

Rapid responses and physiological events of leaf growth in response to water stress induced by poly ethylene glycol in maize (*Zea mays* L.)

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ABSTRACT/ RESUME

Abstract: To follow the rapid physiological events of the osmotic adjustment (OA) known as a mechanism adaptation of leaf growth to water stress, three-weeks old maize plants *Zea mays* subsp. *mays* were exposed to water stress by the addition of polyethylene glycol (PEG) 6000 to the hydroponics solution. The water deficiency caused a quick cessation of elongation rate measured on the 6th growing leaf, as a result of turgor decrease. For this period, the stomatal conductance was maintained temporarily before going down partially, while the photosynthesis continued during stress. After ~30 min. the osmotic potential (ψ_{π}) started to decrease. Thus, the OA was quickly generated, turgor recovered, and returned to its previous level before the stress. Simultaneously, the leaf elongation rate (LER) partially recovered. The recovery of turgor was quite associated with the osmotic adjustment. In parallel, with the fast increase of the abscisic acid (ABA) in growing not transpiring leaf, with the partial opening of stomata in transpiring leaf, that makes the important connotation.

I. Introduction

Water stress, is one of the very first aspects involved in restrictive growth as well as yields; it strongly affects the agricultural production of arid and semi-arid zones, characterized by high radiation and random rains. Maize (*Zea mays* L.) is a versatile crop with wide flexibility to different agro-climatic conditions [1], in situations of high water deficit, the loss of yield in maize can go up to 50-60% [2]. Maize can be produced at yearly precipitation levels as nether as 200 mm [3], and recently, maize cultivation has been steadily expanded to arid and semi-arid zones, which perform a considerable amount of ground resources [4].

The osmotic adjustment (OA) is an important physiological adaptation in plants, as a lowering of osmotic potential (ψ_{π}) due to net solute

accumulation in tissues. OA has been considered to be a beneficial drought tolerance mechanism in some crop species, such as maintenance of turgor under water stress.

The most important in water state is the role of turgor in monitoring leaf growth under water deficit, that it is well discussed [5,6,7]. Others have focused on the role of root signals in controlling the growth under water stress such as the ABA [8,9,10]. But that's still a great debate about the point at which leaf growth is first controlled by the water state, and how far did the abscisic acid (ABA) intervene? However, it is possible that the diversification of plant categories can enrich the challenge, without neglecting the role of wall extensibility on cell growth [11]. The most studies on the OA have been carried out long term under the effect of water or salt stress [12,13]. At short term, through some studies at tissue or at cell level,

indicated also that the OA is a mechanism by which plants decrease their osmotic rapidly allowing the stability of turgor at inferior water status through short time-scale [14,7].

The osmotic adjustment increased rapidly simultaneously with the intervention of ABA under rapid osmotic stress [9, 10]. Sudden environmental modifications cause quick and frequently transient varieties in leaf elongation, by a temporary arrest, followed by a period of partial restoration of leaf elongation [15,6,7].

The pressurization technique has indicated clearly the preference advantage of turgor in the rapid recovery. It is known that, will be no rapid turgor recovery without a rapid OA [15]. The discussion is still about the point at which leaf growth is first controlled by the water state, and how far did the ABA intervene? However, it is possible that the changing the studied of plant categories can enrich the debate, C4 plants for example. The OA mechanism in wheat (C3 plants) occurs after stress application is quickly within 1-2 h; however, the growth recovery is very rapid too. While the maize plant belongs to C4 plants. Its important to know whether there is the same mechanism. The C4 plants have high carbon fixation capacity, because C4 plant leaf has two different chloroplast-containing cell categories: mesophyll and bundle sheath (or Kranz, German for "wreath") cells.

The role of the ABA in controlling growth in reaction to water insufficiency or salinity remains the subject of wide debate, which often leads to contradictory conclusions. At first, the ABA was previously known as a growth inhibitor at long term [16,17]. But other results have reinforced the role of the ABA as a stimulator of leaf growth under stress in maize, by inhibiting the production of ethylene during a water deficiency [18], and by the beneficial effect of the ABA on root hydraulic conductivity (L_p) via the aquaporins activity, which involved in the preservation of a constructive plant water status [19]. The ABA upgrades enough than inhibits plant expansion below stress, and has a major consequence on conservation of adult leaves and might control leaf cell development during a signal transduction pathway. The ABA controls the activity of ion channels that take up crucial solutes for growth, such as potassium (K^+) and amino acids [20]. Before, it was believed that the ABA intervenes slowly [16]. So, according to Munns et al., [15], the root signals had no detectable function over the first little hours! However the recent investigators have indicated at scale time hours that the ABA intervened rapidly in barley and wheat [9,10].

At short term (hours), the rapid osmotic stress permit us to follow some physiological and

biochemical changes better than natural stress. At natural stress, the response mechanisms, and the factors involved in the regulation of growth at long stress, are probably not well distinguished compared to case of rapid stress by osmotic effect (e.g., PEG), where the triggering of these instruments is easier to follow. It's important also to recognize and to track of what was happening during rapid stress in leaves maize as physiological events following an addition of PEG as an osmotic stress, and to make simpler and determine exactly the biophysical changes of response, such as the OA mechanism within hours with biochemical variables. For this purpose, this study attempts to know if they spend the same events in maize (C4) compared with previous studies in wheat and barley (C3) under rapid osmotic and saline medium Respectively, and to know which one who intervenes early, water status or the ABA or together? The synchronization of appearance between them, and their relationship to photosynthesis and stomatal conductivity also are the subject of the current study.

II. Materials and methods

II.1. Growth conditions and rapid plant stress

Maize seeds (*Zea mays* subsp. *mays* 'Dea') were germinated, at room temperature ($23\pm 2^\circ\text{C}$), on a wet filter paper in the dark awaiting the mesophyll reached a length of ~10 mm. Seedlings were placed in tubes (open syringes), and were then grown in continuously aerated standard nutrient solution with the following composition (in mM): 0.5, $\text{CaSO}_4\cdot 2\text{H}_2\text{O}$; 0.8, KNO_3 ; 0.3, KH_2PO_4 ; 0.2, $\text{MgSO}_4\cdot 7\text{H}_2\text{O}$; 0.4, NH_4NO_3 ; 0.02, Fe-EDDHA; 0.008, H_3BO_3 ; 1×10^{-3} , $\text{MnSO}_4\cdot \text{H}_2\text{O}$; 0.1×10^{-3} , $\text{Na}_2\text{MoO}_4\cdot 2\text{H}_2\text{O}$; 0.2×10^{-3} , $\text{ZnSO}_4\cdot 7\text{H}_2\text{O}$; and 0.2×10^{-3} , $\text{CuSO}_4\cdot 5\text{H}_2\text{O}$], was renovated each 4 days. The pH preserved between 5.5 and 5.8. The hydroponic solution was renovated each 3rd to 4th day. Plants were developed in a growth chamber, with a photoperiod of 14 h at photosynthesis photon flux density (PPFD) of 400 $\mu\text{mol}/\text{m}^2/\text{s}$ and day/night temperature of 24/20 $^\circ\text{C}$, as well as a vapor pressure deficiency comprised between 0.8 and 1 kPa. The hydroponic solution was continuously aerated using an air pump. Osmotic stress was induced using PEG₆₀₀₀ at a concentration equivalent to - 0.5 MPa (195.4 g/l), the osmotic potential of PEG was checked using osmometer.

II.2. Physiological measurement

The LER (Leaf Elongation Rate) was measured continuously by Linear Variable Displacement Transducer (LVDT) Used for continuous measurements of growing leaf growth. To determine the elongation zones of leaves, the growing leaf six was disclosed, the relative elongation rate (RER) i.e. elongation rate per tissue length, along the leaf elongation zone (EZ) exhibits a net profile at ~6-8 mm [21], we've demonstrated that by assaying the displacement rates the length of leaf axis by the pricking method under different stresses. While the mature zone (MZ) is the rest of the leaf (Figure 1). The tissues were rapidly cut into little segments put into microtubes containing a little plastic sieve, sealed and rapidly plunged into liquid nitrogen. Samples were defrosted and then spin in the centrifuge (10,000 rpm for 10 min). Next, ~20 μ L of samples were collected and kept in freezer (-20°C) awaiting their analysis (several days). The OP, m mol/kg, was calculated on the expressed sap via vapor pressure osmometer (VAPRO, 5520) following calibration with NaCl of definite osmotic potential. To translate to pressure units it was determined that 40 mmol/kg water = -0.1 MPa. The relative water content (RWC) was ascertained as follows:

$$\text{RWC} = \frac{(\text{FW} - \text{DW})}{(\text{TW} - \text{DW})} \times 100 \quad (\text{Eq. 1})$$

Where: FW, DW, and TW represent the fresh weight, the dry weight, and the turgid weight, respectively.

The osmotic potential (OP) at full plant turgor ($\Psi\pi_{100}$) was calculated as explained by Wilson et al., [22] as follows:

$$\Psi\pi_{100} = [\Psi\pi (\text{RWC} - 10)] / 90 \quad (\text{Eq. 2})$$

The osmotic adjustment was calculated as follows:

$$\Delta\Psi_{100} = \Psi_{S100} - \Psi_{C100} \quad (\text{Eq. 3})$$

The utility of this equation to obtain a suggestion of the component of modifications in OP that is due to solute accretion slightly than cell shrinkage. The turgor was assessed via cell-pressure probe procedure in epidermal cells of the leaf 6 the EZ. The plant was maintained in nutrient solution, taken vertically and then attached by a plastic ribbon on plate plastic bar. To gain admission to the growth zone of the sixth leaf, the coleoptiles, the five outer leaves were estranged. the measurements have been made by cell impalement via a cell pressure probe prepared by a borosilicate microcapillaries (Harvard Apparatus Ltd. Eden bridge, UK) which were sharpened and beveled to acquire a tip diameter of 6 to 8 μ m and replete with silicone oil (Type AS4, Wacker, Munich, Germany). The plant was positioned in front of the pressure probe and a

video microscope system (Leica, Buffalo, NY, USA) was used to adjust the microcapillary in front of the expansion zone of leaf 6. Cell emplacement resulted in the immediate appearance of oil/cell sap meniscus in the capillary. Adjusting the pressure in the microcapillary, the meniscus was brought back to its initial position and the required pressure value read on a digital pressure indicator (DPI 260, Druck, Leicetser, UK).



Figure 1. Experimental device scheme of the test.

The experiment was carried out under ambient conditions: control, hydroponics with the nutrient solution; which was renovated every 4 days. The stress was applied quickly with the substitution of the nutrient solution by the PEG. During both periods, the LER was measured continuously by LVDT, used for continuous measurements of leaf growth, Shown in the figure vertical bars of linear transducers. Thus, in this situation the turgor measurements is then made by the pressure probe, through a small window (hole) cut at the base of the plant, leading to the elongation zone of the sixth leaf the plant.

II.3. Abscisic acid (ABA)

The quantity of (+) *cis-trans*-ABA was predictable from leaf extract, obtained as described above by liquid nitrogen, by means of the procedure described by Quarrie et al., [23] using 3H-ABA (tritiated ABA) specific monoclonal antibodies for (+) -ABA and ammonium sulfate [(NH₄)₂SO₄]. The measurements of ABA from sap (by liquid nitrogen) were very close to those of the classic method [10].

II.4. Stomatal conductance and net photosynthetic rate

The net photosynthetic rate (NPR) was calculated by a unique leaf chamber intended for maize plant and linked to the Gas Analyzers from PP Systems (MA, USA) on four to six individual plants. So, net photosynthetic rate and stomatal conductance on leaf 4 (Expanded), were calculated *via* only leaf chamber, intended for maize and connected to a Gas Analyzer (CIRAS-2; PP Systems, Amesbury, MA, USA), on four to six individual plants.

II.5. Biochemical measurements

The quantity of some carbohydrates e.g., glucose (Glu), fructose (Fru), and sucrose (Suc) were determined following enzymatic transformation to NADH (reducing agent), which is expected at the absorbance of 340 nm ($A_{340\text{ nm}}$). The conversion of glucose was recovered via hexokinase (HK) to yield glucose-6-phosphate (G-6-P) which is changed with the G-6-P dehydrogenase (G6PDH) to yield NADH. The fructose conversion, whereas, necessitate the phospho-glucose-isomerase (PGI) which convert the fructose-6-phosphate (F6P) to the G6P. The sucrose is the primary hydrolysis by the means of β -fructosidase to yield Glu + Fru [24].

The amount of potassium in leaf sap was determined via the flame photometer (SPECTRAA 220fs Varian) following a calibration with the standard solutions of the potassium chloride (KCl). The entire of biochemical sizes were achieved via the expressed sap extracted with liquid nitrogen.

II.6. Statistical analyses

The analysis of variance (ANOVA) unique element between groups was carried out using Microsoft Excel sheet (Version 2007, Microsoft Inc. USA). The distinctions were considered to be statistically significant if $p \leq 0.05$, $p \leq 0.01$, or $p \leq 0.001$. The Student's t-tests were achieved by R package Version 3.0.3 (Vanderbilt University, USA). All experimentations were performed in three independent replicates by referring at the control experiment. The results were expressed with standard Error (mean \pm SE).

III. Results and discussion

III.1. LER and water status changes

The dynamics of leaf elongation previous and following the stress designated the existence of four growth stages (Figure 2): before stress phase 1 rapid drop of LER down to zero after PEG 6000 addition, phase 2, recovery third phase and steady state phase 4. These phases were recorded in real time. Leaf elongation ceased inside 1 to 2 min

following the imposing of PEG 6000. LER recovered rapidly for a period between 3 h. The LER partly recovered, about 70 % of growth rate before PEG 6000 addition.

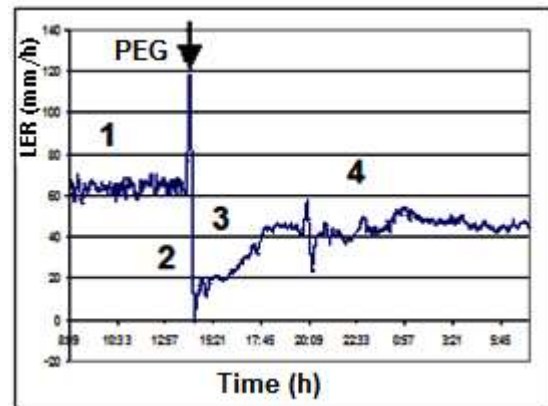


Figure 2. LER kinetics determined by LVDT on the 6th growing leaf of maize, as affected by quick water deficiency imposed by the adding of the PEG 6000 to the hydroponics solution.

The leaf osmotic potential exhibited a gradual decline in both zones (EZ + EM) within 0.5 h after PEG 6000 addition in the 6th growing leaf, its decrease gradually for ~3 h, and then it increased a little value at 4 h subsequent to PEG treatment, which corresponds to resumption of growth. The osmotic potential in increasing leaf six of stressed plants stay considerably lesser at the finish of stress compared to untreated plants (Figure 3).

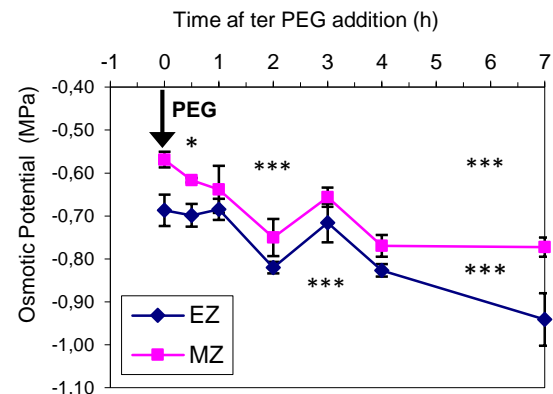


Figure 3. Variations in the OP at full turgor of bulk-tissue in the EZ + MZ of leaf six of maize previous and subsequent to the addition of PEG 6000 to the root medium. Values are the means of 4 repetitions \pm SE (ns, non-significant ; ***, $p \leq 0.001$, versus to time 0 and control 7h).

The osmotic potential at full turgor ($\pi 100$) showed diverse a gradual reduce in the two zones (EZ + MZ) from 0.5 hour following stress. $\pi 100$ continued its decline until the end of stress period. At this moment, the quantity of OA (dissimilarity between stressed period and prior the stress) 0.48

MPa in the EZ, indicates that the OA is generated within 0.5 h (Figure 3). The quantifications of turgor on cells in EZ designated a quick reduce in turgor 30 min after stress. These coincide with the decrease of expansion and water potential, followed by a recuperation stage attainment pre stress values in EZ at end of stress period (Figure 4).

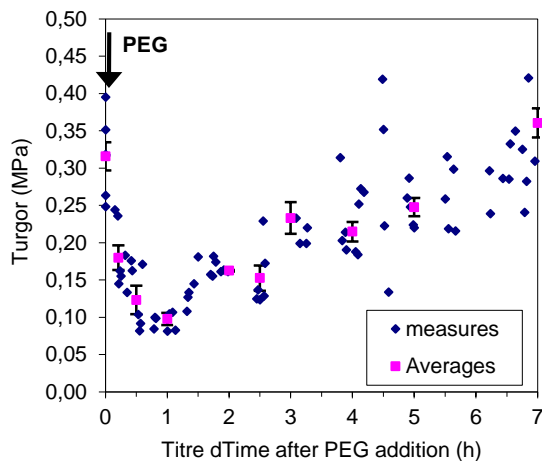


Figure 4. The Variations in turgor pressure measured by pressure investigation in the EZ of leaf six of maize prior and after the addition of the PEG 6000 to the root medium. The data represents the average of at least 6 assays and \pm SE are reported. The vertical bars are SE.

It appears clearly that the LER changes are linked to water states. First, the rapid decrease of water potential is due the sudden disappearance of turgor after PEG 6000 supplementation. Is very obvious the changes of turgor, such a plasmolysis during growth cessation, probably suitable to the loss of water potential rank between the EZ cells and nearby (MZ) tissue and xylem, due to reduce of water potential nutrient solution. Consequently, this reduce will ultimately results in inferior water flow and lead to a withering in the growing tissue of maize similar to wheat [12]. The reduction of gradient water potential is following the application of PEG 6000. This solution adsorbs the water molecules by preventing the entrance of water to the roots. This period was followed by a recovery period of this gradient probably due to rapid decrease of osmotic potential in the EZ, followed by a full in water potential as a consequence of solute accretion, and then the restoration of turgor.

The LER recovered quickly within approximately 3 hours. The origin of the growth recovery is related to the water relations changes and its components. So that, the cells adjusted osmotically into a few minutes, which indicates that the OA is a process generated rapidly by active solute accumulation.

Subsequently, to the water potential gradient returned from the MZ to the EZ cells. Therefore the water flow to elongation cell is returned. So, the turgor recovered rapidly. This led to the cell elongation (Auxesis).

The rapid OA was mentioned in growing tissues of wheat, which gain water and thus turgor recovered immediately [9]. This is the only explanation of LER changes at first few hours. In most previous work, growth at least in time scale hours did not recuperate to pre-stress levels, particularly below conditions where elevated concentrations of salt or PEG are present in the origin medium, at least in the short term. This may be due to the shock applied of PEG or salt in nutrient solution, may beget some changes in cell wall rheology, as mentioned by Muller et al., [21], the hardening of wall and the decrease of the extensibility provokes and leads to an additional turgor against wall hardness. The OA jointly with cell wall extensibility regulates turgor below dehydration [25], where the former is usually more efficient and frequent than the latter.

An accumulation of soluble sugars (Glucose, Fructose and sucrose) were recorded in two zones EZ (Table 1) and MZ (Table 2); The both zones showed the highest accumulation of sugars during the stress period especially sucrose (more than 5 times compared to unstressed plants).

Table 1. Concentrations of Glu, Fru, Suc (m.mol/kg), in the EZ of variety (Dea) of the sixth leaf of maize calculated from the sap of leaf tissue at 0 to 7h of stress, Values are the signify of four replicates \pm SE (ns, non-significant; *, $p \leq 0.05$; **, $p \leq 0.01$; ***, and $p \leq 0.001$, compared to time 0).

Time of stress (h)	Sugars (m mole/kg)		
	Elongation zone (EZ)		
	Glu	Fru	Suc
0	73.25 \pm 4.24	46.32 \pm 3.48	3.18 \pm 1.02
0.5	66.41 \pm 3.31*	36.79 \pm 4.65 *	6.62 \pm 2.75
1	63.85 \pm 2.09*	39.31 \pm 2.00 *	4.33 \pm 0.86
2	73.38 \pm 3.77	43.36 \pm 4.87	11.16 \pm 4.05 **
3	75.32 \pm 4.72	40.27 \pm 5.10	7.65 \pm 3.91 *
4	84.86 \pm 7.21 *	44.99 \pm 9.40	14.71 \pm 4.49 **
7	101.09 \pm 2.29 *	61.29 \pm 7.84 *	17.27 \pm 7.58 ***

Table 2. Concentrations of Glu, Fru, Suc (m.mol/kg), in the MZ of variety (Dea) of the sixth leaf of maize calculated from the sap of leaf tissue at 0 to 7h of stress, Values are the signify of four replicates \pm SE (ns, non-significant; *, $p \leq 0.05$; **, $p \leq 0.01$; ***, and $p \leq 0.001$, compared to time 0).Table 2.

Time of stress (h)	Sugars (m mole/kg)		
	Mature zone (MZ)		
	Glu	Fru	Suc
0	58.79 \pm 7.48	18.76 \pm 2.98	4.52 \pm 2.65
0.5	54.5 \pm 16.72	17.80 \pm 4.20	4.90 \pm 1.90
1	65.2 \pm 5.33	20.07 \pm 2.77	7.52 \pm 0.80 *
2	73.95 \pm 11.18 *	26.19 \pm 2.55 **	12.99 \pm 2.23 ***
3	64.28 \pm 7.04	22.51 \pm 4.58	9.47 \pm 3.05 *
4	71.16 \pm 12.96	30.93 \pm 7.46 *	8.73 \pm 3.78
7	84.18 \pm 15.12 *	34.27 \pm 2.07 ***	13.09 \pm 4.64 *

The increase in K^+ concentration occurred in two tissues (EZ and MZ) through the stress epoch. The involvement of K^+ to osmotic potential was important during and in the end of stress. This contribution to the OA was significantly towards the end of the stress period, between 40 and 80 mM in MZ and EZ, respectively, with an oscillating increase in the case of MZ (Figure 5).

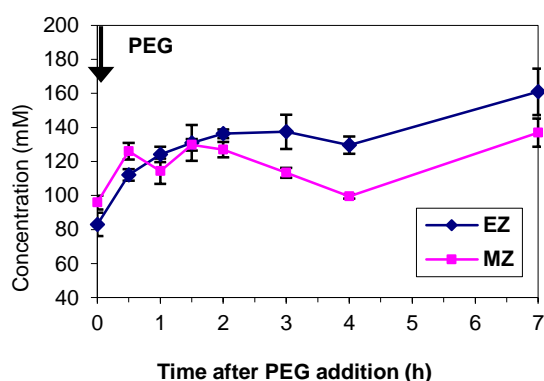


Figure 5. Variations in the K^+ at full turgor of bulk-tissue in the EZ + MZ of leaf six of maize prior and after the adding of the PEG 6000 to the origin medium. Values are the means of four replicates \pm SE. (ns, non-significant; *, $p \leq 0.05$; **, $p \leq 0.01$; ***, and $p \leq 0.001$, compared to time 0 and control 7h). The vertical bars are SE.

III.2. Rapid contribution of solute accumulation

The accumulation of all these solutes has an impact on the EZ through the OA compared with the MZ. The type of accumulated molecules are Sugars and K^+ , which indicates the accumulation so rapid of these solutes (especially sugars) in the OA particularly in the growing leaf tissue, was the result of Cell utilization of photosynthesis products. While photosynthesis *hasn't* been affected much. The enzymatic analysis way used for glucose, glucose and sucrose permitted the assessment of the involvement of these common carbohydrates to osmotic adjustment. Sucrose Fructose and glucose are as photosynthetic products may cross the cell membranes towards EZ, their transport to this zone is clear (Table 1). For these sugars, they seem clear in the EZ is as a sink tissue compared to the MZ, with its remarkable accumulation compared to the MZ of maize. Especially for sucrose, wiche its contribution was more than 5 times at 7 h of stress. The accumulation of sugars was announced first in durum wheat under long-term stress [26], and in several articles until recent years [27]. At short term, it can be considered that the accumulation of total sugars is not a direct consequence of the slowing down of growth or photosynthesis, but rather as an active accumulation related to the osmotic adjustment processes [28]. It also seems here the domination of sucrose as a first product of photosynthesis process.

The important increase of K^+ concentration was registered in both zones, particularly in EZ. The contribution of K^+ to osmotic potential was roughly twice in two zones. Despite the small space occupied by vacuoles in growing cells, these rapid changes in K^+ concentration in both of MZ and EZ could be the result of increased mobilization and fluxes of endogenous solutes (including other ions) from the sap of xylem or phloem across the plasma membrane, including the potassium transporter channels [20].

III.3. Rapid changes of the abscisic acid, stomatal conductance, photosynthesis

The histogram (Figure 6) shows a clear increase in the ABA concentrations in both of the EZ and MZ of the sixth growing leaf of maize in the tissue sap more than 20 times at the seventh hour of stress.

The stomatal conductance of mature transpiring of leaf four was reduced partially after 30 of stress by the PEG 6000 treatment, contrariwise to the transient burst. That was detected at 15 min subsequent to the PEG 6000 application. This transient in stomatal aperture may due to stress sudden shock of PEG. Stomata remained open 75% at the end of the stress period ! In contrast, the photosynthesis was not reduced in along seven hours of stress (Figure 7).

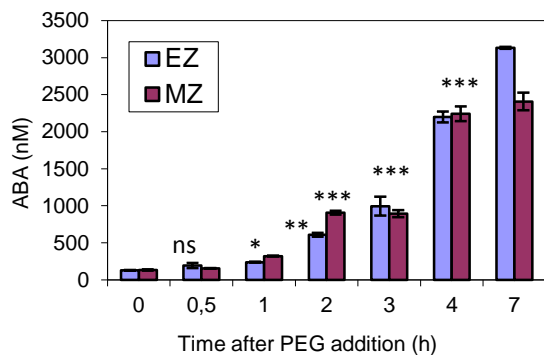


Figure 6. Abscisic acid (ABA) concentration of the EZ leaf six of maize before and following the addition of PEG to the root medium. Results represent means \pm SE of 4-6 samples. (ns, non-significant ; *, $p \leq 0.05$; **, $p \leq 0.01$; ***, and $p \leq 0.001$ compared to time 0 and control 7 h). The vertical bars are SE.

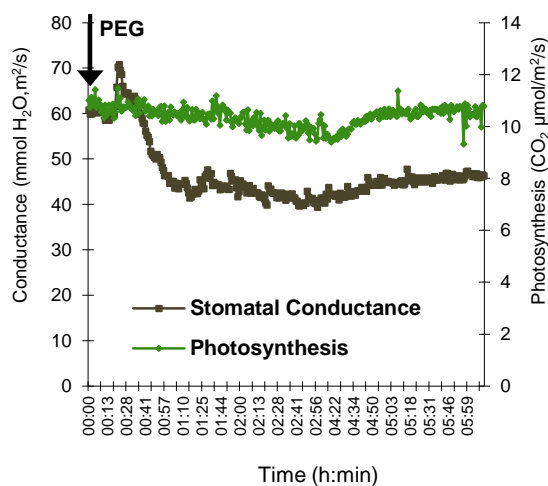


Figure 7. Stomatal conductance and photosynthesis of mature transpiring leaf flour with a diffusion porometer previous and following to the PEG addition, Values are the means of 6 replicates \pm SE.

A rapid accumulation of the ABA was recorded within few hours. This indicates the rapid intervention of this hormone in the regulation of growth in the new water state, as growth control. It appears here that the ABA was synchronized with growth recovery and therefore, was has a positive role in growth during stress.

It became clear the role of the ABA in signalization of several mechanisms ions transport, which can lead to a reduction in osmotic potential and therefore the osmotic adjustment. Its signalization not just on guard cell but includes other cells via ionic flow, causing a diminution in osmotic

potential or Osmotic adjustment, Thus, maintain the water potential gradient and therefore, water entering and in the end, turgid cells. This supports the idea of positive effect of the ABA on growth only during stress.

The venation pattern of xylem and phloem becomes so finely branched that most cells. From the both of veins, water and soluble solutes are drawn into the cells of the leaf of the EZ. The reason of accumulation of their solutes also in the MZ, probably due to the necessary osmotic balances with the EZ, and avoid osmotic imbalance.

In recent years, the role of the ABA has been demonstrated in the activation of the ion flow as a signalization through a series of receptors. Such a PYR/PYL/RCAR-PP2Cs-SnRK2s [29, 30]. A considerable increase of the ABA noted by Qadir et al [31]. in drought tolerant of triticum eastivum, as compared to susceptible genotypes under drought stress condition. So, that, the ABA accretion throughout water stress may frequently function to help preserve suitable shoot size in accordance to root growth, rather than to inhibit growth as is usually believed. As well as plant use other adaptation mechanism like cell shrinkage to decreased water content. Cell membrane stability index and increased proline content of the leaves more remarkably differ in tolerant genotypes as compared to susceptible one. Hashim and Ahmed [32], drew attention to the positive role of exogenous the ABA on growth parameter of wheat. As a conclusion, it is clear that photosynthesis has not decreased and has not been affected during rapid and short water stress, with the relative closure of the stomata; he has continued to play its role in supplying the tissues with the organic assimilates. Continued photosynthesis may indicate that the synthetic compound may contribute to the OA without affecting the growth, which shows the partial aperture of stomata.

IV. Conclusion

It can be concluded from the results of this study LER are partly due to plant water status. The results also suggested that OA was a rapid response and that this adjustment resulted in turgor recovery under a short term water deficit induced by PEG. The OA generated using mainly organic solutes 'sugars' in similarly to studies of wheat [7]. The results of this study indicated the role of turgor recovery in leaf elongation recovery; however, full turgor recovery alone does not necessarily result in full growth recovery which suggested that other factors such as cell wall extensibility changes may also be rapid and play a major role in expansion growth recovery under short term stress. Finally,

ABA content was increased rapidly under rapid water stress by PEG. ABA, which accumulated in the growing tissues, may have facilitated solute transfer and build up in these tissues leading to osmotic adjustment.

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