

Original Research

Effect of salinity on the composition of total lipids in the two varieties of banana (*Musa acuminata* L.) dwarf and small dwarf in Morocco

Authors:

Meriem BELFAKIH¹,
Mohammed IBRIZ¹
Abdelmjid ZOUAHRI² and
Said HILALI¹

Institution:

1. Laboratoire de Génétique
et Biométrie, Faculté des
sciences, Université Ibn
Tofail, BP: 133. 14 000.
Kenitra.

2. Unité de Recherche «
Environnement et
Conservation des Ressources
Naturelles », INRA, CRRA
de Rabat, BP6356 Rabat
Instituts, Maroc.

Corresponding author:

Meriem BELFAKIH

ABSTRACT:

Objectives: The present study aims to determine the effect of salt stress on the total lipid composition for two varieties of banana (*Musa acuminata*) viz., great dwarf and small dwarf variety. The presence of different concentrations viz., triglycerides and diglycerides did not influenced the increasing salt concentration in the medium. However, monoglycerides and free fatty acids were more affected by the effect of salinity.

Regarding the large dwarf variety, the absence of triglycerides noted, especially in the stressed plants and also in the control plants. In the light of our results we saw that the membrane lipids in the vast dwarf were less affected by salinity compared to the small dwarf

Email Id:

m_belfakih2000@yahoo.fr

Article Citation:

Meriem BELFAKIH, Mohammed IBRIZ Abdelmjid ZOUAHRI and Said HILALI
Effect of salinity on the composition of total lipids in the two varieties of banana (*Musa acuminata* L.) dwarf and small dwarf in Morocco
Journal of Research in Biology (2016) 6(1): 1952-1958

Dates:

Received: 07 Oct 2015 Accepted: 02 Dec 2015 Published: 12 Feb 2016

Web Address:

[http://jresearchbiology.com/
documents/RA0578.pdf](http://jresearchbiology.com/documents/RA0578.pdf)

This article is governed by the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which gives permission for unrestricted use, non-commercial, distribution and reproduction in all medium, provided the original work is properly cited.

INTRODUCTION

Salt stress is the main environmental factor that causes serious damage to plants and thus food production. When the plants are exposed to salinity, they face two obstacles; external low water potential and high internal concentration of toxic ions (Verslues *et al.*, 2006; Hirayama and Mihara, 1987). These barriers generally lead to the disturbance of various enzymatic processes; changes in the composition of membrane lipids and inhibition of growth (Brown and Smith, 1989; Elkahoui *et al.*, 2004; Munns, 2005; Sui *et al.*, 2010; Shu *et al.*, 2012). Some authors suggested that lipids are involved in the protection of plants against salt stress (Khamutov *et al.*, 1990. Ritter and Yopp, 1993) and numerous studies have shown that stress can induce changes in the lipids of cell membranes (Huflejt *et al.*, 1990; Elkahoui *et al.*, 2004; Sui *et al.*, 2010). Indeed, when the photosynthetic organisms are exposed to salt stress, the fatty acids of the cell membrane are denatured because the membranes of plant cells are dynamic in their behavior with a lipid composition that changes during variations in the environment. These variations may affect the fluidity and integrity of the membrane and change the activity of the proteins due to the alteration of lipid composition (Quartacci *et al.*, 2000). For many plants, changes in the lipid composition as galactolipids, phospholipids and sulpholipids are observed under the salinity conditions (Hamed *et al.*, 2005).

Indeed, the cell membrane is susceptible to numerous physiological and biochemical activities and plants easily change the lipid composition of their membrane in response to the environmental stress (Harwood and Russell, 1984). Thus, the lipid peroxidation of the cell membrane is the symptom related to oxidative damage caused to the cell during stress and it is therefore used as an indicator of the effects caused to the cell membrane under conditions of saline stress (Campo *et al.*, 2014). The aim of this experiment is to demonstrate the effect of salinity on the

lipid composition in the two varieties of banana at *in vivo* conditions.

MATERIALS AND METHODS

The objective of this work is the demonstration of the effect of salt stress on the composition of total lipids of two banana varieties; the tall and the dwarf.

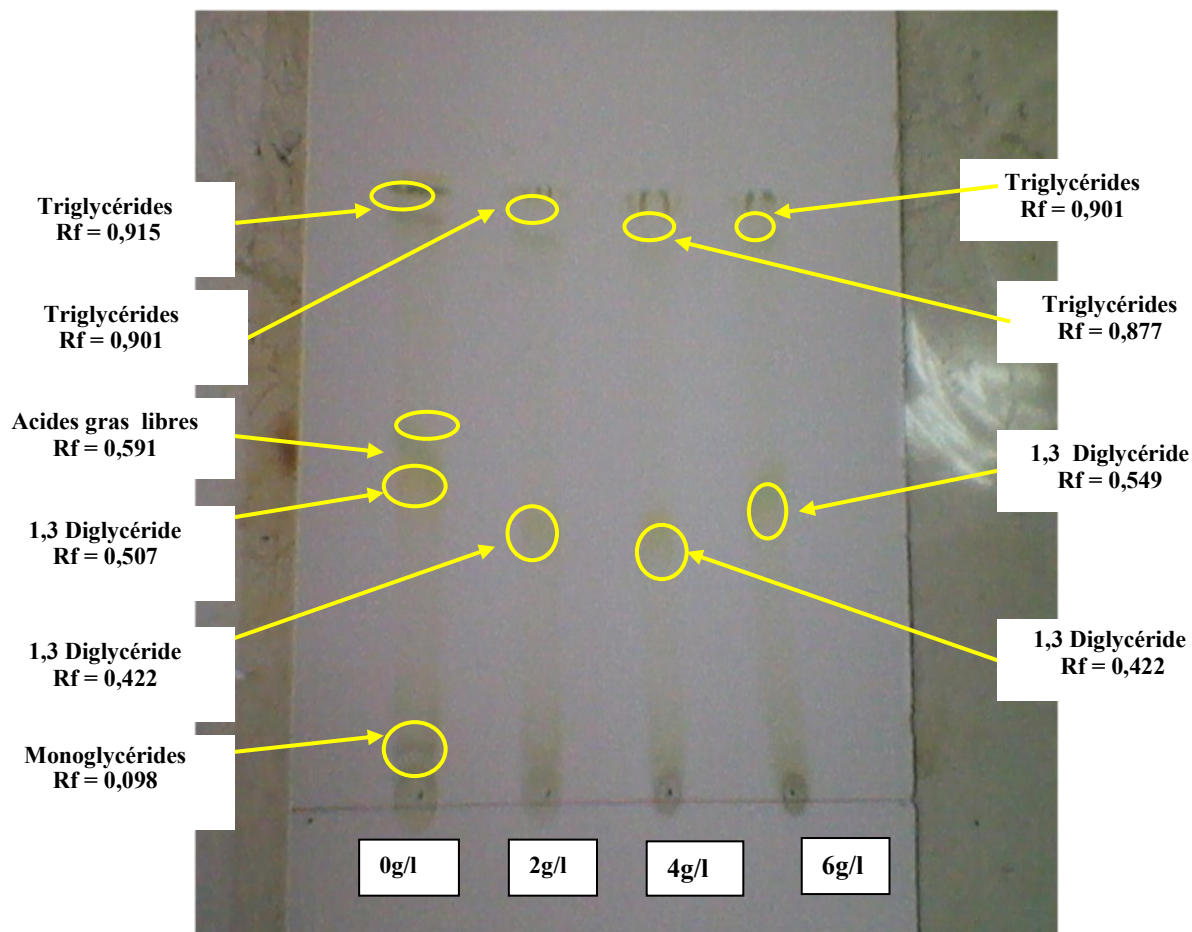
Plant Material

The study of the effect of salinity was carried out on the two varieties of banana (*Musa acuminata L*), the "tall" and "dwarf" from *in vitro* culture. After the vegetative propagation of banana explant, and after rooting stage, seedlings (plantlets) from the laboratory were transferred for acclimatization to be greenhouse under black polyethylene bags of 20 cm long and 12 cm in diameter which contains peat. The plants were kept moist by regular watering at the rate of one out of two. The tested varieties were treated with four levels of NaCl (0, 2, 4 and 6 gl / l). The adopted experimental type is a factorial of two factors (variety, salinity) ets three repetitions.

Total lipid extraction from whole plant

The lipid extraction was made from the leaves according to the method of Bligh and Dyer (1959) using the mixture: chloroform / methanol / brine ; 2/2/2 ; v / v / v. The banana leaves were put in boiling water for 3-5 minutes to stop any enzymatic reaction. Tissues were crushed using a porcelain mortar initially with chloroform and added methanol (2/1, V / V). Another volume of chloroform is then added followed by two volumes of brine (1% NaCl) comprising water binding. Each addition was followed by mortar grinding. Centrifugation at 2000 rpm / min for 15 min yielded two phases separated by a protein coat.

The lower phase (chloroform) containing the lipids was removed and evaporated to dryness under a stream of nitrogen or under vacuum at 50°C using a rotary evaporator. Total lipids were taken up in a toluene – ethanol/ (4/1 ; v / v) or chloroform, and stored at - 20°



C (Bligh and Dyer, 1959).

Chromatographic separation of the lipid

Different lipid phases were separated from the total extract by one-dimensional thin layer chromatography. The lipid extract was deposited under nitrogen on a silica gel plate with 0.20mm thickness (Silicagel G 60, Marek). The deposition consists of 50 of the lipid phase recovered from the ethanol-toluene phase using a Hamilton syringe.

The one-dimensional migration was performed in a hermetically closed vessel containing chloroform, acetone, methanol, acetic acid and distilled water in the proportions: of 50/20/10/10/5 volumes (Lepage, 1967).

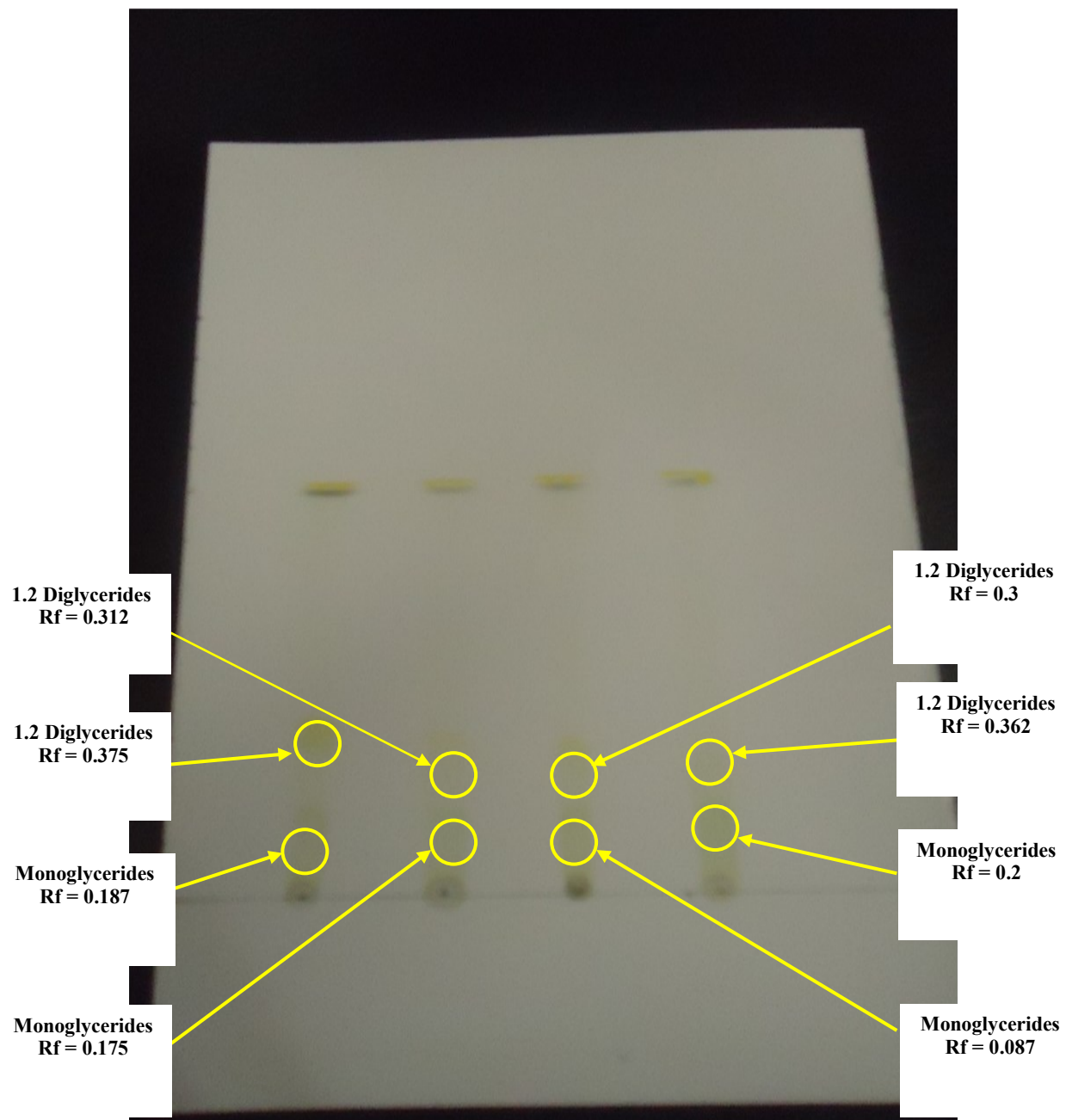
After migration (As the migration front reached 1 cm from the top of the plate) the plates were removed and then dried. The various classes of lipids were disclosed by iodine vapor and observed under ultraviolet light. Iodine binded to the double bonds and stained the

lipid tan. It was easily removed in the air or by gently heating the plate above a water bath under the stream of nitrogen. For each lipid class, Rf (reference edge) was calculated. Rf is equal to the distance traveled by the lipid divided by the distance traveled by the solvent (mobile phase).

RESULTS

The analysis of lipids by the method of Bligh and Dyer (1959) from the leaves of control and stressed banana varieties showed that the chromatographic profile of the total lipids in the absence of the salt is not the same for both the varieties of banana. The topography of the chromatograms (photo 3.4) showed the presence of the main classes of components of lipid membranes.

For the dwarf variety (picture 7), the presence and concentrations of triglycerides and diglycerides (1,3) were not influenced by increasing the salt concentration



in the medium. However, monoglycerides and free fatty acids had mostly been affected by the effect of salinity as being present in the control plants; they were removed from leaves at all salt concentrations. Thus, salinity had no effect on triglycerides having an R_f between 0.877 and 0.915, when we note the occurrence of 1, 3 diglycerides with $R_f = 0.422$ for salt concentrations of 2 and 4 g / l. It should be noted that the triglycerides with (1, 3) of R_f 0.507 and 0.549 respectively were detected in the control and in the concentration of 6 g / l.

Concerning the great dwarf variety (photo 4), we note the absence of triglycerides in stressed plants and also in the control plants. In addition, the presence of 1,2 diglycerides and monoglycerides and their migration heights are identical for both control for all salt concentrations.

DISCUSSION

The majority of work in the literature about the effect of salt stress on lipids, were particularly interested

in is the peroxidation and desaturation of fatty acids (Berberich *et al.*, 1998; Mikami and Murata, 2003) and very little work was on the evolution of, di- and monoglycerides in plants under salt stress. Based on the migration of membrane lipids in the tall (photo 8), it is seen that they were less affected by salinity in relative to the dwarf. This could be explained by the fact that the lipids of the genotype of the tall are relatively less sensitive to salt stress. Zenoff *et al.*, (1994) reported the same point on two soybean genotypes. These authors suggested that salt stress differently affects lipid metabolism at the genotype level of the same soybean species. Because the phospholipid content was not affected by increasing the salinity while that of free sterols and triglycerides increases in two soybean genotypes.

Among the dwarf. Plants triglycerides persistence was noted in both the control plants and stressed plants but with a slight difference in their Rf. This showed that, probably lipids in their triglyceride form have not been influenced by salt. Indeed, high contents of phosphorus and glycolipids in case of stress indicates that the plant probably synthesized membrane lipids in response to the adverse effects of stress (Munshi *et al.*, 1986). These authors reported that the relative content of glycolipids, triglycerides and free fatty acids in *Raya* remain constant while at *Toria*, phospholipids and other lipid compounds remained more or less constant. Similarly, Walker *et al.*, (1984) postponed the starch content and triglycerides were seen similar between the roots of control plants of *Citrus reticulata* and those exposed to the salt stress of 100 mM. In the plant *Chloris Gayana* these accumulated a high concentration of soluble sugars and body fat, compared to controls ; when the plant was subjected to a salt stress of 0.2 mol / liter (Córdoba *et al.*, 2001).

By cons, many worked agrees that glycerolipids were reduced on giving salt stress (Smaoui and Cherif, 2000). These results are not surprising as they reflect the

considerable decrease in the number of chloroplasts in certain plants like *Arabidopsis* (Deeken *et al.*, 2006) and sunflower (Gee *et al.*, 1967). In fact, phospholipids and glycolipids which are used as nonproteinaceous primary compounds of plant membranes, while triglycerides (fats and oils) are efficient forms carbon storage at different stages of development of the plant and particularly in grain (Taiz and Zeiger, 1991). Thus, almost 70% of the total proteins and 80% of the total lipids of plant cells are found in chloroplasts and any change in the membrane of chloroplasts are reflected by corresponding changes in the content and quality of the total lipids in leaves (Harwood and Russell, 1984).

The maintenance or increase of membrane lipids is a response to abiotic stress. Because this reaction results in a significant accumulation of lipid droplets in the cytoplasm of cells of leaves in sorghum subjected to salt stress (Arafat *et al.*, 2009). This accumulation, which disappears when the stress conditions cease (Poljakoff Poljakoff-Mayber, 1981) was also observed in cells exposed to saline treatment and other tissues exposed to water stress (Rahman *et al.*, 2000) . However, they have not been found in the cytoplasm of control leaves suggesting a loss of lipids of the membrane to the cytoplasm under stress (Olmos and Hellin, 1996). The accumulation of lipid droplets is then regarded as a reserve of energy that is used by cells to satisfy a growing demand for metabolic energy to tolerate salinity (Rahman *et al.*, 2000). In fact, some studies have suggested that the lipids are probably involved in the protection of the plant against saline stress (Huflejt *et al.*, 1990; Khamutov *et al.*, 1990; Ritter and Yopp, 1993).

CONCLUSION

The application of the salt has caused changes in the fluidity of the membrane, which is largely controlled by lipids and proteins Generally, lipid proportions different from one variety to the next. In the dwarf variety, the presence and concentrations of triglycerides

and diglycerides ie., (1, 3) were not influenced by increasing the salt concentration in the medium. However, monoglycerides and free fatty acids have been mostly affected by the effect of salinity.

Regarding the tall variety, one could notice the absence of triglycerides in the stressed plants and also in the control plant. In light of our results, we see that the membrane lipids in the tall were less affected by salinity relative to the dwarf. This can be explained by the fact that the lipids of the genotype of the tall are relatively less sensitive to salt stress.

REFERENCES

- Arafa AA, Khafagy MA and El-Banna MF. (2009).** The effect of glycinebetaine or ascorbic acid on grain germination and leaf structure of sorghum plants grown under salinity stress. *Australian Journal of Crop Science*, 3(5):294-304.
- Ben Hamed K, Ben Youssef N, Ranieri A, Zarrouk M and Abdelly C. (2005).** Changes in content and fatty acid profiles of total lipids and sulfolipides in the halophyte *Crithmum maritimum* under salt stress. *Journal of Plant Physiology*, 162(5):599–602.
- Berberich T, Harada M, Sugawara K, Kodama H, Iba K and Kusano T. (1998).** Two maize genes encoding omega-3 fatty acid desaturase and their differential expression to temperature. *Plant molecular biology*, 36(2):297–306.
- Bligh EG and Dyer WJ. (1959).** A rapid method for total lipid extraction and purification. *Canadian Journal of Biochemistry and Physiology*, 37(8):911-917.
- Brown DJ and Dupont FM. (1989).** Lipid composition of plasma membranes and endomembranes prepared from roots of barley (*Hordeum vulgare* L.): effects of salt. *Plant physiology*, 90(3):955–961.
- Campo S, Baldrich P, Messegue J, Lalanne E, Coca M and San Segundo B. (2014).** Overexpression of a Calcium-Dependent Protein Kinase Confers Salt and Drought Tolerance in Rice by Preventing membrane Lipid Peroxidation. *Plant physiology*, 165(2):688-704.
- Córdoba A, García Seffino L, Moreno H, Arias C, Grunberg K, Zenoff A and Taleisnik E. (2001).** Characterization of the effect of high salinity on roots of *Chloris gayana* Kunth: carbohydrate and lipid accumulation and growth. *Grass and Forage Science*. 56 (2):162–168
- Deeken R, Engelmann JC, Efetova M, Czirjak T, Müller T, Kaiser WM, Tietz O, Krischke M, Mueller MJ and Palme, K, Dandekar T and Hedrich R. (2006).** An integrated view of gene expression and solute profiles of *Arabidopsis* tumors: a genome-wide approach. *Plant Cell*, 18(12):3617–3634.
- Elkahoui S, Smaoui A, Zarrouk M, Ghrir R and Limam F. (2004).** Salt-induced lipid changes in *Catharanthus roseus* cultured cell suspensions. *Phytochemistry*, 65(13):1911–1917.
- Gee MM, Sun CN and Dwyer JD. (1967).** An electron microscope study of sunflower crown gall tumor. *Protoplasma*, 64(2):195–200.
- Harwood JL and Russell NJ. (1984).** Lipids in plants and microbes. George Allen and Unwin, London.
- Hirayama O and Mihara M. (1987).** Characterization of membrane lipids of higher plants different in salt-tolerance. *Agricultural and biological chemistry*, 51 (12):3215–3221.
- Huflejt ME, Tremolieres A, Pineau B, Lang JK, Hatheway J and Packer L. (1990).** Changes in membrane lipid composition during saline growth of the fresh water *Cyanobacterium Synechococcus* 6311. *Plant physiology*, 94(4):1512–1521.

- Khamutov G, Fry IV, Huflejt ME and Packer L. (1990).** Membrane lipid composition, fluidity, and surface charge changes in response to growth of the freshwater Cyanobacterium *Synechococcus* 6311 under high salinity. *Archives of Biochemistry and Biophysics*, 277(2):263–267.
- Lepage M. (1967).** Identification and composition of turnip root lipids. *Lipids*, 2(3):244-250.
- Mikami K and Murata N. (2003).** Membrane fluidity and the perception of environmental signals in cyanobacteria and plants. *Progress in Lipid Research*, 42(6):527–543.
- Munns R. (2005).** Genes and salt tolerance: bringing them together. *The New phytologist*, 167(3):645–663.
- Munshi SK, Bhatia N, Dhillon KS and Sukhija PS. (1986).** Effect of moisture and salt stress on oil filling in *Brassica* Seeds. *Proc. Indian nain. Sci. Acad.* 52(6):755-759.
- Olmos E and Hellin E. (1996).** Cellular adaptation from a salt-tolerant cell line of *Pisum sativum*. *Journal of plant physiology*, 148(6):727-734.
- Poljakoff-Mayber A. (1981).** Ultrastructural consequence of drought. In L.G. Paleg and D. Aspinall eds., the Physiology and Biochemistry of Drought Resistance in Plants. Academic Press, New York. 389-403.
- Quartacci MF, Pinzino C, Sgherri CLM, Dalla Vecchia F and Navari-Izzo F. (2000).** Growth in excess copper induces changes in the lipid composition and fluidity of PSII-enriched membranes in wheat. *Physiol Physiologia Plantarum*, 108 (1):87–93.
- Rahman MS, Matsumuro T, Miyake H and Takeoka Y. (2000).** Salinity-induced ultrastructural alterations in leaf cells of rice (*Oryza sativa* L.). *Plant Production Science*, 3 (4):422-429.
- Ritter D and Yopp JH. (1993).** Plasmam Membrane lipid composition of the halophilic cyanobacterium *Aphanothece halophytica*. *Archives of Microbiology*, 159(5):435–439.
- Shu S, Guo SR, Sun J and Yuan LY. (2012).** Effects of salt stress on the structure and function of the photosynthetic apparatus in *Cucumis sativus* and its protection by exogenous putrescine. *Physiologia plantarum*, 146(3):285–296.
- Smaoui A and Chérif A. (2000).** Changes in molecular species of triacylglycerols in developing cotton seeds under salt stress. *Biochemical Society transactions*, 28(6):902-905.
- Sui N, Li M, Li K, Son J and Wang BS. (2010).** Increase in unsaturated fatty acids in membrane lipids of Suaeda salsa L. enhances protection of photosystem II under high salinity. *Photosynthetica* 48:623–629
- Taiz L and Zeiger E. (1991).** *Plant Physiology*. The Benjamin/Cummings. Publishing Company, California, 265-291.
- Verslues PE, Agarwal M, Katiyar-Agarwal S, Zhu J and Zhu JK. (2006).** Methods and concepts in quantifying resistance to drought, salt and freezing, abiotic stresses that affect plant water status. *The Plant journal: for cell and molecular biology*, 45(4):523–539
- Walker RR, Sedgley M, Blesing MA and Douglas TJ. (1984).** Anatomy, Ultrastructure and Assimilate Concentrations of Roots of Citrus Genotypes Differing in Ability for Salt Exclusion. *Journal of Experimental Botany*, 35(10):1481-1494.
- Zenoff AM, Hilal M, Galo M and Moreno H. (1994).** Changes in Roots Lipid Composition and Inhibition of the Extrusion of Protons during Salt Stress in Two Genotypes of Soybean Resistant or Susceptible to Stress. Varietal Differences. *Plant and Cell Physiology*, 35(5):729-735.

Submit your articles online at www.jresearchbiology.com

Advantages

- **Easy online submission**
- **Complete Peer review**
- **Affordable Charges**
- **Quick processing**
- **Extensive indexing**
- **You retain your copyright**

submit@jresearchbiology.com

www.jresearchbiology.com/Submit.php