



# Effect of drought stress on epicuticular wax load in peanut genotypes

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## ABSTRACT

The changes in epicuticular wax load (EWL) and the leaf temperature in two peanut genotypes such as JL-24 (drought susceptible) and K-9 (drought tolerant) was studied under drought stress and its recovery. The drought stress was induced at different soil moisture level (100%, 60%, 40% and 20% FC) for 7 days and subsequent recovery for 7 days. In both JL-24 and K-9 genotypes, the EWL was found to be increase with the increase in the intensity of drought stress. However, the increase in EWL in K-9 genotype was found to be significantly higher than JL-24 genotype. Moreover, the leaf temperature was also found to be increased with the increase in the intensity of drought stress and the degree of increase in leaf temperature was more pronounced in JL-24 compared to K-9 genotype. Further, after the recovery of drought stress, the EWL and leaf temperature decreased consistently in both the genotypes. The results are discussed in terms of the differences in EWL and temperature of leaf in two genotypes studied under drought stress.

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## 1. INTRODUCTION

Peanut (*Arachis hypogaea* L.), commonly known as groundnut is source of the fourth most important edible oil and the third most important vegetable protein in the world [1]. However, over 97.6% of world peanut growing area and about 95.5% of the total production is concentrated in developing countries, predominantly in Asia and Africa, where the crop is grown mostly under rain-fed conditions [2]. Low rainfall and prolonged dry spells during growth and development stages of crop are the main reasons for low yields in these areas. To cope with the adverse effects of drought, plants have evolved strategies that enable them to survive under stress conditions [3]. Under drought stress, prevention of water loss through aerial parts of the plant is considered as one of the most important mechanism to avoid drought. As a consequence of water-deficit condition, the stomata tend to close to minimize the loss through transpiration. It has been known that the epicuticular wax minimizes cuticular transpiration [4]. Under water-deficit condition, the leaf temperature have been recognized as an indicator of overall plant-water status [5] and used to asses yield under drought stress [6,7]. Therefore, the present work aims to study the effect of drought stress on epicuticular wax load and leaf temperature in peanut genotypes.

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## 2. MATERIALS AND METHODS

### 2.1 Plant material and stress imposition

Seeds of peanut genotypes, namely JL-24 (drought susceptible) and K-9 (drought tolerant) were collected from Acharya N. G. Ranga Agricultural University (ANGRAU), Agriculture Research Station (ARS), Kadiri, Andhra Pradesh, India. The seeds were sown in plastic pots and plants were maintained gravimetrically [8]. All the pots were maintained at 100% FC (field capacity) and the water was supplied by keeping check on the water loss by ET (Evapo-Transpiration) rate twice a day for 30 days. At the end of 30<sup>th</sup> day, the pots were allowed to reach 60%, 40% and 20% FC by controlled irrigation with an average decrease of 15% FC/day for the next five days. At the end of 35<sup>th</sup> day, drought stress were imposed by maintaining pots at different soil moisture content (100% FC, control; 60% FC, mild stress; 40% FC, moderate stress and 20% FC, severe stress) for 7 days. After 7 days of drought imposition, the leaf temperature was recorded and the leaves were harvested for the estimation of epicuticular wax load. Following induction of stress, all the pots were supplemented with water upto 100% FC for the recovery phase for 7 days. At the end of recovery phase, leaf temperature was recorded and the leaves were harvested for the estimation of epicuticular wax load. Throughout the experiment, all the pots were maintained at rain-free condition.

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## 2.2 Measurement of epicuticular wax load (EWL)

Leaf discs were excised from fresh leaf samples with cork borer. For extraction of wax, five (05) discs per sample (area of a disc, 1.54 cm<sup>2</sup>) were dipped for 15 sec in the tube containing 15 ml of chloroform and taken out. The tubes were kept in hot water bath maintained at 60°C for 8 h until the evaporation of chloroform and the wax remained at the bottom in the sticky form. The wax content was estimated by the method as described by Ebercorn [9] with slight modification. A 5 ml of wax reagent was added to each tube and kept at 60°C in water bath for 30 min. After cooling, 12 ml of deionized water was added to each tube and the solution was filtered through a filter paper. The absorbance was measured at 590 nm with spectrophotometer (Shimadzu, UV-1800). The wax content was calculated using standard carnauba plant wax and the EWL is expressed as wax load per unit area of leaf (mg/dm<sup>2</sup>).

## 2.3 Measurement of leaf temperature

The leaf temperature was measured using infrared thermometer (Raytec, Mini Temp.) between 12 pm to 1 pm. The IR gun was placed about an inch from the adaxial leaf surface and the temperature (in °C) was recorded.

## 3. Statistics analysis

The experiments were performed in triplicate (n=3). The results are presented as Mean with Standard Deviation (SD). Significance between the results among different treatments was analyzed by multivariate ANOVA followed by Duncan's Multiple Range Test (DMRT) using IBM SPSS Statistics V20.0 software and represented as lower case letters placed at superscript of Mean ± SD values along every row in the table.

## 4. RESULTS

The effect of drought stress on epicuticular wax load (EWL) and leaf temperature in peanut genotypes was studied and the results are presented as table and figure.

### 4.1 Changes in the epicuticular wax load

The EWL in JL-24 genotype under drought stress ranged from a minimum of 0.903 mg/dm<sup>2</sup> at 100% FC to a maximum of 1.327 mg/dm<sup>2</sup> at 20% FC, whereas, in K-9 genotype, it ranged from a minimum of 1.140 at 100% FC to a maximum of 2.147 mg/dm<sup>2</sup> at 20% FC. Under recovery of drought stress, the EWL was found to 0.873 mg/dm<sup>2</sup> and 1.160 mg/dm<sup>2</sup> at 100% FC in JL-24 and K-9 genotypes respectively, whereas, it reached to 1.157 mg/dm<sup>2</sup> and 1.347 mg/dm<sup>2</sup> at 20% FC in JL-24 and K-9 genotypes (Table 1, Fig. 2).

### 4.2 Changes in the leaf temperature

In JL-24 genotype, the leaf temperature under drought ranged from a minimum of 30.13°C at 100% FC to a maximum of 39.47°C at 20% FC. Whereas, in K-9 genotype, it ranged from a minimum of 29.70°C at 100% FC to a maximum of 34.50°C at 20% FC. Under recovery of drought stress, the leaf temperature

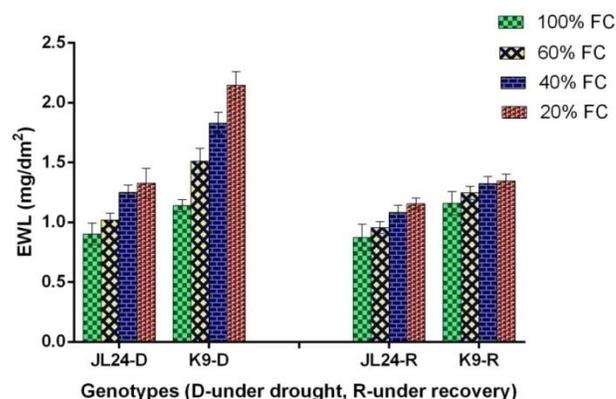
reached to 32.20°C and 31.00°C in JL-24 and K-9 genotypes at 20% FC (Table, Fig. 2).

**Table 1:** Changes in the epicuticular wax load (EWL) and leaf temperature under drought stress and its recovery.

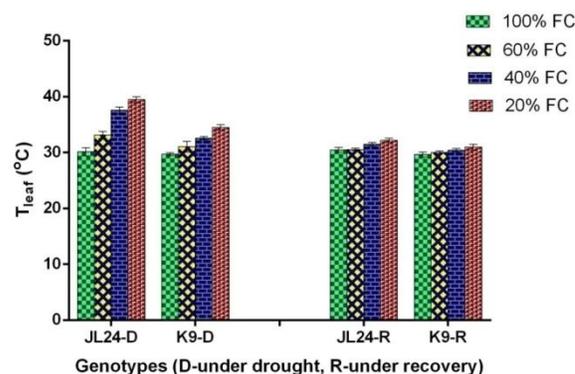
Sl. No.	Genotypes	Treatment FC (in %)	EWL	T <sub>leaf</sub>	EWL	T <sub>leaf</sub>
			(Under drought) (mg/dm <sup>2</sup> )	(Under drought) (°C)	(Under recovery) (mg/dm <sup>2</sup> )	(Under recovery) (°C)
			Mean SD	Mean SD	Mean SD	Mean SD
1	JL-24	100	0.903 ± 0.090 <sup>a</sup>	30.13 ± 0.74 <sup>ab</sup>	0.873 ± 0.112 <sup>a</sup>	30.47 ± 0.45 <sup>bc</sup>
2		60	1.020 ± 0.056 <sup>ab</sup>	33.13 ± 0.65 <sup>c</sup>	0.957 ± 0.050 <sup>a</sup>	30.57 ± 0.21 <sup>bc</sup>
3		40	1.250 ± 0.060 <sup>cd</sup>	37.57 ± 0.59 <sup>e</sup>	1.083 ± 0.060 <sup>b</sup>	31.47 ± 0.35 <sup>d</sup>
4		20	1.327 ± 0.125 <sup>d</sup>	39.47 ± 0.51 <sup>f</sup>	1.157 ± 0.045 <sup>bc</sup>	32.20 ± 0.36 <sup>e</sup>
5	K-9	100	1.140 ± 0.050 <sup>bc</sup>	29.70 ± 0.26 <sup>a</sup>	1.160 ± 0.096 <sup>bc</sup>	29.67 ± 0.45 <sup>a</sup>
6		60	1.512 ± 0.107 <sup>c</sup>	31.03 ± 0.96 <sup>b</sup>	1.247 ± 0.055 <sup>cd</sup>	30.03 ± 0.25 <sup>ab</sup>
7		40	1.830 ± 0.092 <sup>f</sup>	32.60 ± 0.26 <sup>c</sup>	1.323 ± 0.061 <sup>d</sup>	30.43 ± 0.32 <sup>bc</sup>
8		20	2.147 ± 0.112 <sup>f</sup>	34.50 ± 0.46 <sup>d</sup>	1.347 ± 0.057 <sup>d</sup>	31.00 ± 0.50 <sup>cd</sup>

Values not sharing same alphabets are significantly different at p ≥ 0.05.

FC – Field Capacity (in %), EWL – Epicuticular Wax Load, T<sub>leaf</sub> – Leaf temperature.



**Fig. 1:** Changes in the epicuticular wax load (EWL) in the leaf of peanut genotypes under drought stress and its recovery.



**Fig. 2:** Changes in the leaf temperature (°C) of peanut genotypes under drought stress and its recovery.

## 5. DISCUSSION

The results of the present study showed that both JL-24 (drought susceptible) and K-9 (drought tolerant) genotypes differed in EWL on their leaf surface. In control set (100% FC), the EWL in K-9 genotype was found to be slightly higher than JL-24 genotype. Under drought stress, the EWL was found to increase consistently with the intensity of drought stress in both the genotypes. Moreover, the increase in EWL was found to be more significant in K-9 compared to JL-24 genotype. At 20% FC, the WUE in JL-24 genotype was found to increase by 46.84% whereas, in K-9 genotype, it increased by 88.33%. Under recovery of drought stress, both the genotypes showed considerable decrease in the EWL. After recovery, the EWL in K-9 genotype at 20% FC was found to be 16.42% more than JL-24 genotype. The leaf temperature did not show much change in control sets. Whereas, with the increase in the intensity of drought stress, the leaf temperature was found to increase in both the genotypes. However, the extent of increase in leaf temperature in JL-24 (drought susceptible) was found to be more significant than K-9 (drought tolerant) genotype. Under recovery of drought stress, the leaf temperature decreased in all the stress conditions in both the genotypes. The present findings showed that the tolerant peanut genotype (K-9) showed adaptive response to drought stress by increasing their EWL in the leaf which may assist in lowering the leaf temperature compared to susceptible genotype (JL-24). Although the role of EWL pertaining to drought stress is not clear, it may be predicted that involvement of EWL in drought tolerance is either due to reduction in cuticular transpiration or maintaining lesser leaf surface temperature under water-deficit condition by providing reflective surface for sunlight [10]. Under drought stress, the EWL was found to be increase in several crops such as wheat and pea [11,12]. Moreover, epicuticular waxes are known to improve the efficiency of water use through suppressing transpiration under dehydration in sorghum and peanut [13,14]. Studies have also suggested that in peanut genetic variability for leaf-canopy temperature exists [15,16] and it was negatively related with the yield [17].

## 6. CONCLUSION

The present study showed that K-9 (drought tolerant) genotype responds better than JL-24 genotype in their physiological characteristics such as enhancing their EWL in the leaf and lowering leaf temperature under drought stress, which may signifies its relative importance in drought-conditions. Although a thorough investigation is required to assess the contribution of EWL in providing drought tolerance in peanut plant under field conditions.

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