

## Behavior of Esterase, Peroxidase, Protein Profile and Growth Parameters of *Zea maize* and *Vicia faba* Cultivar under Heat Stress

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**I**N THE present study, the effect of temperature (20/10 °C and 35/20 °C) on the activities of esterase, peroxidase, protein profile and growth parameters in two different plants (*Vicia faba* cv. Assuit 95/2 and *Zea maize* cv. Hi Tech 20\*30) was investigated. *Vicia faba* cv. Assuit 95/2 was grown in pots at optimum temperature 20/10 °C exposed to heat stress 35/20 °C. While *Zea maize* cv. Hi Tech 20\*30 was grown in pots at optimum temperature 35/20 °C exposed to cooling stress 20/10°C for 15 days. Behavior of esterase, peroxidase, protein profile, growth parameters, and metabolic constituents of *Vicia faba* and *Zea maize* plants varied between increase and shortage under heat stress and also between two organs shoot and root. Under high temperature, the growth parameters (fresh weights, dry weights, length and leaf area) were greatly reduced. It can be observed that the two plants responded to the unfavorable conditions (heat stress) by the change in their activities of enzymes, in the number of isoenzymes which include esterase (EST) and peroxidase (PX) and also changes in their biochemical composition actor in the amount of protein, carbohydrates and proline.

**Keywords:** Heat stress, Protein profile, Isoenzymes, Proline, *Vicia faba* cv. Assuit 95/2, *Zea maize* cv. Hi Tech 20\*30.

### Introduction

Growth, physiology and metabolism of plants are known to be altered by heat stress (high temperature and chilling (Laudencia-Chingcuanco et al., 2011 and Han et al., 2014). The degree of these alterations depends mainly on the plant type and the plant growth stage. In most cases, there are quantitative and qualitative differences in the degree of plant responses to heat stress (Bita & Gerats, 2013).

Changing temperature beyond limits decreased elongation, shoot length, shoot dry weight and fruit yield (Audusseau et al., 2013). This depression of the relative growth rate varied in various types (Allen et al., 2013). Morphologically the most typical symptom of temperature stress injury to plant is the reduction of growth, and also the reduction of the leaf growth rate and shortening of the period of rapid leaf elongation by producing shorter leaves.

The chlorophyll and total carotenoids contents of leaves decrease in general under temperature

stress. Photosynthesis, one of the most important metabolic pathways in plants, is a target of temperature stress. Abscisic acid produced in response to temperature stress decreases turgor in guard cells and thus limits the CO<sub>2</sub> available for photosynthesis (Vikender et al., 2016). During temperature stress, reduction of chloroplast stromal volume and generation of active oxygen species also are thought to play an important role in inhibiting photosynthesis (Vikender et al., 2016). Photosynthesis depends on leaf chlorophyll content and stomatal conductance (Borjigidai & Yu, 2013).

Photosynthesis involves a long chain of mechanisms, enzymes and intermediate products and is regulated by several external and internal factors. Photosynthetic efficiency depends on the sequence of metabolic events such as photochemical reactions on the enzymes involved in carbon assimilation, on the structure of the photosynthetic apparatus and on the transport of photosynthetic intermediates between the subcellular compartments (Vikender et al., 2016). Photosynthetic rate is lower in the temperature-treated plants, but the photosynthetic

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potential is not greatly affected when the rates are expressed with regard to chlorophyll or leaf area. Decreases in photosynthetic rate are due to enhanced photooxidation and changes of enzyme activity induced by temperature stress (Waraich et al., 2012).

Temperature influences most plant processes, including photosynthesis, transpiration, respiration, germination, and flowering (Vikender et al., 2016). As temperature increases (up to a point), photosynthesis, transpiration, and respiration increase. When combined with day-length, temperature also affects the change from vegetative (leafy) to reproductive (flowering) growth. Depending on the situation and the specific plant, the effect of temperature can either speed up or slow down this transition (Collins & Parent, 2017).

Heat stress causes oxidative stress which is marked by the generation of reactive oxygen species (ROS). These species include singlet oxygen, superoxide anion, peroxide hydrogen and hydroxyl radical; all of them are said to damage the cell membrane (Kumar et al., 2012). Plants have developed a series of enzymatic and non-enzymatic detoxification systems to counteract ROS, and protect cells from oxidative damage (Sairam et al., 2002 and Nagesh Babu & Devaraj, 2008). The Antioxidant enzymes such as SOD, CAT, PX, and GR function in detoxification of super oxide and H<sub>2</sub>O<sub>2</sub> (Mittler, 2002 and Adak & Datta, 2005). Moreover, He (2010) found that differential responses to heat stress in activities and isozymes of four antioxidant enzymes for two cultivars of *Kentucky bluegrass* contrasting in heat tolerance.

Therefore, the present work was carried out to study behavior or mechanism tolerance of two plant *Zea maize cv. Hi Tech 20\*30* and *Vicia faba* under heat stress. This study can help in identification of heat tolerance of the plant and better understanding of physiological and biochemical defenses mechanisms associated with heat tolerance.

## Materials and Methods

The present work was carried out to study the effect of different temperature degree (20°C day/10°C night) and (35°C day /20°C night) on *Zea maize cv. Hi Tech 20\*30* and *Vicia faba cv. Assuit 95/2*.

*Zea maize cv. Hi Tech 20\*30* and *Vicia faba cv. Assuit 95/2* were obtained from the breeding program of Seeds Center, Beni Suef, Egypt; which were screened for germination responses to the different temperature degrees (20°C day/10°C night) and (35°C day /20°C night). *Zea maize cv. Hi Tech 20\*30* grains and *Vicia faba cv. Assuit 95/2* seeds were surface sterilized by immersion in a mixture of ethanol 96% and H<sub>2</sub>O<sub>2</sub> (1:1) for 3 min, followed by several washings with sterile distilled water. Five seeds were sown per Plastic pot. Each pot contained 500g of garden clay soil. All pots were irrigated with tap water kept at the field capacity. The grains were left to grow under these temperature degree (20°C day/10°C night) and (35°C day /20°C night) for 15 days. At the end of the experimental period, the dry matter yields of roots and shoots were determined. To determine the dry matter yields of roots, shoots were dried in an oven at 80°C until the constant dry weights were reached. Length of roots and shoots of two plant under study were determined. Leaf area in *Zea maize cv. Hi Tech 20\*30* was measured according to Norman & Campbell (1994) but Leaf area in *Vicia faba cv. Assuit 95/2* was carried by Wiersma & Bailey (1975). The soluble proteins were determined according to the method adopted by Lowry et al. (1951). Free proline was determined according to Bates et al. (1973).

The electrophoresis of protein profiles was carried out in vertical polyacrylamide gels, using the slab gel apparatus "SE 600, vertical slab gel" according to Laemmli (1970).

Data were obtained by Total Lab version 1.10 electrophoresis data system program (Scanalytics Inc.). The molecular weights of protein bands were determined against the protein marker 10 to 200 kDa. Isozymes were analyzed using the polyacrylamide gel electrophoresis (Native-PAGE 7.5%) the following enzymes (PX peroxidase, and EST esterase) were detected according to the methods described by Guikema & Sherman (1980), and Tanksley & Orton (1983), respectively.

## Results

### *Fresh and dry weight*

#### *Vicia faba cv. Assuit 95/2*

The data presented in Fig.1 reveal that, in root and shoot of *Vicia faba cv. Assuit 95/2*,

there is decrease in fresh and dry matter. This reduction was more pronounced in root than shoot. The percent of reduction in fresh weight of root and shoot were about 33.1 % and 84.5%, respectively in comparison with the corresponding

control plant at the (35°C /20°C). Moreover, the percent of reduction in dry matter yield was about 49% and 78.8 % in root and shoot, respectively in comparison with the corresponding control plant at the (35°C /20°C).

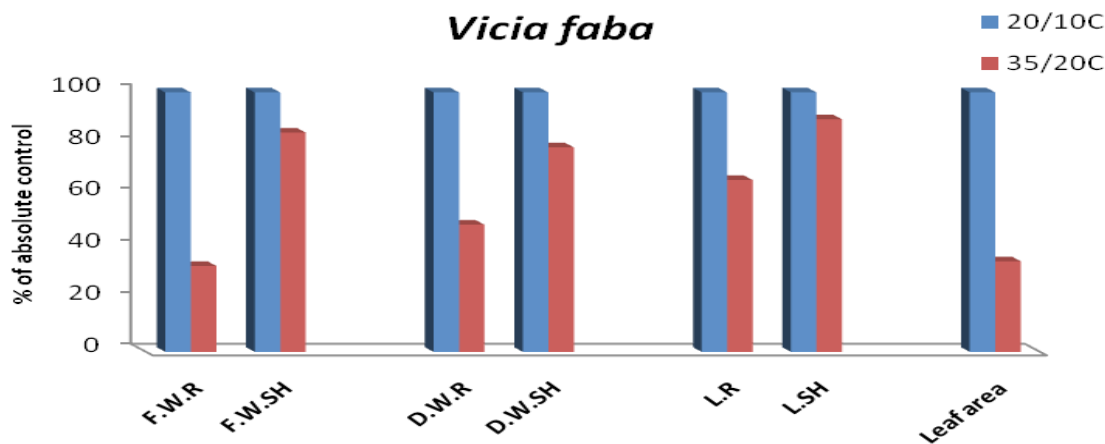


Fig.1. Fresh and dry weights (g), shoot and root lengths (cm/organ) and leaf area (Cm<sup>2</sup>/plant) of *Vicia faba* cv. Assuit 95/2 under different degrees of temperature (35/20°C and 20/10°C).

*Zea maize* cv. Hi Tech 20\*30:

Fresh, dry weights of shoot and dry weight of root were sharply decreased. On other hand, fresh weight of root was slightly increased under (20°C day/10°C night). The percentage of increasing in fresh weight was about 104.7 % in root, while the

percentage of reduction in fresh weight of shoot was 69.6%. And also, the percentage of reduction in dry matter was about 65.3 % and 55.6 % in root and shoot, respectively in comparison with the corresponding control plant (Fig 2).

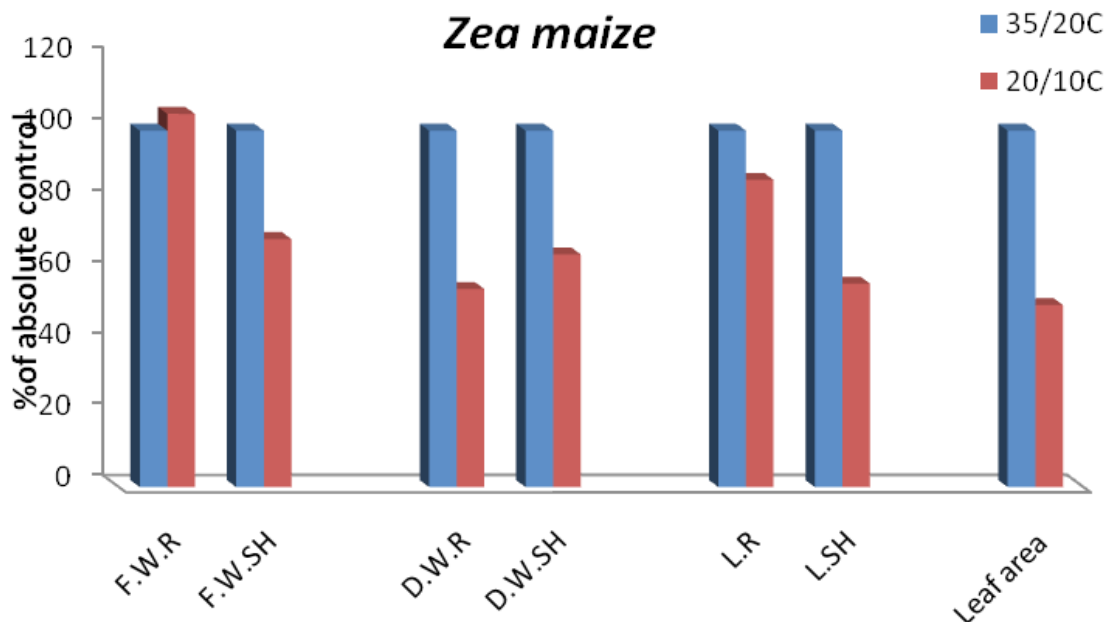


Fig. 2. Fresh and dry weight (g), shoot and root lengths (cm/organ) and leaf area (Cm<sup>2</sup>/plant) of *Zea maize* cv. Hi Tech 20\*30 different Under degrees of temperature(35/20°C and 20/10°C).

*Length of root and shoot:**Vicia faba cv. Assuit 95/2*

Shoot length was more or less unchanged at (35°C/20°C) degree. On other hand, the root length was sharply decreased with increasing temperature (35°C /20°C) (Fig.1).

*Zea maize cv. Hi Tech 20\*30:*

The lengths of root and shoot were sharply decreased with decreasing temperature (20°C/10°C). The percentage of reduction in length was about 86.1 % and 57.1% in root and shoot, respectively as compared with the corresponding control plant (Fig. 2).

*Leaf area:**Vicia faba cv. Assuit 95/2*

The results concerning the leaf area (cm<sup>2</sup>/ plant) of the various treatments are presented in

Fig. 1. It revealed that the leaf area of cv. Assuit 95/2 exhibited a marked and progressive decrease with increasing temperature and the percentage of this reduction was 34.9 % relative to the control.

*Zea maize cv. Hi Tech 20\*30:*

The leaf area of *Zea maize* exhibited a marked and progressive decrease with decreasing temperature and the percentage of reduction was 51.1 % as compared with the control (Fig 2).

*Photosynthetic pigments**Vicia faba cv. Assuit 95/2*

The concentration of Chl.a, Chl. b and carotenoids were decreased at high temperature and the percentage of reduction was 46.7%, 20.6% and 34.5%, respectively as compared with absolute control (Fig. 3).

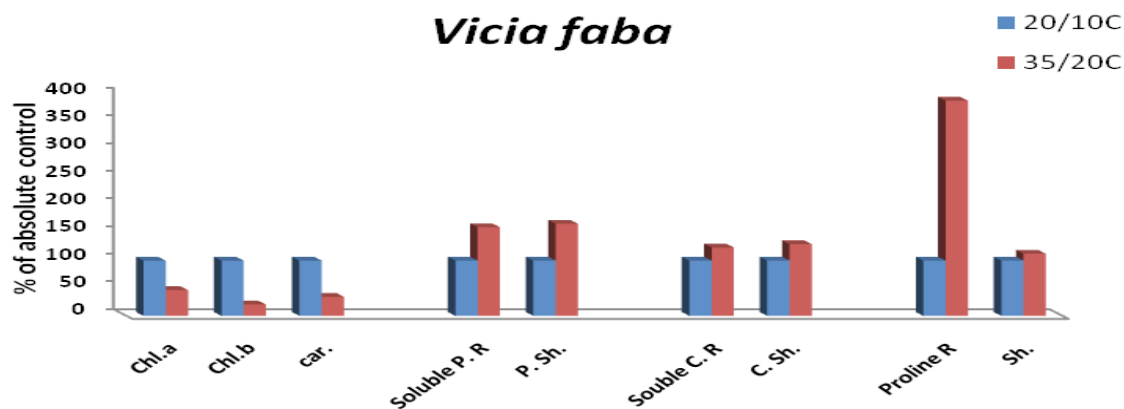


Fig. 3. Photosynthetic pigments (mg plant<sup>-1</sup>), soluble protein and soluble carbohydrate (mg plant<sup>-1</sup>) of shoot and root of *Vicia faba* cv. Assuit 95/2 under different degrees of temperature (35/20°C and 20/10°C).

*Zea maize cv. Hi Tech 20\*30*

The concentration of Chl.a decreased at low temperature the percent of reduction were 16.2 % as compared with absolute control. On other hand, the content of Chl. b and carotenoids increased at low temperature (Fig. 4).

*Soluble carbohydrate**Vicia faba cv. Assuit 95/2:*

Soluble carbohydrate increase in root and shoot. This accumulation was more pronounced in shoot than in root. The percent of increasing 129.6% and 123.7% in shoot and root as compared with absolute control, respectively (Fig. 3).

*Zea maize cv. Hi Tech 20\*30*

Soluble carbohydrate a slight reduced at low temperature in shoot. While in root, the content

of soluble carbohydrate highly reduced at low temperature. The percent of reduction 98% and 70% in shoot and root as compared with absolute control, respectively (Fig. 4).

*Soluble protein**Vicia faba cv. Assuit 95/2:*

Soluble protein, in root and shoot marked and progressive increase. The accumulation of soluble protein more pronounced in shoot than root as compared with control (Fig. 3).

*Zea maize cv. Hi Tech 20\*30:*

In root and shoot a marked and progressive decrease in soluble protein. These reductions more pronounced in shoot than root as compared with control (Fig. 4).

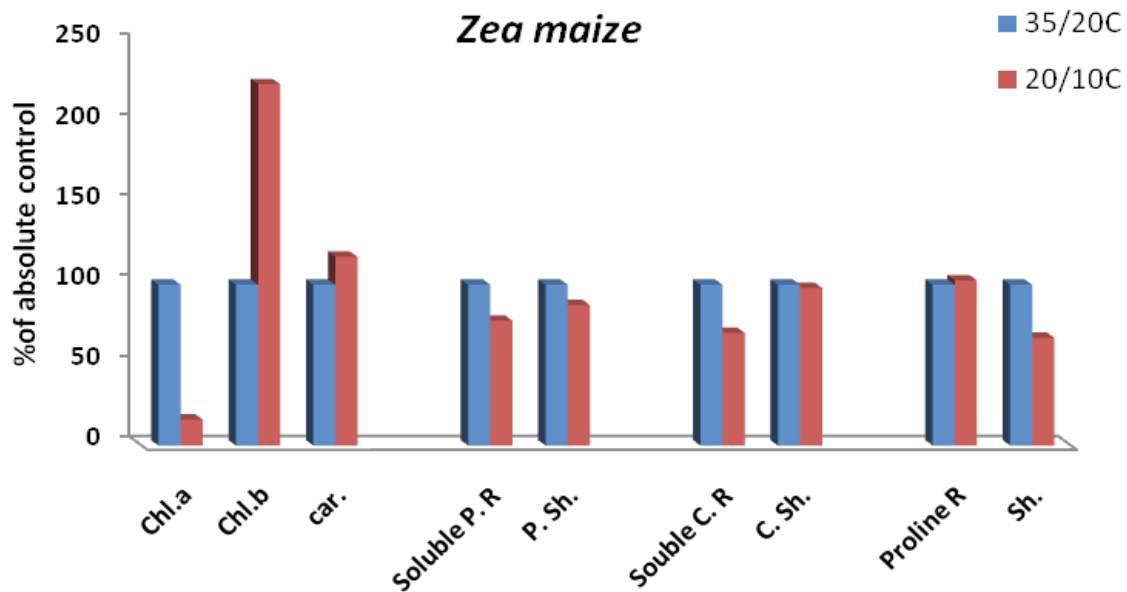


Fig. 4. Photosynthetic pigments (mg plant<sup>-1</sup>), soluble protein and soluble carbohydrate (mg plant<sup>-1</sup>) of shoot and root of *Zea maize* cv. Hi Tech 20\*30 under different degrees of temperature (35/20°C) and 20/10°C).

#### Proline

##### *Vicia faba* cv. Assuit 95/2:

The accumulation of proline was observed in two organs but it was more pronounced in root than shoot under high temperature (Fig. 3).

##### *Zea maize* cv. Hi Tech 20\*30:

In shoot a marked and progressive decrease in proline, while the accumulation of proline more or less unchanged in root at low temperature as compared with control (Fig. 4).

#### Protein profile

Heat stress caused an induction in the synthesis of some new polypeptides in two plant compared to control one (Tables 1,2 and Fig. 5). Generally, the electrophoretically separated protein under heat stress as compared with control revealed (i) Quantitative decline in certain proteins, (ii) Rise in levels of other proteins (density of protein bands), (iii) Some proteins remained unchanged, and (iv) *de novo* induction of specific proteins. In *Vicia faba* cv. Assuit 95/2, levels of proteins with molecular weights 19.766, 13.106 and 11.687 kDa polypeptides in shoot and 60.843, 15.369, 12.982 and 9.777 kDa in root, were common bands under different temperature. Application of high temperature (35/20°C) caused changes in the levels of proteins with molecular weights of 46.858, 35.973 and 23.178 kDa in root however,

54.947 and 29.81 kDa in shoot. On other hand in *Zea maize* cv. Hi Tech 20\*30, Levels of proteins with molecular weights 19.392, 12.258, 9.871 and 8.663 kDa polypeptides in shoot and 13.836 and 9.871 kDa polypeptides in root, were common bands under different treatment. Application of low temperature (20/10°C) caused changes in the levels of proteins with molecular weights of 176.845, 89.45, 53.39 and 29.81 kDa in shoot while, 167.542, 101.928, 59.691 and 33.753 kDa in root. These alterations ranged in molecular weight from as low as 8 kDa to as high as 200 kDa. Moreover, the intensity of bands was changed in two organs of both plants (Fig.5). From the general picture of stress proteins emerging from this work, one point is noteworthy, more protein alterations were scored in stressed plant than unstressed plant in both plant, moreover, these alterations in protein highly observed in two oranges under study in both plant. It is possible that this differential response in both plants reflect their relative sensitivities to stress conditions.

#### Isozymes activity

The electrophoresis profiles of PX and EST isozymes showed that isozymes activity was affected by heat stress and a differential PX and EST isozymes profiles between two plants and also in two organs were observed (Fig. 6 and 7).

**TABLE 1. SDS electrophoresis analysis of protein bands produced by *Vicia faba* cv. Assuit 95/2 from the shoot and root under different temperature 35/20°C and 20/10°C.**

<i>Vicia faba</i>	Shoot		Root	
	20/10°C	35/20°C	20/10°C	35/20°C
MW/KDa				
Band1	81.298	54.947	99.998	60.843
Band2	59.123	29.810	60.843	46.858
Band3	33.432	19.766	33.753	35.973
Band4	19.766	13.106	19.766	23.178
Band5	13.106	11.687	15.369	15.369
Band6	11.687		12.982	12.982
Band7	9.777		9.777	9.777
Number of bands	7	5	7	7

**TABLE 2. SDS electrophoresis analysis of protein bands produced by *Zea maize* cv. Hi Tech 20\*30 from the shoot and root under different temperature 35/20°C and 20/10°C.**

<i>Zea maize</i>	Shoot		Root	
	35/20°C	20/10°C	35/20°C	20/10°C
MW/KDa				
Band1	140.158	176.845	176.845	167.524
Band2	56.365	89.450	37.733	101.928
Band3	31.569	53.394	23.401	59.691
Band4	25.020	29.810	13.836	33.753
Band5	19.392	19.392	9.871	13.836
Band6	12.258	12.258	-----	9.871
Band7	9.871	9.871	-----	-----
Band8	8.663	8.663	-----	-----
Number of bands	8	8	5	6

Electrophoresis patterns of esterase EST and peroxidase PX isozymes showed differences in density and number of bands among control and under heat stress in two plants and also in root and shoot. And also, this difference in density and number of bands are more pronounced under heat stress (Tables 3, 4 and Fig. 6, 7). Peroxidase PX electrophoresis patterns are illustrated in Table 1 and Fig. 6. In *Zea maize* cv. Hi Tech 20\*30, two bands were present in all treatments (monomorphic bands) at Rf 0.091 and 0.646 in shoot and root under control and heat stress. In *vicia faba*, two bands were present in all treatments at Rf 0.071 and 0.616. In addition, one unique band was detected in root under heat stress treatment at Rf 0.293.

Esterase electrophoresis patterns are illustrated in Table 4 and Fig. 7. In *Zea maize* cv. Hi Tech 20\*30, six bands (monomorphic bands) exhibited with different densities and intensities in root under control and heat stress. In shoot, six bands with different intensities and densities were observed among the profiles of all treatments (control and heat stress). One band was absent at Rf 0.529 in shoot under heat stress. In *Vicia faba*, isozyme of esterase under the heat stress show some increase in activity and in the number of bands in root and shoot. Also, one and two isozymes bands were induced in shoot and root under heat stress treatments at Rf 0.529 and (0.234 and 0.529), respectively.

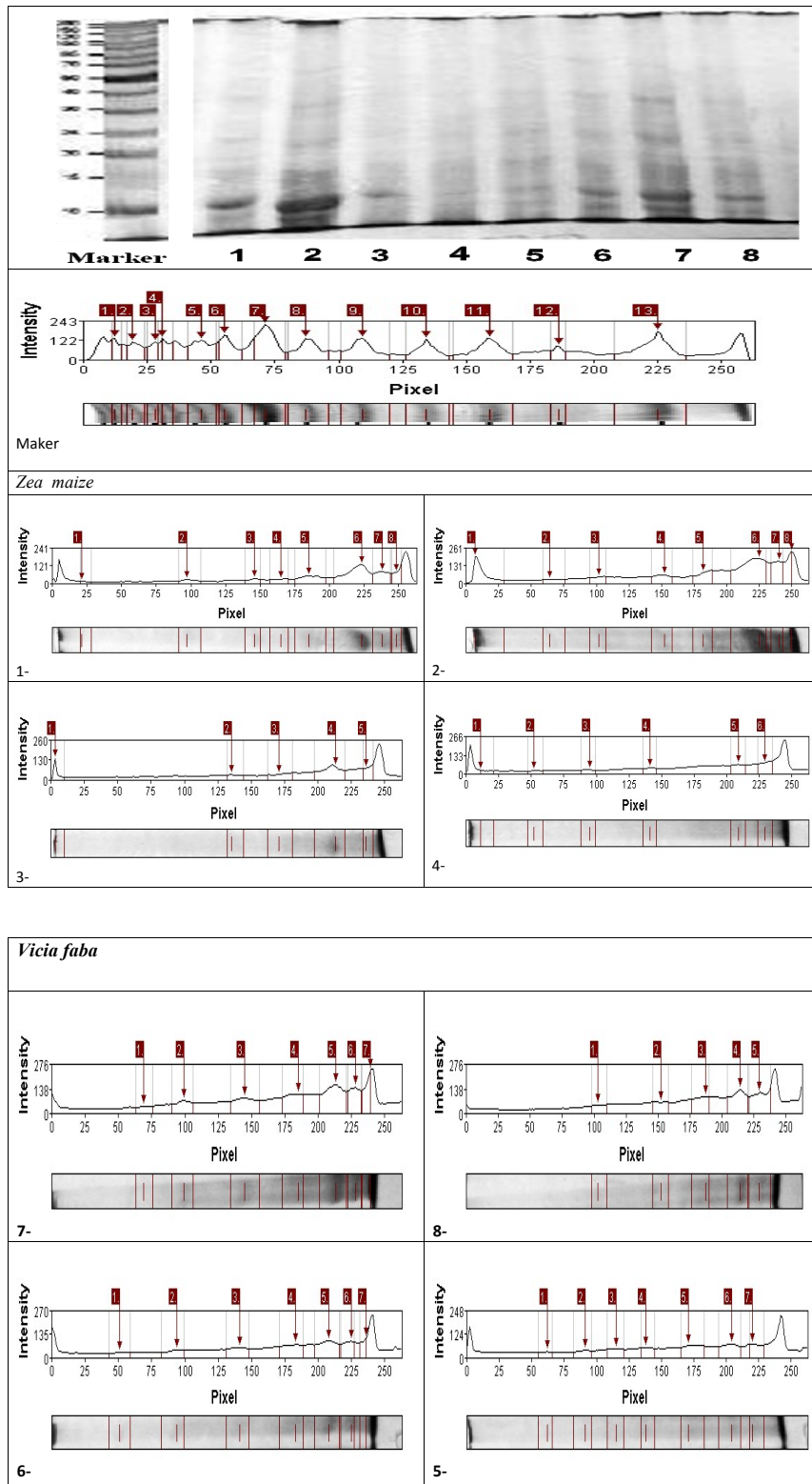


Fig. 5. Electrophoretic banding profile of protein and Scanogram of protein profiles extracted from the leaves and root of *Vicia faba* cv. Assuit 95/2 and *Zea maize* cv. Hi Tech 20\*30 under different temperature 35/20°C and 20/10°C. Lane 7: shoot under 20/10°C, lane 8: shoot under 35/20°C, lane 6: root under 20/10°C and lane 5: root under 35/20°C of *Vicia faba* cv. Assuit 95/2 .

Lane 1: Shoot under 35/20°C , lane 2: shoot under 20/10°C, lane 3: root under 35/20°C and lane 4: root under 20/10° C of *Zea maize* cv. Hi Tech 20\*30.

TABLE 3. RF analysis of electrophoretic banding profile of peroxidase produced by *Vicia faba* cv. Assuit 95/2 and *Zea maïze* cv. Hi Tech 20\*30 from the shoot and root under different temperatures 35/20°C and 20/10°C.

Rf	<i>Zea maïze</i>				<i>Vicia faba</i>			
	Shoot		Root		Shoot		Root	
	35/20°C	20/10°C	35/20°C	20/10 °C	35/20 °C	20/10°C	20/10°C	35/20°C
PX3	0.091	0.091	0.091	0.091	0.071	0.071	0.071	0.071
PX1	0.646	0.646	0.646	0.646	0.616	0.616	0.616	0.616
PX2	-----	-----	-----	-----			-----	0.293

TABLE 4. RF analysis of electrophoretic banding profile of esterase produced by *Vicia faba* cv. Assuit 95/2 and *Zea maïze* cv. Hi Tech 20\*30 from the shoot and root under different temperature 35/20°C and 20/10°C.

RF	<i>Zea maïze</i>				<i>Vicia faba</i>			
	Shoot		Root		Shoot		Root	
	35/20°C	20/10°C	35/20°C	20/10°C	20/10°C	35/20°C	20/10°C	35/20°C
EST 7	0.189	0.189	-----	-----	0.189	0.189	-----	
EST 6	0.243	0.243	0.243	0.243	0.243	0.243		0.243
EST 5	0.304	0.304	0.304	0.304	0.304	0.304	0.307	0.307
EST 4	0.376	0.376	0.376	0.376	0.376	0.376	-----	-----
EST 3	0.529	----	0.529	0.529		0.529		0.529
EST 2	0.629	0.629	0.629	0.629	0.629	0.629	0.629	0.629
EST 1	0.739	0.739	0.739	0.739	0.739	0.739	0.739	0.739

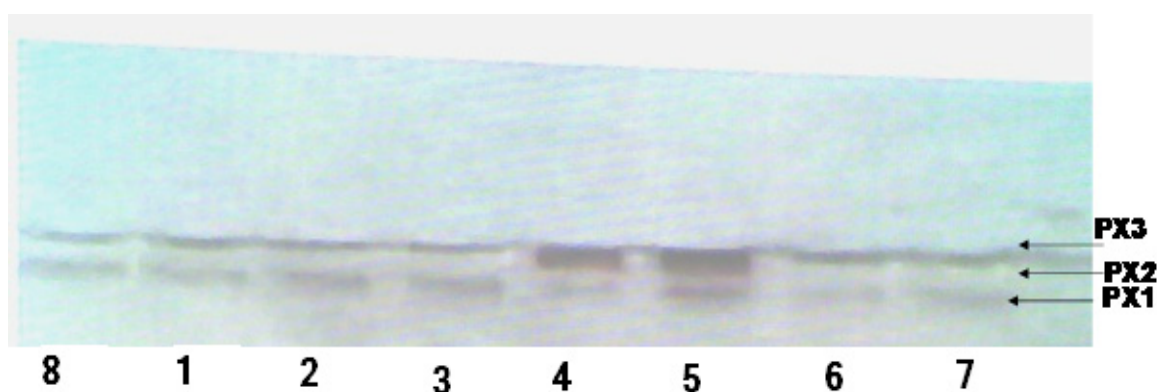
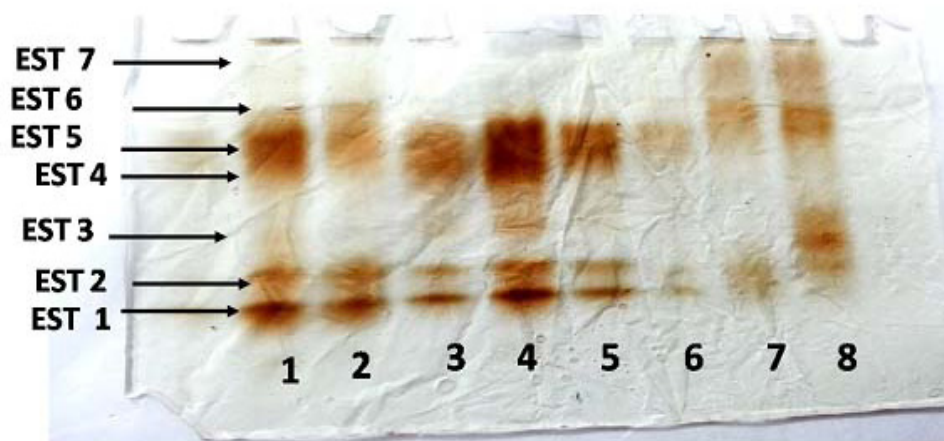


Fig. 6. Electrophoretic banding profile of peroxidase from the shoot and root of *Vicia faba* cv. Assuit 95/2 and *Zea maïze* cv. Hi Tech 20\*30 under different temperature 35/20°C and 20/10°C. Lane 7: shoot under 20/10°C, lane 8: shoot under 35/20°C, lane 6: root under 20/10°C and lane 5: root under 35/20 C of *Vicia faba* cv. Assuit 95/2.

Lane 1: Shoot under 35/20°C, lane 2: shoot under 20/10°C, lane 3: root under 35/20°C and lane 4: root under 20/10°C of *Zea maïze* cv. Hi Tech 20\*30.





**Fig. 7.** Electrophoretic banding profile of esterase from the leaves and root of *Vicia faba* cv. Assuit 95/2 and *Zea maize* cv. Hi Tech 20\*30 under different temperature 35/20°C and 20/10°C. Lane 7: shoot under 20/10°C, lane 8: shoot under 35/20°C, lane 6: roots under 20/10°C and lane 5: root under 35/20°C of *Vicia faba* cv. Assuit 95/2 .

Lane 1: shoot under 35/20°C, lane 2: shoot under 20/10°C, lane 3: roots under 35/20°C and lane 4: root under 20/10°C of *Zea maize* cv. Hi Tech 20\*30.

### Discussion

The plants under study differ in their acclimation to heat stress which exerts adverse effects on plant growth and development. One of the major environmental factors limiting the productivity of crops is heat stress which negatively affects the metabolism of plants and causes important modification in different biochemical and molecular processes (Baurle, 2016). The two plants agree on the negative effects of heat stress on root/shoot length, fresh and dry mass production (Baghour et al., 2002 and Waheed et al., 2007). Reduced growth in stressful medium is a typical phenomenon that has been interpreted as a change in metabolism initiated to resist stress. Moreover, heat stress may induce osmotic stress, oxidative stress and protein denaturation in two plants, which lead to cellular adaptive responses and accumulation of compatible organic solutes such as soluble carbohydrates, amino acids and proline (Dufooo-Hurtado et al., 2013). In the present study, adaptive responses in *Vicia faba* cv. Assuit 95/2 were represented in accumulation of soluble protein in root and shoot, soluble carbohydrate in two organs and also proline. On other side, in *Zea maize* cv. Hi Tech 20\*30, it was achieved in accumulation of soluble protein in shoot only and reduction of soluble protein in root. On the other hand, soluble carbohydrate and proline in two organs were reduced. In addition to their role in cell water relations, organic solute

might helped towards the removal of free radicals, and stabilization of macromolecules, such as proteins, protein complexes and membranes (Bohnert & Shen, 1999; Bray et al., 2000 and Bita & Gerats, 2013). Metabolic imbalances caused by ionic toxicity under heat stress may also lead to oxidative stress and cause accumulation of reactive oxygen species (ROS). Plants employed antioxidants compounds and detoxifying enzymes e.g., (superoxide dismutase, catalase, and enzymes of ascorbate-glutathione cycle) to resist oxidative stress (He, 2010 and Mansoor & Naqv, 2013). Transgenic plants over expressing ROS scavenging enzymes, such as superoxide dismutase (Alscher et al., 2002), ascorbate peroxidase (Wang et al., 1999) and glutathione S-transferase/glutathione peroxidase (Roxas et al., 2000) showed increased tolerance to osmotic, temperature, and oxidative stresses.

The results showed that, three bands were exhibited with different densities and intensities in two plants grown under controlled temperature and under heat stress treatment. Two bands of peroxidase are common bands. Peroxidase activity in roots of two plants under study increased under heat stress treatment. These results are similar to those of Mohamed & Abdel-Hamid (2013) who found that three bands were exhibited with different densities and intensities in cotton genotypes grown under control temperature and heat stress treatment. Two bands are common

bands which detected at Rf 0.30 and 0.55. Peroxidase activity in the tolerant genotypes of cotton (Giza 85 and Giza 92) increased under heat stress treatment (40°C). Peroxidases are heme-containing oxidoreductases that participate in a number of metabolic processes, such as regulation of cell elongation, lignifications, cross linking of cell wall structural proteins and phenolic oxidation (Kumar et al., 2012 and Silva et al. 2015). Seven bands of esterase electrophoretic patterns were observed among the profile of all treatments. Five bands were common present in some treatments with substantial differences in their intensities and densities. Also, two isozyme bands were induced in two organs of *Vicia faba cv. Assuit 95/2* and *Zea maize cv. Hi Tech 20\*30* under heat stress treatments. These results are in accordance with those of Mohamed & Abdel –Hamid (2013) who found four bands in some treatments and absence of others (polymorphic) with substantial differences in their intensities and densities in cotton plants. One band which has Rf 0.02 was present in all treatments (monomorphic bands). In addition, two unique bands were detected in the tolerant genotype (Giza 92) under heat stress treatment (Silva et al., 2015)

Three types of modifications are observed in the protein patterns of two organs of *Vicia faba cv. Assuit 95/2* and *Zea maize cv. Hi Tech 20\*30* some protein bands disappeared, other proteins selectively increased and synthesis of new set of proteins was induced. Some of these responses were observed under heat stress treatments. These results are in accordance with those of Mohamed & Abdel –Hamid (2013). These new protein bands may be the HSPs or the enzyme of the antioxidant systems which plays very important role in providing tolerance against oxidative burst which is in conformity with the observation made by Singh & Khurana (2016). Another approach to understand the molecular basis of heat stress tolerance is to identify stress induced changes in the protein expression (Bita & Gerats, 2013 and Silva et al. 2015).

#### Final conclusion

In conclusion, both non enzymatic and enzymatic antioxidant mechanisms responded distinctly to temperature stress. In two plant under study have divergent of response mechanisms to heat stress. Response two plant (*Zea maize cv. Hi Tech 20\*30* and *Vicia faba cv. Assuit 95/2*) to heat stress involves few common enzymatic and non

enzymatic components. This is reflected in plant growth and plant resistance to stress.

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#### References

- Adak, S. and Datta, A.K. (2005) Leishmania major encodes an unusual peroxidase that is a close homologue of plant ascorbate peroxidase: a novel role of the transmembrane domain. *Biochem. J.* **390**, 465-474.
- Allen, C.R., Stein, O.R., Hook, P.B., Burr, M. D., Parker, A.E. and Hafila, E.C. (2013) Temperature, plant species and residence time effects on nitrogen removal in model treatment wetlands. *Water Sci. Technol.* **68**, 2337-2343.
- Alscher, R.G., Nevel, E. and Heath, S.L. (2002) Role of superoxide dismutases (SODs) in controlling oxidative stress in plants. *J. Exp. Bot.* **372**, 1331-1341.
- Audusseau, H., Nylin, S. and Janz, N. (2013) Implications of a temperature increase for host plant range: predictions for a butterfly. *Ecol. Evol.* **3**, 3021-3029.
- Baghour, M., Moreno, D.A., Villora, G., Lopez-Cantarero, I., Hernandez, J., Castilla, N. and Romero, L. (2002) Root-zone temperature influences the distribution of Cu and Zn in potato-plant organs. *J. Agric. Food Chem.* **50**, 140-146.
- Bates, L.S., Waldren, R.P. and Teare, I.D. (1973) Rapid determination of free proline for water stress studies. *Plant and Soil*, **39**, 205-207.
- Baurle, I. (2016) Plant heat adaptation: priming in response to heat stress. *F1000Res.*, **5**, F10005.
- Bita, C.E. and Gerats, T. (2013) Plant tolerance to high temperature in a changing environment: scientific fundamentals and production of heat stress-tolerant crops. *Frontiers in Plant Science*, **4**.
- Bohnert, H.J. and Shen, B. (1999) Transformation and compatible solutes. *Sci. Hortic.* **78**, 237- 260.
- Borjigidai, A. and Yu, G.R. (2013) Temperature dependence of parameters of plant photosynthesis models: A review. *Ying Yong Sheng Tai Xue Bao*, **24**, 3588-3594.
- Bray, E.A., Bailley-Serres, J. and Weretilnyk, E. (2000) Responses to abiotic stresses. In: "*Biochemistry and*

- Molecular Biology of Plants*" , Gruissem, W. et al. (Ed.), pp. 1158–1203, American Society of Plant Physiologists. Rockville, Md.
- Collins, N.C. and Parent, B. (2017) Diverging temperature responses of CO<sub>2</sub> assimilation and plant development explain the overall effect of temperature on biomass accumulation in wheat leaves and grains. *AoB Plants.*, plw 092
- Dufoo-Hurtado, M.D., Zavala-Gutierrez, K.G., Cao, C.M., Cisneros-Zevallos, L., Guevara-Gonzalez, R.G., Torres-Pacheco, I., Vazquez-Barrios, M.E., Rivera-Pastrana, D.M. and Mercado-Silva, E.M. (2013) Low-temperature conditioning of "seed" cloves enhances the expression of phenolic metabolism related genes and anthocyanin content in 'Coreano' garlic (*Allium sativum*) during plant development. *J. Agric. Food Chem.* 61, 10439-10446
- Guikema, J.A. and Sherman, L.A. (1980) Electrophoretic profiles of cyanobacterial membrane polypeptides showing heme dependent peroxidase activity. *Biochem. Biophys. Acta*, **637**, 189-201.
- Han, Z., Ku L., Zhang, Z., Zhang, J., Guo, S., Liu, H., Zhao, R., Ren, Z., Zhang, L., Su, H., Dong, L. and Chen, Y. (2014) QTLs for Seed Vigor-Related Traits Identified in Maize Seeds Germinated under Artificial Aging Conditions. *PLoS ONE.*, **9**, e92535.
- He, Y. (2010) Differential responses to heat stress in activities and isozymes of four antioxidant enzymes for two cultivars of Kentucky bluegrass contrasting in heat tolerance. *Crop Sci.* **135** (2), 116–124.
- Kumar, R.R., Goswami, S., Sharma, Singh K., Gadpayle, K.A., Kumar, N., Rai, G.K., Singh, M. and Rai, R.D. (2012) Protection against heat stress in wheat involves change in cell membrane stability, antioxidant enzymes, osmolyte, H<sub>2</sub>O<sub>2</sub> and transcript of heat shock protein. *Int. J. Plant Physiol. Biochem.* **4**, 83-91.
- Laemmli, U.K. (1970) Cleavage of structural proteins during the assembly of the head of T4 bacteriophage. *Nature*, **227**, 680-685.
- Laudencia-Chingcuanco, D., Ganeshan, S., You, F., Fowler, B., Chibbar, R. and Anderson, O. (2011) Genome-wide gene expression analysis supports a developmental model of low temperature tolerance gene regulation in wheat (*Triticum aestivum* L.). *BMC Genom.* **12**, 299.
- Lowry, O.H., Rosebrough, N.J., Farr, A.R. and Randall, R.J. (1951) Protein measurement with Folin- Phenol reagent. *J. Biol. Chem.* **193**, 265-275.
- Mansoor, S. and Naqv, F.N. (2013) Effect of heat stress on lipid peroxidation and antioxidant enzymes in mung bean (*Vigna radiata* L) seedlings. *African Journal of Biot.* **12**, 3196-3203.
- Mohamed, H.I. and Abdel-Hamid, A.M.E. (2013) Molecular and biochemical studies for heat tolerance on four cotton genotypes. *Romanian Biotechnological Letters*, **18** (6), 8823-8831.
- Mittler, R. (2002) Oxidative stress, antioxidants and stress tolerance. *Trends Plant Sci.* **7**, 405-410
- Nagesh Babu, R. and Devaraj, V.R. (2008) High temperature and salt stress response in French bean (*Phaseolus vulgaris*). *Australian Journal of Crop Sci.* **2** (2), 40-48.
- Norman, J., Campbell, G. S. (1994) Canopy structure. In: "*Plant Physiological Ecology*", Percy R. W., Ehleringer J., Moony H. A., Rundel P. W. (Ed.), pp. 301-326. Chapman & Hall, London. *Physiol. Plant*, **115**, 251–257.
- Roxas, V.P., Lodhi, S.A., Garrett, D.K., Mahan, J. R. and Allen, R.D. (2000) Stress tolerance in transgenic tobacco seedlings that overexpress glutathione S-transferase/glutathione peroxidase. *Plant Cell Physiol.* **41**, 1229-1234.
- Sairam, R.K., Rao, K.V., Srivastava, G.C. (2002) Differential response of wheat genotypes to long term salinity stress in relation to oxidative stress, antioxidant activity and osmolyte concentration. *Plant Sci.* **163**, 1037-1046.
- Silva, L. A., Fatima Sales, J. , Oliveira, J.A. , Resende Von Pinho, É.V., Santos, H.O. and Soares M. A. (2015) Desiccation tolerance of Rhamnium elaeocarpum Reissek (Rhamnaceae) seeds. *Acta Scient.* **37**(2), 181-189.
- Singh, A. and Khurana, P. (2016) Molecular and Functional Characterization of a Wheat B2 Protein Imparting Adverse Temperature Tolerance and Influencing Plant Growth. *Frontiers in Plant Science*, **7**, 642.
- Tanksley, S.D. and Orton, T.J. (1983) "*Isozymes in Plant Genetics and Breeding*". parts A & B. Elsevier, Amsterdam.
- Vikender, K., Sunder, S. and Rishi, K.B. (2016) Heat and drought tolerance in wheat: integration of physiological and genetic platforms for better performance under stress . *Ekin. Journal of Crop Breeding and Genetics*, **2** (1), 1-14.
- Waheed, A., Gelani, S., Ashraf, M. and Foolad, M.R. (2007) Heat tolerance in plants: An overview. *Environ. Exp. Bot.* **61**
- Wang, J., Zhang, H. and Allen, R.D. (1999) Overexpression of an Arabidopsis peroxisomal

- ascorbate peroxidase gene in tobacco increases protection against oxidative stress. *Plant Cell Physiol.* **40**, 725-732.
- Waraich, E.A., Ahmad, R., Halim, A. and Aziz, T. (2012) Alleviation of temperature stress by nutrient management in crop plants: a review. *Journal of Soil Science and Plant Nutrition*, **12**, 221-244.
- Wiersma, J.V. and Bailey, T.B. (1975) Estimation of leaflet, trifoliolate and total leaf area of soyabean. *Agron. J.* **67**, 26–30.

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## سلوك انزيم استريز، البيروكسيديز، التفريد الكهربى للبروتين ودلات النمو في نبات الفول ونبات الذرة تحت الإجهاد الحراري

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قسم النبات والميكروبيولوجي – كلية العلوم – جامعة المنيا – المنيا - مصر.

تم زراعة نبات الفول البلدي أسبوط 95/2 لمدة خمسة عشر يوماً وتم تقسيمه إلى مجموعتين، المجموعة الأولى تحت تأثير درجة الحرارة المثلى (20/10 درجة مئوية) والمجموعة الثانية تعرضت لإجهاد حراري 35/25 درجة مئوية، أما نبات الذرة هاي تك 30\*20 فقد قسم أيضاً إلى مجموعتين. المجموعة الأولى تحت تأثير درجة الحرارة المثلى (35/25 درجة مئوية) والمجموعة الثانية تعرضت لإجهاد حراري (20/10 درجة مئوية) لمدة 15 يوماً.

لقد أظهرت الدراسة تأثير درجة الحرارة (20/10) درجة مئوية و35/20 درجة مئوية على انزيم استريز، البيروكسيديز، التفريد الكهربى للبروتين ودلات النمو في نبات الفول البلدي صنف أسبوط 95/2 ونبات الذرة هاي تك 30\*20. أظهر سلوك استريز، البيروكسيديز، التفريد الكهربى للبروتين، ودلات النمو والإيض في نبات الذرة ونبات الفول اختلافاً واضحاً ما بين الزيادة والنقص تحت الإجهاد الحراري.

وأيضاً وجد تأثير واضح على طول الجذور والسيقان في كلا النباتين تحت الإجهاد الحراري، بينما أظهر النمو متمثلاً في (الوزن الجاف والرطب ومساحة الأوراق) نقصاً واضحاً في كلا النباتين.

وقد أظهرت النباتات قيد البحث استراتيجيه واضحة في مواجهة الإجهاد الحراري عن طريق التغير في نشاط وعدد المشابهات الأنزيميه لانزيمى الاستريز والبيروكسيديز وايضا تراكم العديد من المركبات الأسموزيه ممثله في البروتين والبرولين والكاربوهيدرات.