Essential nutrients and oxidative stress of durum wheat under water stress.
Issaad Ghoziène., Djebar Mohammed-Réda, Berrebbah Houria and Charga Ali
Corresponding author, Email: semsem-dj@hotmail.fr
Laboratory of Cellular Toxicology, University of Annaba, 23000, Annaba, Algeria.

ABSTRACT
Oxidative stress in plants is the subject of numerous reviews of the literature (Apel and Hirt, 2004; Noctor and Foyer, 2005a; Pitzschke et al, 2006; Wormuth et al, 2007; Sbartai et al, 2012.) And of several books (Inze and Montagu, 2001; Smirnoff et al, 2005.).

This work focuses on studying the effects of water stress on the roots of a plant model: durum wheat (Triticum durum) variety GTA. After the germination, the plants were subjected to water stress during (03, 05, 07 and 09 days).

Our results demonstrate abiochemical and metabolic disturbances of the seed subjected to water deficit. The lack of water has caused significant changes in the GTA durum variety.

Keywords: Nutrients; Durum wheat; Water stress; Soluble sugars; Oxidative stress.
Introduction

Plants take minerals from the soil to produce organic compounds. It is recognized that several components are necessary for the normal operation of the biochemical machinery of the plant. Nutrients must be present in an available form for the plants.

To grow a plant needs water, light, oxygen, carbon minerals and also more or less minerals present in the soil. To grow, plants need several essential nutrients in varying amounts and at different steps of their development. Elements required in relatively large amounts (nitrogen, phosphorus, potassium, calcium, magnesium and sulfur) are called macronutrients and the elements required in small quantities are called micronutrients or trace elements. Water stress causes a disruption of the nutrient content in the roots, which leads to the generation of reactive oxygen species (Fenton reaction). The objective of this study is to demonstrate the effect of water stress on the nutrients and in the generation of oxidative stress.

Material and methods

The biological material used in this work is the durum family of Poaceae more specifically it is called Triticum durum Desf. The organ chosen for this study is the root. The samples come from the Algerian office inter cereals (CATO) El Hajar, Annaba, Algeria.

We used the variety of GTA hard.

Performing the test

The tests are done at the Laboratory of Cellular Toxicology of Badji Mokhtar Annaba University and the Laboratory of Fertil annaba complex.

After 4 days of germination, wheat germ suffer from water stress by stopping of watering in addition wheat samples were analyzed at 3, 5, 7 and 9 th day after cessation of watering. A part continues to be watered normally and is considered as a witness.

Germination was carried out at a temperature of 21 ° C day and 17-21 ° C night with artificial lighting from 6 am to 22 H.

Determination of soluble sugars

The determination of sugars is carried out according to the method of Shields and Burnet (1960) which uses the anthrone reagent in a sulfuric environment (150mg anthrone, 75 ml of sulfuric acid and 25ml of distilled water) and a stock solution of glucose at 50 μg / ml. This method comprises extracting where 100mg of sample (MF) were weighed. 3 ml of 80% ethanol was then added, the mixture is left to room temperature for about 48h. Mixture is heated in a water bath at 70 ° C for 30 min, and 2 ml of sample is taken for the assay. The determination of total sugars is performed in an aliquot (100µl). The absorbance is measured with a spectrophotometer (Jenway- 6300) and playback is made at a wavelength of 585nm.

Determination of nitrogen.

Take a test portion of 0.1 to 0.15 g sample of plant crushed and sieved to 500 microns, then put it in aluminum foil and wrap tightly. Next, the sample is injected into the furnace of elemental analyzer LECO nitrogen: the apparatus determines the level of nitrogen contained in the sample by infrared using a TCD detector (detector conductivity thermal). Finally the unit displays the level of nitrogen content in plants.

Determination of average contents of copper, iron, zinc and phosphorus.

Weigh 0.4 g of plant powder crushed at 500 microM, the dried vegetal is crushed with a grinder to reduce the sample to a fineness of less than 500 microM.

Transfer to a Teflon tube numbered and add 5 ml of HNO3 56% 5mL 30 of H2O230% using a dispenser. Close hermetically tubes with screw caps, put them in a tray, then place them in the oven. At the end of the cycle of mineralization, filter the tube contents in volumetric flasks. The reading is performed using a plasma torch spectrophotometer.
Fig 1: Effect of water stress on the average total sugar content in the roots. SSH: No Water Stress; ASH: With Water Stress.

Fig 2: Nitrogen content at root level. SSH: No Water Stress; ASH: With Water Stress.

Fig 3: Copper content at root level. SSH: No Water Stress; ASH: With Water Stress.
Fig 4: Iron content at root level. SSH: No Water Stress; ASH: With Water Stress.

Fig 5: Zinc content at root level. SSH: No Water Stress; ASH: With Water Stress.

Fig 6: Phosphorus content at root level. SSH: No Water Stress; ASH: With Water Stress.
Discussion

The results obtained on root of durum wheat after a water stress indicate a net increase in sugar content of stressed compared to control series and this increases according to the intensity of water stress. These results are consistent with those of some researchers, including Ben Abdellah Ben Salem (1993). Indeed sugars, even if they represent less powerful osmoticums, they are also involved in maintaining the balance of osmotic strength to keep turgor and cytolgal volume as high as possible (Bouzoubaa et al., 2001). They also enable the preservation of organs in membrane integrity and protection of the dry proteins (Darbyshire, 1974). Carbohydrates indispensable for living organisms energy are used immediately as a glucose, or in reserves such as starch, they also have a structural role (reinforcement material of the plant cell wall) such as cellulose, chitin or hyaluranique acid. Enrichment sugars seems protect membranes drying. (Binet, 1989).

The nitrogen content of dry roots has increased as a result of water deficit. In soybean, it was observed that water stress reduced nitrogen metabolism (Calmes et al., 1985). This reduction does not necessarily lead to the decrease in nitrogen content, because it depends on the ratio between carbon and nitrogen metabolisms. The large increase in the nitrogen content could be explained in the roots by a higher sensitivity of the transfer and the synthesis of carbohydrate to stress than the nitrogen compounds (amino acids, proteins, etc.). However, the drought has led to a decline about the phosphorus content of the organs studied. These results indicate that the mass and the biochemical composition of the plant is affected by water deficit but not identically for all compounds. According to Ingestad (1979), the nitrogen stress generally also increases the partition of major nutrients in the roots. This implies, in comparison with well supplied with nitrogen plants, an increase in the proportion of nitrogen absorbed and accumulated in roots (Ruffy et al., 1990) and may also involve an increase in nitrogen remobilization from Part air to the roots (Cruz et al. 2004).

The determination of the average copper content in roots subjected to water stress causes a significant increase from the 3rd day, this recording shows that copper plays an important role in plant defense against stress. Copper is involved in protein synthesis, photosynthesis, respiration and non early chlorophyll degradation (Issaad et al., 2013), plants keep longer green and youthful appearance. Thus, it is necessary for the proper functioning of many enzymes including plastocyanine (PC) or cytochrome oxidase and is involved in the reactions redoxes enzymes and proteins (George, 1996; Barker and Pilbeam 2007).

On the other hand, the results indicate an increase in the average iron content in durum wheat under water deficit. According to INRA, iron is an essential cofactor in biological processes involving electron transfers: photosynthesis, respiration, DNA synthesis and assimilation of nitrogen and sulfur, for example. There is an optimal window of the iron concentration for optimal growth and development. In the presence of water stress, free iron can lead to the production of highly toxic reactive oxygen species, for cell (Issaad et al., 2013). The iron homeostasis must be finely regulated to prevent deficiency, detrimental to the metabolism, and excess ROS generator. In excess, iron is stored in the vacuole and apoplasm, and the synthesis of storage proteins of iron, the ferritin, is induced. Ferritins are ubiquitous proteins which form a hollow sphere capable of storing iron in a form non-toxic and remobilised for metabolic needs.

On the average zinc content analysis of samples revealed a high accumulation of this molecule in stressed roots. According Chaoui and In excess al. (1997), zinc induces oxidative stress type and therefore the activation of enzymes Asada-Halliwell-Foyer cycle due primarily to the ascorbate and glutathione and regulate the redox status of the cell (Noctor and Foyer, 2005; Halliwell, 2006). Stress caused an accumulation of this ion in the plant.

Regarding phosphorus, water stress led to a significant drop from the 3rd day in stressed organs. According Broadley et al. (2007), a high content of Zn leads to a reduction of the synthesis of chlorophyll, degradation of chloroplasts and interference that limit the absorption elements such as phosphorus.

According to the results obtained by Issaad et al., 2013; Noctor and Foyer, 2005; Halliwell, 2006 and the significant increases in mean nutrient contents that we have demonstrated in this article, we can conclude that one of the effects of water stress in roots of durum wheat is the disruption of nutrients that are at the origin of the emergence of oxidative stress.

Références bibliographiques

1. Apel, K. et Hirt, H. 2004. Reactive oxygen species: Metabolism, Oxidative Stress, and Signal Transduction. Annual Review of Plant Biology 55(1): 373-399.
2. Barker, A.V. et Pilbeam, D.J. 2007. Handbook of Plant Nutrition. CRC/Taylor & Francis, Boca Raton, FL. 11-18.
3. Ben Abdellah, N. et Ben Salem, M. 1993. Paramètres morphophysiologiques de sélection pour la résistance à la sécheresse des céréales. INRA, Montpellier. 266 pages.
4. Binet, P. 1989. Métabolisme et adaptation des végétaux supérieurs aux contraintes hydriques, thermiques et salines. Bull. Ecof. T.20.1 : 41-49.
5. Bouzoubaa, Z., El Mourid, M., Karrou, M., et El Ghrous, M. 2001. Manuel d’analyse chimique et biochimique des plantes. INRA. Maroc.
6. Broadley, M. R., White, P. J., Hammond, J. P., Zelko, I., and Lux, A. 2007. Zinc in plants. New Phytologist 173, 677-702.
7. Calmes, J., Viala, G., Gelf, N., Blanchet, R. 1985. Influence d’un déficit hydrique sur trois variétés de soja : effet sur la protéogénèse des graines. Agronomie. 5 : 169-76.
8. Chaoui, A., Mazhoudi, S., Ghorbal, M. H., and El Ferjani, E., 1997. Cadmium and zinc induction of lipid peroxidation and effects on antioxidant enzyme activities in bean (Phaseolus vulgaris L.). *Plant Science* 127: 139-147.
9. Cruz, J.L., Mosquim, P.R., Pelacani, C.R., Araujo, W.L., DaMatta, F.M., 2004. Effects of nitrate nutrition on nitrogen metabolism in cassava. *Biologia Plantarum* 48: 67-72.
10. Darbyshire, B., 1974. The function of carbohydrate units of tree fungal enzymes in their resistance to dehydration. *Plant Physiol.* 54: 717-721.
11. Foyer, C.H. and Noctor, G. 2005. Oxidant and antioxidant signalling in plants: a re-evaluation of the concept of oxidative stress in a physiological context. *Plant, Cell and Environment* 28: 1056-1071.
12. George, E.F. 1996. Plant propagation by tissue culture (Parts 1 and 2). Exegetics Ltd., Edington, UK. 561 pages.
13. Halliwell, B., 2006. Reactive Species and Antioxidants. Redox Biology Is a Fundamental Theme of Aerobic Life. *Plant Physiology* 141(2): 312-322.
14. Ingestad, T., 1979. Nitrogen stress in birch seedlings. *Physiologia Plantarum* 45: 149-157.
15. Issaad, G., Djebar, M.R., Berrebah, H., Rouabhi, R. 2013. ROS and antioxidant system of Triticum durum after water stress. *Annual research and review in biology*. 4(8): 1241-1249, 2014.
16. Pitzschke, A., Forzani, C. and Hirt, H., 2006. Reactive oxygen species signaling in plants. Antioxidant Redox Signal 8(9-10): 175-1764.
17. Rufty, T.W., MacKown, C.T., Volk, R.J., 1990. Alterations in nitrogen assimilation and partitioning in nitrogen-stressed plants. *Physiologia Plantarum* 79: 85-95.
18. Sbartai, H., Djebar, M.R., Sbartai, I. et Berrabah, H. 2012. Bioaccumulation du Cd et du Zn chez les plants de tomates (Lycopersicon esculentum L.). Bioaccumulation of cadmium and zinc in tomato (Lycopersicon esculentum L.). *Biologie et Pathologie végétales*. 585-593.
19. Schields, R. and Burnett, W. 1960. Determination of protein bound carbohydrate in serum by a modified anthrone method. *Anal. Chem.* 32: 885-886.
20. Smirnoff, N., Foyer, C., Dietz, K., Mittler, R., Feierabend, J., Grace, S., Desikan, R., Jones, M., Vreeburg, R., Logan, B. and Jaspers, P. 2005. Antioxidants and Reactive Oxygen Species in Plants, Blackwell publishing. *Sciences (CMLS)* 57(5): 779-795.
21. Van Breusegem, F., Vranova, E. Dat, J.F. and Inze, D. 2001. The role of active oxygen species in plant signal transduction. *Plant Science* 161(3): 405-414.
22. Wormuth, D., Heiber, I., Shaikali, J., Kandlbinder, A., Baier, M. and Dietz, K.J. 2007. Redox regulation and antioxidant defense in Arabidopsis leaves viewed from a systems biology perspective. *Journal of Biotechnology* 129(2): 229-248.