

Study of Drought Stress on ABA Accumulation and Proline among in Different Genotypes Forage Corn

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Abstract: In order to investigate the role of physiological factors, proline and ABA in the drought resistance of forage corn varieties, an experiment was carried out under drought (medium stress and high stress) and control conditions. A split plot design experiment using randomized complete block with four replication was used, so that irrigation treatment was considered as main plots and varieties forage corn as sub plots. Significant difference was found among the varieties at the proline and ABA. Results also indicated that drought stress dose affect different activity levels of proline and ABA. Therefore, selection for drought resistance by evaluation of proline and ABA in these varieties is useful.

Key words: Forage corn, drought, stress, proline, ABA, physiological factors, varieties

INTRODUCTION

Annually, great expenses are spent for researching on drought in crops. Though, the results show that simple and expensive methods are not presented yet, so, varieties resistance to drought be characterized. If one can describe sorghum drought resistance varieties in a simple way, that would enhance in the development of agricultural affairs. In all, drought is one of the most important constraints to the production of crops in droughty region, so it is of much importance to choose varieties with high efficiency in stress conditions. Researchers verify that in the crops which are under the effects of different stresses, the level of free radicals of peroxide in the tissue increases. Therefore, in this study the effect of the drought on the agronomical, physiological and biochemical properties of forage corn varieties is examined. Plants maybe subjected to environmental stresses that affect growth, metabolism and yields (Lawlor and Cornic, 2002). Drought is one of the most important stresses in crop production because it affect almost all plant function (Hernandez *et al.*, 2001). Crops have evolved several mechanisms to tolerance with drought stress (Zhu, 2001). Under water stress conditions, the lowering of the cellular water potential and the ABA accumulation give rise to a reorganization of the cellular metabolism, accumulation of osmolytes, such as proline, glycine, betaine can be one of the most molecular responses to water stress.

Under drought stress, plants stop growing completely and accumulate solutes in cells in order to maintain the

cell volume and turgor against dehydration which is named osmotic adjustment (Nomani, 1998; Patakas *et al.*, 2002). The principal role of proline probably is not to reduce the osmotic potential, but to protect enzymes against dehydration and salt accumulation (Thomas, 1990).

Nieves *et al.* (2001) has pointed out that ABA treatment caused an increase in the levels of proline compared to control. Plants stop growing completely and accumulation solutes on cells to maintain pressure of turgor dehydration (Nomani, 1998; Patakas *et al.*, 2002) stress may induce expand responses such as enhancement of plant hormones. Many kinds of stresses such as salt and water stress induct ABA synthesis (Swamy and Smith, 1999).

Increase in ABA content of leaves can be resulted during drought stress. However, the extent of the ABA response stress (Luo *et al.*, 1992). However, the extent of the ABA response depends on varietal differences (Wrightman, 1979). ABA levels in vegetative tissues can be elevated in response to various environmental stresses, Particularly under drought stress. Therefore, ABA is referred to as a stress hormone (Taylor *et al.*, 2000).

Objectives: Objectives of this study are:

- To determine effects of ABA and proline under normal irrigation and drought stress in corn
- To determine differences in the responses to drought stress between zea maize varieties

- To determine possibility of physiological role of proline and ABA for selection of corn's varieties

MATERIALS AND METHODS

This research was carried out in 2007 in the research Land of Agriculture College of Azad University of firouzabad fars province. A split plot design experiment using randomized complete block in which irrigation treatment as main plots and three varieties forage corn as sub plot with four replication was used.

The Relative Water Content (RWC) of leaves was measured with the following equation:

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

Freeze-dried tissue samples were homogenized and extracted in 80% methanol. Extracts were passed through a Sep-pak C18 cartridge. Methanol was removed under reduced pressure and the aqueous residue partitioned three times against ethyl acetate at pH 3.0. The ethyl acetate of the combined organic fractions was evaporates under reduced. The residue was taken up in TBS-buffer (TRIS-buffered saline; 150 mM NaCl, 1 mM MgCl_2 , 50 mM TRIS, pH 7.8) and subjected to an immunological ABA assay (ELISA), which described earlier (Peuke *et al.*, 1994). The accuracy of the ELISA was verified for maize in earlier investigations (Hartung *et al.*, 1994).

Extraction and determination of proline was preformed according to the method of Bates *et al.* (1973). Leaf samples (1 g) were extracted with 3% sulphosalicylic acid. Extracts (2 mL) were held for 1 h boiling water by adding 2 mL ninhydrin and 2 mL glacial acetic acid, after which cold toluene (4 mL) was added. Proline content was measured by a spectrophotometer (Shimadzu UV 1601) at 520 nm and calculated as $\mu\text{mol g}^{-1}$ against standard proline. The data were analyzed by mstat-c and minitab software.

RESULTS AND DISCUSSION

Drought treatment had significant effects on ABA and proline in compare with normal and mild irrigation (Fig. 1 and 2). The results showed significant differences ($p < 0.01$) between varieties for ABA and proline (Fig. 3 and 4). In fact, it can be observed that varieties of NS540 and BC666 had more ABA than varieties of BC582 and as shown in Fig. 4. On the other hand, it can suggested that drought stress stimulated ABA and proline accumulation and varieties of NS540 and Bc666 accumulated more proline and ABA with increasing

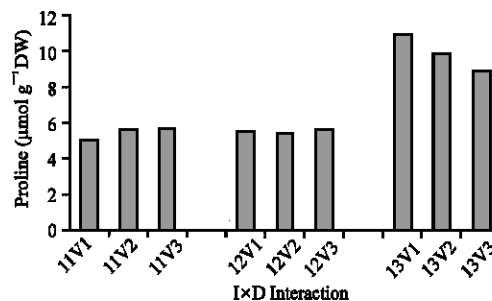


Fig. 1: Free proline contents in corns varieties under normal, mild, irrigation and high stress, I1 normal irrigation, I2 mild irrigation, I3 high stress, V1 = NS540, V2 = BC582 and V3 = BC666

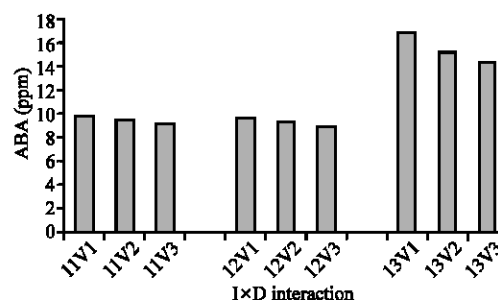


Fig. 2: Free ABA contents in corns varieties under normal, mild, irrigation and high stress, I1 normal irrigation, I2 mild irrigation, I3 high stress, V1 = NS540, V2 = BC582 and V3 = BC666

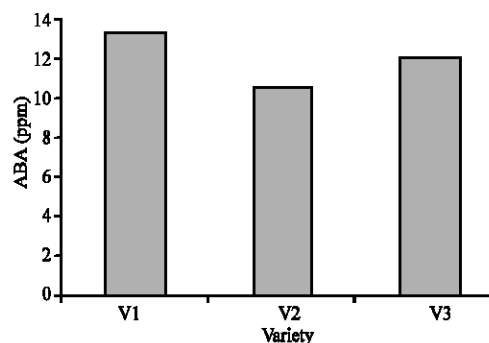


Fig. 3: ABA content in corns varietie, V1=NS540, V2 = BC582, V3 = BC666

drought comparatively high amounts of proline than other varieties (Fig. 4) and variety Bc582 Possessed less amounts of ABA, providing evidence for an efficient role of these metabolites as osmoprotectants under drought stress. The present data indicate that there are close relations between proline accumulation, ABA accumulation and drought stress and two physiological indices: ABA and proline including one of the best

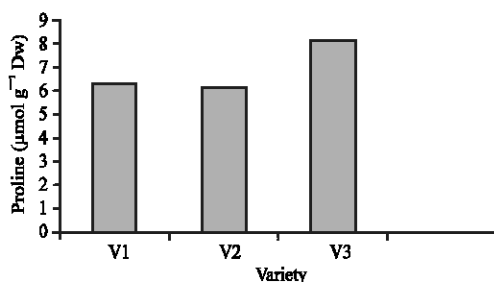


Fig. 4: Proline content in corns varieties, V1=NS540, V2 = BC582, V3 = BC666

indices to choosing, proline and ABA accumulation are increased by drought in *Zea mays*, Meyer and Boyer (1981) indicated that water deficiency caused a decreased growth in higher plants.

It is resulted that both proline and ABA are increased under high drought stress and genotypes of NS540 and BC582 have high amount of proline and ABA in drought stress and can be one of the best indices to selection resistance varieties of corn under drought, proline is a dominant organic molecule that acts as a mediator of osmotic adjustment under drought stress (Rontein *et al.*, 2002).

The osmolytes are known to function in protecting macromolecules by stabilizing protein structure under stress conditions (Mitysik *et al.*, 2002).

We demonstrated that of proline and ABA were significantly higher under drought stress, it may be due to the fact that under conditions of stress, growth is restricted and delivery of phosphate is impaired, resulting in the activation of the cellular phosphatase that release soluble of the cells (Thakur and Thakur, 1993).

CONCLUSION

Selection of tolerance genotypes *zea mays* can be considered in this study and two genotypes including high amount of ABA and proline can be in breeding programs, one of the mechanisms that plants use to combat the detrimental effects of water loss is to synthesis compatible solutes, typically certain polyols, sugar, amino acids, proline and related compounds (Bohnert and Jenson, 1996).

The properties of compatible solutes and proline facilitate the maintenance of favorable turgor pressure during water stress and in addition may serve as protective agents stabilizing proteins (Akashi *et al.*, 2001). Hence, increasing of ABA and proline in this study can be coordinate to high tolerance under drought stress.

ACKNOWLEDGEMENT

This research was supported by the Islamic Azad University of Kazeroun.

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