

## Classification and Evaluation of Epicuticular wax in Wheat (*Triticum aestivum* L.) for Heat stress Screening

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**ABSTRACT:** Global temperatures have increased since the dawn of the century and are predicted to continue rising under climate change. This increases in temperature can cause heat stress: a severe threat to wheat production globally. In this research we describe a particular morphological response of wheat to elevated temperature. This experimental research was carried out to determine the response of different wheat genotypes (*Triticum aestivum* L.) on epicuticular wax under timely and late sown condition. The research was conducted at Seed Breeding Farm, AICRP on Wheat & Barley, Department of Plant Breeding & Genetics, JNKVV (M.P.) during Rabi season in year 2021-22, in randomized complete block design with two replications. Result of morphological traits revealed that tolerant genotypes had higher epicuticular wax under late sown condition. Genotypes JW 3336, JW 3288, DBW 71, RAJ 3765 and WH 730 show maximum glaucousness and increase tendency of epicuticular wax synthesis from normal to late sown environmental condition. This study explains the inter-relationship between epicuticular wax improving wheat adaptability to high temperature stress.

**Keywords:** *Triticum aestivum* L., Epicuticular wax, Heat stress.

### INTRODUCTION

Wheat, the second most important staple food consumed by almost 35% of the world population and providing 20% of the total food calories. It occupies nearly about 32% of the total acreage under cereals crops in the world. Wheat belongs to the genus *Triticum* of the family *poaceae* and it is believed to be originated from South West Asia (Bingham and Lupton, 1987). There are three well known species of wheat namely, *Triticum aestivum* (Bread), *Triticum durum* (Macaroni) and *Triticum dicoccum* (Emmer) grown for commercial purpose in India. Of these species, *T. aestivum* is the most dominant species computing about 90-95% of whole wheat area of the country and is grown in almost all the wheat growing regions of the states.

Global climate models predict that rise in mean ambient temperature between 1.8 to 5.8°C by the end of this century (Masson *et al.*, 2021). Developing new crop varieties adaptable to future climate, we need to know how crops respond to elevated temperatures and how heat tolerance can be improved (Halford, 2009). One of the most important abiotic agent limiting plant growth and productivity worldwide in agriculture is heat stress. It is recognizable that two thirds of the potential main crops yields were drastically affected by undesirable environmental factors. Heat response variables, such as membrane thermostability, epicuticular wax increased

osmotic adjustment production, and so on cause to neutralize or minimize heat stress effects.

Over the period of time plants have evolved to survive in conditions which are hardly ideal for maintenance of normal physiology and may be at the limit for existence. In response, plants can avoid, adapt and overcome the stress by means of different biochemical and physiological mechanisms. Glaucousness is the whitish or grayish appearance of leaf blades, glumes, sheaths, and stems of plants. This appearance is because of the epicuticular wax exudates secreted by plant organ. As the primary interface between plant and its environment the cuticle plays a vital role in maintaining the plant's integrity within an inherently hostile environment. Outermost surface is covered with a hydrophobic layer of a long chain aliphatic molecules, collectively referred to as cuticular wax.

Leaf epicuticular wax is frequently define as a bluish-green covering on the adaxial and abaxial surfaces of leaf and appears during the early reproductive stages. It is associated with increased stress tolerance in rice (*Oryza sativa* L.) (Haque *et al.*, 1992), maize (*Zea mays* L.) (Meeks *et al.*, 2012), barley (*Hordeum vulgare* L.) (Febrero *et al.*, 1998), wheat (Bennett *et al.*, 2012), sorghum (*Sorghum bicolor* L.). In addition to the leaf surface, epicuticular wax also present on the peduncle (culm), leaf sheath, stem sheath, glumes, and other areas of the plant, performing as a hydrophobic barrier

between the surrounding environment and leaf epicuticle (Bird *et al.*, 2007).

Availability of sufficient genetic variability is very crucial for any crop improvement programme. Amount of sufficient variability in experimental material is desirable characteristics for any successful breeding programme. All varieties usually do not maintain the same kind of relationship under variable temperatures. Screening genotypes and their use in research experiment for heat tolerance will enable us to develop thermos-insensitive or temperature insensitive varieties which will ultimately boost up the production of wheat in temperature prone countries. The present research describes morphological trait particularly, epicuticular

wax, responses and to explore how these relation can be exploited to improve heat tolerance in wheat crop.

## MATERIAL AND METHODS

The present investigation was carried out at Breeders Seed Production Unit, Department of Plant Breeding and Genetics, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur, Madhya Pradesh (India) in the rabi season 2021-22. Fifteen wheat genotypes were grown in a heat stress and optimal growth conditions in 2021-2022. Each genotypes was replicated 2 times for both control and high temperatures conditions in RCBD.

**Table 1: Experimental material.**

Sr. No.	Genotypes	S. No.	Genotypes	S. No.	Genotypes
1.	GW366	2.	GW 273	3.	GW322
4.	JW1201	5.	JW1202	6.	JW1203
7.	JW 3336	8.	JW 3382	9.	JW 3211
10.	DBW 14	11.	DBW 71	12.	MP 4010
13.	JW 3288	14.	RAJ 3765	15.	WH 730

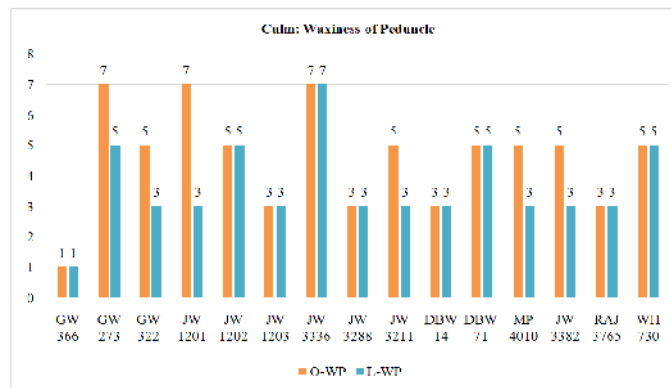
**Glaucousness scores.** The data was taken in the form of descriptor codes with slight modification assigned by PPV and FRA & UPOV for the crop wheat. Glaucousness was scored visually at vegetative and anthesis stage. Visual rating of glaucousness was recorded. Genotypes were classified as (1=Low, 3=Medium, 5=Moderate, 7=High and 9=Very high glaucous) based on the visual rating of glaucousness appearance of culm, leaf sheath, ear and flag leaf.

## RESULT

Significant differences were observed for epicuticular wax between the cultivars. The interaction between cultivar and environment for the morphological traits namely Culm: waxiness of peduncle, Flag leaf:

waxiness of leaf sheath, Ear: waxiness and Flag leaf: waxiness of leaf blade were observed and showed in Fig. 1 to 4.

**Culm: waxiness of peduncle.** JW 3336 showed high glaucousness under both timely and late sown condition. DBW 14, RAJ 3765, and DBW 71, WH 730 showed medium and moderate glaucousness with equal magnitude in both timely and late sown condition. Sharpest decline was observed in JW 1201 for late sown condition, while GW 273 declines from high to moderate and genotypes GW 322, MP 4010 and JW 3382 declines from moderate to medium glaucousness in normal to late sown conditions.



**Fig. 1.** O-WP= wax peduncle at optimum, L-WP= wax peduncle at late sown.

**Flag leaf: waxiness of sheath.** Genotypes GW 273, DBW 14, DBW 71, RAJ 3765 and WH 730 showed increase in epicuticular wax for leaf sheath in normal to late sown condition. DBW 71 and WH 730 falls under very high glaucous category and GW 273 and Raj 3765 falls under high glaucous category in late sown

conditions. JW 3336 maintains a constant glaucousness in both the environmental conditions. Decline was observed from moderate to medium level for GW 322 and JW 1201 from normal to late sown conditions, while deepest decline was observed for genotype JW 3211.

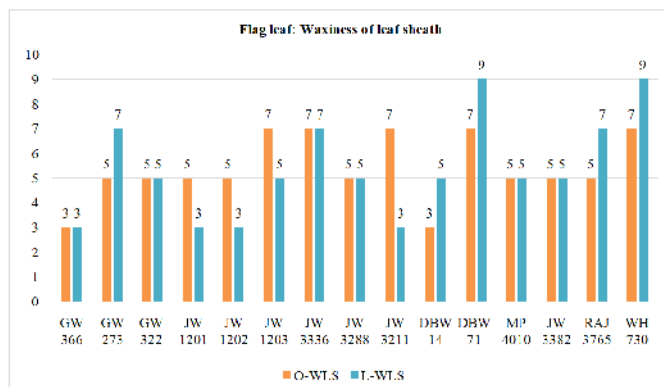


Fig. 2. O-WLS= waxiness of leaf sheath at optimum and L-WLS= waxiness of leaf sheath at late sown.

**Ear: waxiness.** JW 3288, DBW 71 and WH 730 showed increase in glaucousness in late sown conditions as compared to other genotypes. Sharpest decline was observed in RAJ 3765 for late sown conditions, while low glaucousness was observed for GW 366. DBW 71 showed very high glaucousness and JW 1203, JW 3288, MP 4010 and WH 730 showed moderate level of glaucousness.

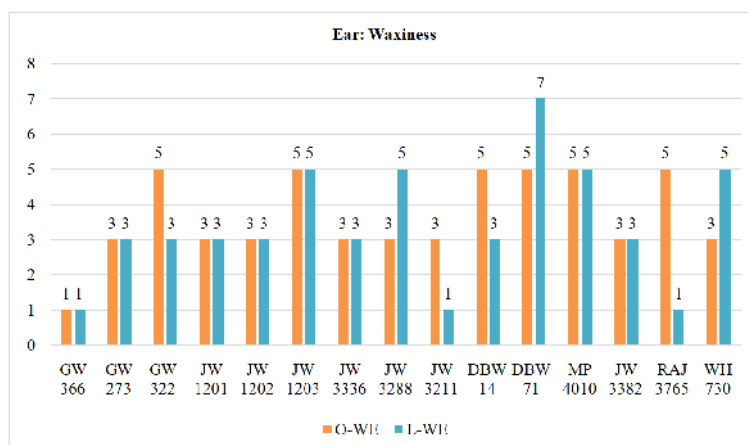


Fig. 3. O-WE= ear waxiness at optimum and L-WE= ear waxiness at late sown.

**Flag leaf: waxiness of blade.** While talking about another important morphological traits, flag leaf: waxiness of blade, genotypes DBW 71, RAJ 3765 and WH 730 increases and showed very high glaucousness in late sown condition. JW 3336, DBW 14 and MP 4010 increases from normal to late sown condition and showed high glaucousness. GW 322 and JW 1201 declines to medium glaucousness from normal to late sown condition.

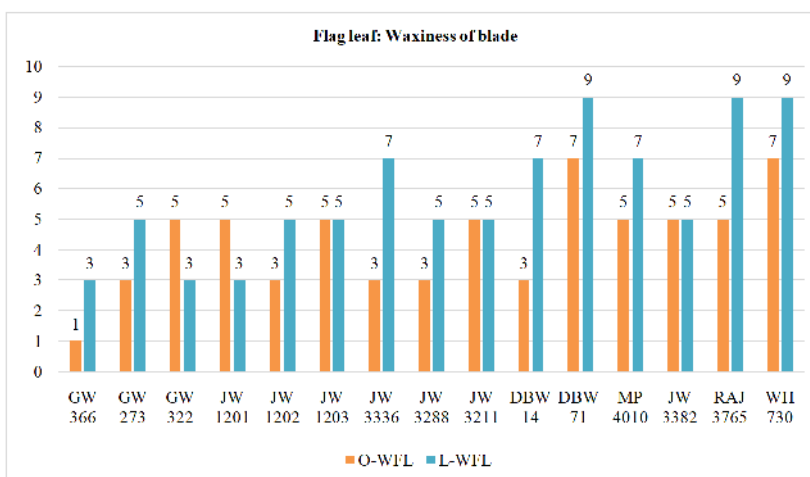


Fig. 4. O-WFL= waxiness in flag leaf blade at optimum& L-WFL= waxiness in flag leaf blade at late sown.

## DISCUSSION

Epicuticular wax is not only the product of genetics of plant body but is also greatly influenced by the ambient environmental conditions and its composition and concentration differs drastically across and within the species (Elham *et al.*, 2012; Kim *et al.*, 2007).

Result obtained in this study also showed that epicuticular wax content declines in genotypes GW 366, GW 322, JW 1201, JW 1202 under heat stress environments. Similar result were supported by Huggins *et al.* (2018) where some wheat genotypes produced lower amount of wax content in both the environments.

Wheat genotypes *viz.*, JW 3336, WH 730, RAJ 3765, and DBW 71 performed better in terms of epicuticular wax synthesis in culm, flag leaf sheath, ear and flag leaf in late sown condition as compared to other genotypes. This result inference that heat stress increase the amount of epicuticular deposition on the leaf surface. Clarke and Richards 1988 also showed higher amount of epicuticular wax in wheat in response to heat stress condition. In addition to this epicuticular wax can reduce the amount of irradiation entering the leaf, thus reducing thermal load by reflecting back the extra light and act as a barrier in hot and dry environments (Sanchez *et al.*, 2001). Huggins *et al.* (2018) found similar pattern of higher epicuticular wax synthesis in high temperature stress as compared to control treatment. As these genotypes are mentioned as heat tolerant genotypes and epicuticular wax is interconnected with various physiological traits influencing plant water use & for this reason it may decrease leaf CT and increases heat tolerance thereby resulting in an improved yield stability (Pinto *et al.*, 2010). Samuels *et al.* (2008); Bi *et al.* (2017) also suggested that glaucousness has been associated with several traits and physiological processes, mainly related to increased drought and heat tolerance and thereby higher yield under heat and dry conditions. So accumulation of epicuticular wax in tolerant genotypes can be screened as an indirect selection criterion for heat stress condition. Furthermore it has been also mentioned (Elham *et al.*, 2012) that under both water and heat stress conditions wheat genotypes produce 20 percent more amount of epicuticular wax. This study reasonably suggest that there is interaction between epicuticular wax layer and the corresponding environment similar to findings reported in peanuts (Samdur *et al.*, 2003).



Fig. 5. Variation in waxiness of flag leaf.



Fig. 6. Variation in waxiness of leaf sheath.



Fig. 7. Highly waxed genotype.



Fig. 8. A non-waxed genotype.

## CONCLUSION AND FUTURE SCOPE

Wheat breeding strategies to develop stress-tolerant varieties are based on the development of a population with ensuing selection within the population. Past decade has seen new evolution in methods specific for yield selection, incorporating morphology into the yield selection criteria. Epicuticular wax offers safeguards against both abiotic & biotic stresses to plants. EW tempers the leaf microenvironment to heat stress, indicated by the positive beneficial relationship between leaf temperature depression and epicuticular wax. This reduction is probably result of increase

reflectance at the leaf surface. The results specify that heat tolerant cultivars can productively use epicuticular wax as one of the physiological components. This unique morphological trait could makes the identification easy between heat susceptible and resistant genotypes.

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**Conflict of Interest.** None.

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