurement during flowering to boll development stage is shown in Fig 2. The results of RWC indicated that the relative water content of the stressed leaves from high temperature recorded lower values (80.4%) as compared to ambient conditions (85.7%). Pawar and Patil showed similar results. (1982) in *Phaseolus aureus* and *Vigna catieng*, Singh and Sahay (1989) in cotton. Mahalakshmi *et al.* (1999) reported that

foliar application of kaolin at 5% in chilli pepper for two seasons induced higher RWC (80.8% and 83.0%, first and second season, respectively) compared to control.

Chlorophyll Index (SPAD value)

SPAD values for assessing chlorophyll index were carried out from the plants exposed under high-temperature conditions. The results indicated that the chlorophyll intensity was significantly reduced when the plants exposed to high temperature might be due to the desiccation of chlorophyll pigments due to high temperature.

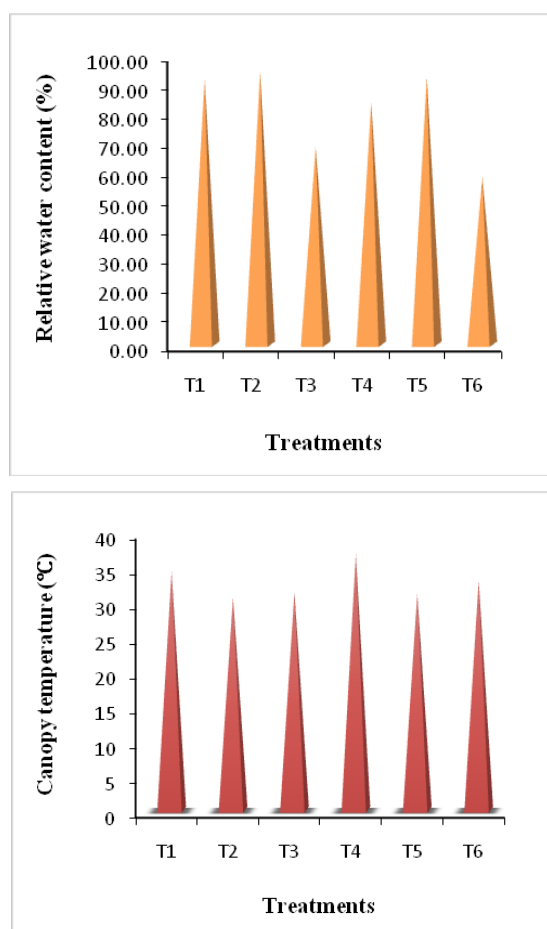


Fig 2. Effects of high-temperature stress and foliar spray on relative water content (%) and canopy temperature (°C)

Lombardini *et al.* (2005) used kaolin to improve the chlorophyll index and the rise in chlorophyll content observed due to lower light reflectance. Similar results were obtained with the conformity of the findings of Dinis *et al.*, 2016.

Chlorophyll fluorescence (Fv/Fm ratio)

Chlorophyll fluorescence parameter is a measure to utilize to know the plant responses under stress conditions. Photosynthetic events can be monitored using chlorophyll fluorescence, which is a non-invasive process. It is a good indicator of

photochemistry and electron transport rate, and linked to photosynthetic performance. Table 1 shows that the plants were grown under ambient conditions had recorded higher chlorophyll fluorescence (0.69) as compared to high temperature exposed plants (0.64). The kaolin @ 3% sprayed both in ambient and high temperature recorded the maximum chlorophyll fluorescence (0.74, 0.66) as compared calcium carbonate @ 5% (0.70, 0.65). Jifon *et al.*, 2003 reported that Fv/Fm values were increased in leaves at 3% to 6% of foliar spray of kaolin than control leaves because lower photoinhibition might improve the Fv/Fm ratio.

CONCLUSION

The present study concluded the effects of foliar spray under high temperature stress in cotton a variety of KC 3 during flowering to boll development stage. It was evaluated that the morpho-physiological characters viz., plant height, leaf area, relative water content, canopy temperature, SPAD and chlorophyll fluorescence showed that kaolin @ 3% treated plants had better performance when compared to calcium carbonate @ 5% under both in ambient and high temperature condition. The use of foliar spray of antitranspirants helps to mitigate the effects of high temperature stress through reduction of leaf temperature, transpiration rate and increases the water potential on plants during flowering to boll development stage.

REFERENCES

- Almeida Barros da Silva, Juliana ; Perez, Laura and Fraser, Paul. 2020. A transcriptomic, metabolomic and cellular approach to the physiological adaptation of tomato fruit to high temperature. *In: Plant, Cell and Environment*.
- Barrs, H.D. and P.E. Weatherley. 1962. A re-examination of relative turgidity for estimating water deficits in leaves. *Aus.J.Biol.Sci.*,**15**: 413-428.
- Bedrech, S.A. and S. G. Farag. 2015. Usage of some sunscreens to protect the Thompson Seedless and Crimson Seedless grapevines growing in hot climates from sunburn. *Nature and Science*,**13**(12):35- 41.
- Cantore, V., Pace, B. and R. Albrizio. 2009. Kaolin-based particle film technology affects tomato physiology, yield and quality. *Environmental and Experimental Botany*,**66**: 279-288.
- Conde, A., Pimentel, D., Neves, A., Dinis, L. T., Bernardo, S., Correia, C. M., Gerós, H. and J. Moutinho-Pereira. 2016 Kaolin Foliar Application Has a Stimulatory Effect on Phenylpropanoid and Flavonoid Pathways in Grape Berries. *Front. Plant Sci.*,**7**:1150. doi: 10.3389/fpls.2016.01150
- Dinis, L.T., Bernardo S. and A. Conde. 2016b. Kaolin exogenous application boosts antioxidant capacity and phenolic content in berries and leaves of grapevine under summer stress. *J. Plant Physiol.*, **191**:45–53.

- El-Said. 2015. Effect of irrigation intervals and some antitranspirants on growth, yield and fruit quality of Eggplant, *J. Plant Production*, Mansoura Univ. Egypt., **6** (12): 2079- 2091.
- Gharaghani, A., Mohammadi A. and K. Vahdati. 2018. Kaolin particle film alleviates adverse effects of light and heat stresses and improves nut and kernel quality in Persian walnut. *Sci. Hortic. (Amsterdam)*, **239**:35–40. <https://doi.org/10.1016/j.scienta.2018.05.024>.
- Gomez, K.A. and A.A. Gomez. 1984. Statistical procedures for agricultural research. (2nd Ed.) John Wiley and sons, New York, USA.p. 680
- Glenn, D. M., Puterka, G. J., Vanderzwet, T., Byers, R, E. and C. Feldhake. 1999 Hydrophobic particle films: a new paradigm for suppression of arthropod pests and plant diseases. *Journal of Economic Entomology*, **92**:759-771
- Glenn, D. M. 2009. Particle film mechanisms of action that reduce the effect of environmental stress in 'Empire' apple. *J Am Soc Hortic Sci.*, **134**:314–21.
- Hasanuzzaman, M., Hossain, M. A. and M. Fujita. 2010. Physiological and biochemical mechanisms of nitric oxide induced abiotic stress tolerance in plants. *Am J Plant Physiol.*, **5**:295–324.
- Hedhly, A., Hormaza, J.I., and M. Herrero. 2009. Global warming and sexual plant reproduction. *Trends Plant Sci.*, **14**:30-36.
- Janská, A., Mar ík, P., Zelenková, S. and J. Ovesná. 2010. Cold stress and acclimation-what is important for metabolic adjustment? *Plant Biol.*, **12**:395–405.
- Jifon, J.L. and J.P. Syvertsen. 2003. Foliar sprays of kaolin clay can increase photosynthesis and water use efficiency of citrus leaves. *J. Amer. Soc. Hort. Sci.*, **128**: 107-112.
- Khalil, S.E. 2006. Physiological study on sesame plants grown under saline water irrigation condition. PhD thesis, Cairo University 229
- Lombardini, L., Glenn, D. M. and M. K. Harris. 2005. Effects of particle film application on Leaf gas exchange, water relations, nut yield, and insect populations in mature pecan trees. *Hort Science*, **40**:1376–1380
- Lobell, D.B. and G. P. Asner. 2003. Climate and management contributions to recent trends in US agricultural yields. *Science*, **299**:1032
- Lu, J., Lu, Z., Li, X., Xu, H. and X. Li. 2015. Recycling of shell wastes into nanosized calcium carbonate powders with different phase compositions. *J. Clean. Prod.*, **92**: 223-229.
- Mahalakshmi, B. K, Rao, D. V. S., Rao, G. R. and P. J. Reddy 1999. Management of drought with anti-transpirants in chilli (*Capsicum annum* L.). *South Indian Horticulture*, **47**: 86-88.
- Maxwell, K. and G.M. Johnson. 2000. Chlorophyll fluorescence – A practical guide. *J. Exp. Bot.*, **51**: 659-668.
- Monje, O.A. and Bughree. 1992. Inherent limitation of non-destructive chlorophyll meters. A comparison of two types of meters. *Hort. Sci.*, **27**:71-89.
- Minolta, C. 1989. Manual for Chlorophyll meter SPAD-502. Minolta Cameraco., Osaka, Japan.
- Nakano, A. and Uehara, Y. 1996. The effects of kaolin clay on cuticle transpiration in tomato. *Acta Hortic.*, **440**: 233-238.
- Omran, M.A. 2013. Maximizing olives productivity under insufficient chilling requirements conditions. Ph.D. Thesis, *Fac. Of Agric.*, Ain Shams Univ., Egypt
- Pawar, A. B. and B. B. Patil 1982. Effect of reflectant (Kaoline) on yield, relative turgidity of leaf and albedo of the canopy of summer mung (*Phaseolus aureus*) and cowpea (*Vigna catiang*). *Journal of the Maharashtra Agricultural University*, **7**: 12-14.
- Saeb, M.R., Ramezani-Dakheel, H., Khonakdar, H.A., Heinrich, G., and U. Wagenknecht. 2013. A comparative study on curing characteristics and thermomechanical properties of elastomeric nanocomposites: the effects of eggshell and calcium carbonate nanofillers. *J. Appl. Polym. Sci.*, **127**:4241–4250. <https://doi.org/10.1002/app.38022>.
- Sarwar, M., Saleem, M.F. and N. Ullah. Role of mineral nutrition in alleviation of heat stress in cotton plants grown in glasshouse and field conditions. *Sci Rep* **9**:13022 (2019).
- Saour, G. 2005. Morphological assessment of olive seedlings treated with kaolin-based particle film and biostimulant. *Advances in Horticultural Science*, **19** (4):193-197.
- Singh D. and R. K. Sahay 1989. Effect of pix and kaolin on growth and yield of upland cotton (*Gossypium hirsutum*). *Indian Journal of Agricultural Sciences*, **54**:247-250.
- Twoorkoski, T.J., Glenn, D.M. and G.J. Puterka. 2002. Response of bean to applications of hydrophobic mineral particles. *Can. J. Plant Sci.*, **82**:217-219.
- Wahid, A. and T. J. Close. 2007. Expression of dehydrins under heat stress and their relationship with water relations of sugarcane leaves. *Biol Plantarum*, **51**:104–109.
- Wahid A. 2007. Physiological implications of metabolites biosynthesis in net assimilation and heat stress tolerance of sugarcane (*Saccharum officinarum*) sprouts. *J Plant Res.*, **120**:219– 228.
- Wang, Y., Liu, A., Ye, R., Wang, W. and X. Li. 2015. Transglutaminase-induced crosslinking of gelatin-calcium carbonate composite films. *Food Chem.*, **166**: 414–422. <https://doi.org/10.1016/j.foodchem.2014.06.062>.
- Xu, S., Li, J., Zhang, X., Wei, H. and J. Cui. 2006. Effects of heat acclimation pretreatment on changes of membrane lipid peroxidation, antioxidant metabolites and ultrastructure of chloroplasts in two cool-season turfgrass species under heat stress. *Environ Exp Bot.*, **56**:274–285.
- Yuan, M., Liu, C., Liu, W. S., Guo, M. N., Morel, J. L., Huot, H., Yu, H. J., Tang, Y. T. and R. L. Qiu 2017. Accumulation and fractionation of rare earth elements (REEs) in the naturally grown *Phytolacca americana* L. in southern China. *Int. J. Phytoremediat.*