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Responses of Biochemical Traits in un-or Mycorrhizal *T. vogelii* Hook F. Under Drought Stress at an Early Growth Phase

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Abstract

This work was carried out on un-or mycorrhizal *Tephrosia vogelii*, at an early growth phase, to study responses of biochemical traits under drought stress. A completely randomized blocks design was used and the various levels of water stress were adjusted daily by moisten each pot with the amount of water loss by transpiration. The various parameters measured were: total amino acid content and proline; total soluble sugar content; total soluble proteins content and acid phosphatases specific activity. The results obtained show that, under water stress conditions, mycorhization significantly improves all the parameters studied. The improvement of water status, nutritional status and water stress tolerance by mycorhization, provided to the plant a normal functioning and better yield where water is scare. On the basis of these results, we can advise mycorrhizal biofertilizers like sure means to fight against drought and to increase agricultural yield on grounds victims of the dryness.

Key words: Biochemical traits**,** mycorrhizal, *Tephrosia vogelii*, drought stress, early growth phase

1. Introduction

Leguminous plants are characterized by their fruit called pod or vegetable (Westphal and al., 1985).Thanks to their autotrophy for nitrogen they play the role of pioneer's plants on poor and strongly degraded lands (Azzouz, 2009; Akédrin and al., 2010).They improve soil fertility and would play increasing roles in natural ecosystems, in agroecosystems, in sylviculture and agroforestry (Hindumathi and Reddy, 2012; Gao and al., 2012). Several work put forward the importance of leguminous plants in human and animal nutrition, in consolidation of food safety, in economic, in medicine, in decorative point of view (Duke, 1981; Visitpanich and al., 1985a; Vance and al., 2000; Westphal and al., 1985; Makoshi and Arowolo, 2011; El Sohaimy, 2012). Since work of National Academy of Sciences (NAS) in 1979 published in the book title "tropical legumes: resources for the future ", research has been intensified on leguminous plants.

Leguminous plants have to play foreground roles: in mitigation of the effects of climatic change (Jensen and al., 2012); like raw material for biofuels and biorefineries (Jensen and al., 2012). Considering huge services provided to ecosystems, their skill to reduce green-house gas emission, to lower the use of fossil fuel, to accelerate the speed of carbon sequestration in grounds and feedstock for biorefineries, leguminous plants could be regarded as significant components in the development of the futures agroecosystems (Jensen and al., 2012).

For several decades, efforts have been made everywhere in the world to highlight leguminous plants, for the optimal exploitation of their huge potential; this work concerns *T. vogelii*, shrubby leguminous plant whose importance is clarified in several former works (Makoshi and Arowolo, 2011; Orowa and al., 2009; Tsoata and al., 2015). It is in this context that UNO in its sixty-eighth General Assembly proclaimed 2016 international year of leguminous plants (Anonyme, 2013). However leguminous plants culture is confronted with several obstacles.

Among constraints of leguminous plants production water stress is most complex and devastator on a total scale (Pennisi, 2008).There is water stress when water requirements of plant are higher than the quantities available for roots (Gaye and Bloc, 1992).The frequency of this stress should increase as consequence of climatic change (Ceccarelli, 2010). Water is a determining element for plants growth, particularly in areas where this resource is scare; withholding of water can impair crops yield up to 50% even more (UNDP BCPR, 2003; Gholamhoseini and al., 2013). It constitutes a serious threat for everlasting food safety in actual conditions and will be more than a danger in next year because forecast provide that climatic changes will cause frequent and high temperatures, as well as drought conditions in many world areas (Ficher and Schär, 2010 ;Smith, 2011).

In Cameroon where agriculture is primarily rain feed, water stress leads to falls in agricultural production everywhere, especially during small rainy season where rains can stop suddenly or become sporadic. Roots symbiosis realized with arbuscular mycorrhizal fungi (AMF) can in general protect plants and leguminous plants in particular, against harmful effects of water stress (Gholamhoseini and al., 2013). Research relating to the physiology of leguminous plants under drought stress are scare, furthermore those bearing on their first stages of development. The goal of this work is to evaluate the effect of water stress on some biochemical parameters of un-or mycorrhizal *T. vogelii* at an early stage.

2. Materials and Methods

Healthy seeds of *T. vogelii* are disinfected, germinated in darkness; seedlings are **sowed** on sterilized substrate inoculated or not with AMF and various levels of stresses applied immediately and during 31 days. A completely randomized blocks design is used and various levels of water stress were adjusted daily by moisten each pot with the amount of water loss by transpiration. The different measured parameters are: total amino acid content and proline; total soluble sugar content; total soluble proteins content and acid phosphatases specific activity using Tsoata and al. (2015) method.

Data analysis

Data analysis is carried out with software SPSS 18.0 for ANOVA and correlations between various measured parameters. The averages obtained are compared according to water stress level for each treatment (mycorrhizal or not) by DUNCAN's test; differences are declared significant at $p < 0.05\%$. The results, presented in the form of tables and histograms are produced using Microsoft Excel 2007 software.

3. Results

3.1 Total amino acids

The TAA (Fig. 1A) produce by unmycorrhizal control are significantly ($p < 0.05$) lower than that of mycorrhizal ones; it increase by 31 % for mycorrhizal blank. Foliar TAA increase significantly by 18 and 38 % for soft and average stress respectively; but drop by 21 % for severe stress for unmycorrhizal seedlings. For mycorrhizal seedlings this parameter records a non significant lessen of 3 % for soft stress and a significant increase of 15 and 9 % respectively for average and severe stress. Mycorhization increases the TAA significantly, but this increment is weak for soft and average stress, very significant 81 % for severe stress, nearly 10 times compared to soft and average stress.

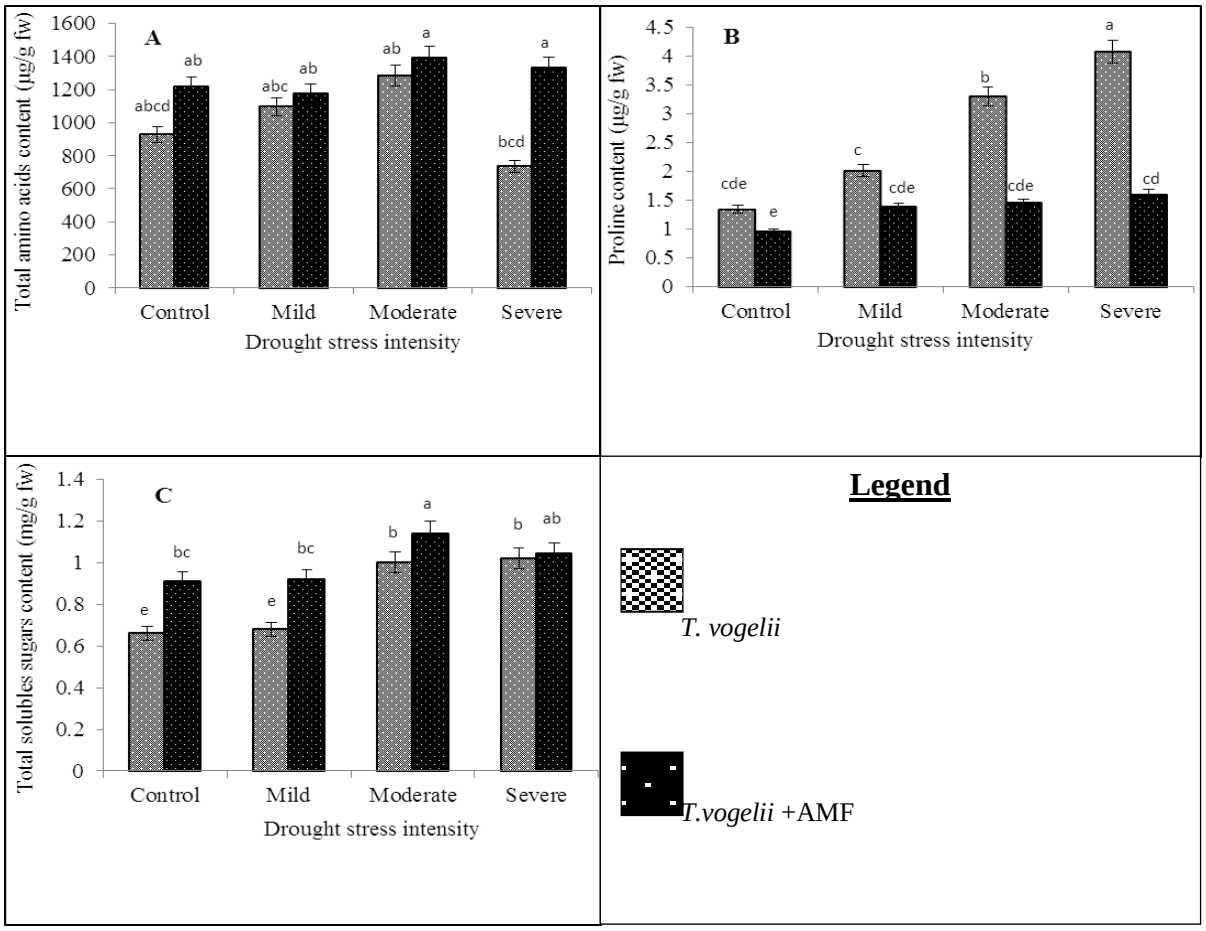

Figure1. Total amino acids (A); proline (B) ; solubles sugars (C) content in mycorrhizal and non-mycorrhizal *T. vogelii* plants under severe, moderate ; mild and no drought stress (control) conditions.

3.2 Proline

Proline produced by unmycorrhizal control seedlings (Fig. 1B) are high compared to that of mycorrhizal ones; nearly 29 % reduction for mycorrhizal ones. When water stress level increases for unmycorrhizal seedlings, proline content raise; the increase is 68, 145, 204 % for soft, average and severe stress respectively. For mycorrhizal seedlings, the increase in proline content is weak compared to unmycorrhizal ones and is 45, 53, 68 % for soft,

average and severe water stress respectively. The mycorhization stimulates the production of proline in mycorrhizal seedlings compared to unmycorrhizal control, this stimulation declines when the water stress level rises; it is: 41, 19, 15, 8 % for blank, soft, average and severe stress.

3.3 Total soluble sugars (TSS)

The **TSS** (Fig. 1C) produced by unmycorrhizal control seedlings is weak compared to that of mycorrhizal ones: 38 % of increase for mycorrhizal seedlings. When water stress level increases, the **TSS** content increases significantly for unmycorrhizal *T. vogelii*; for about: 3, 52, 55 % respectively for soft, average and severe stress. This increase is relatively weak for mycorrhizal *T. vogelii* where highest increase is observed for average stress and weakest for control; the increment is: 1, 14, and 25 % for soft, severe and average stress respectively. Mycorhization stimulates production of total soluble sugars, but the increment lessens when level of stress rise; increment is: 38, 35, 14, 2 % for control, soft, average and severe stress respectively.

3.4 Total soluble proteins (TSP)

The TSP amount produces by unwater stressed seedlings of *T. vogelii* (Fig. 2A) is always significantly weak for unmycorrhizal seedlings compared to mycorrhizal ones; the observed increase is 35 %. For all seedlings an increment of total soluble protein is noticed, it rises with water stress level, for soft and average stress: 16, 24 % and 6, 11 % for unmycorrhizal and mycorrhizal seedlings respectively. For severe stress there is inhibition of total soluble proteins production whatever the treatment; this inhibition is strong 10 % for mycorrhizal seedlings and weak 0.5 % for unmycorrhizal ones. Mycorhization stimulates production of total soluble protein for all stress levels; increment being high for control compared to stressed seedlings: 35, 23, 21, and 23 % for control, soft, average and severe stress respectively.

3.5 Specific activity Acid phosphatases ($SAAP_{ases}$)

It is significantly ($p < 0.05$) weak for unmycorrhizal control seedlings compared to mycorrhizal ones (Fig. 2B), where it increases by 40 %. This activity rise with water stress level whatever the treatment: 96, 113, 367 % and 455, 881, 318 % for soft, average and severe stress respectively for unmycorrhizal and mycorrhizal seedlings. However for severe stress increase is weak compared to the other levels of stress. Mycorhization stimulates this activity for mycorrhizal seedlings comparatively with unmycorrhizal ones. This $SAAP_{ases}$ is high for mycorrhizal control: 40 %; rise with the level of stress: 4, 23, 25 % for soft, average and severe stress respectively.

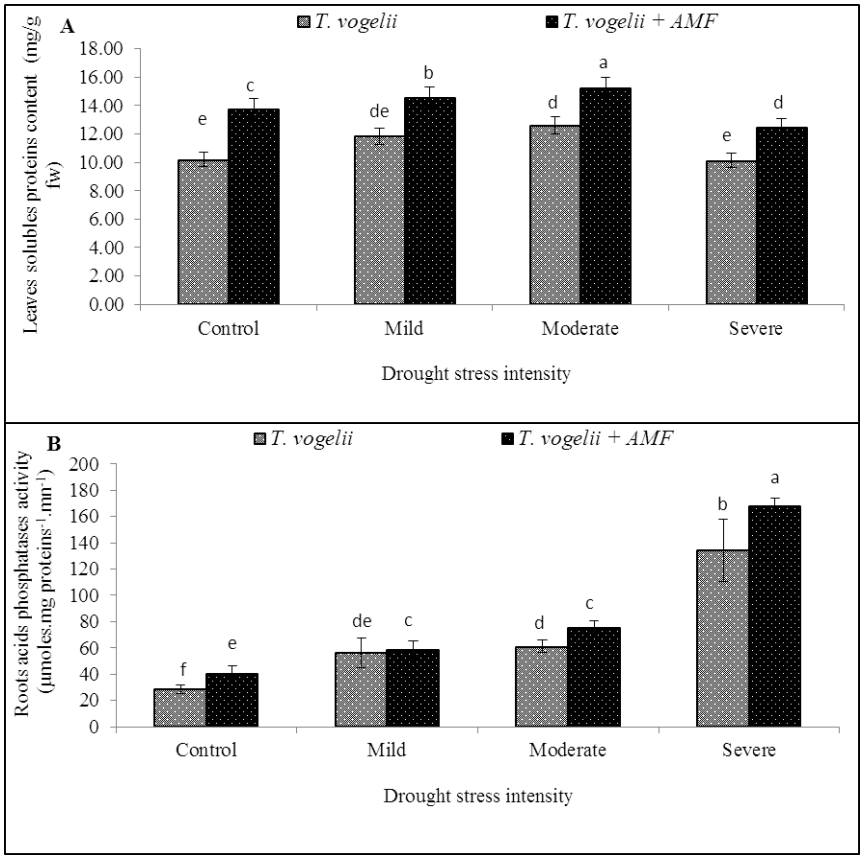

Figure 2. Total solubles proteins content (A); Specific activity of acids phosphatases (B) in mycorrhizal and non-mycorrhizal *T. vogelii* plants under severe, moderate, mild and no drought stress (control) conditions.

Table 1. Pearson correlation of evaluted parameters: Roots colonisation (RC), Total solubles sugars (TSS), Proline, Specific activity of acids phosphatases (SAA Pases), solubles proteins (TSP) and Total amino-acids (TAA

	T. vogelii					
	RC	TSS	Proline	SAA Pases	TSP	TAA
RC	1	0.179	- 0.561**	- 0.193	0.516*	0.363
TSS		1	0.241	0.541**	0.173	0.340
Proline			1	0.351	- 0.498*	- 0.483*
SAA Pases				1	- 0.351	- 0.010
TSP					1	0.563**
TAA						1

Note :* significant correlation at 5 % ; ** significant correlation at 1 %

3.6 Pearson correlation

The Pearson correlation (Table 1) between root colonization: RC (Tsoata et al., 2015) is negative for proline content (-0.561**) and acid phophatase activity (-0.193) in one hand; weak and non significant for TAA (0.363) and TSS (0.179) in the other hand. For TSP, the correlation is positive and significant (0.516*).

4. Discussion

Nitrogenized compound and amino acids are essential in plant metabolism as first products of assimilation of molecular nitrogen and as precursor of proteins and nucleic acids (Shao and al., 2009). The significant increase in the TAA observed for mycorrhizal control (31 %) compared to unmycorrhizal control, could be correlated with multiple beneficial roles that mycorrhizae renders to host plant (Ruiz-Lozano, 2003; Rillig, 2004; Wright, 2005).Under water stress, for all plants, mycorrhizal or not, there is significant accumulation of TAA in *T. vogelii*. This accumulation of TAA is a common answer of plants subjected to environmental stresses (Ranieri and al., 1980; Aspinall and Paleg, 1981). It was observed in groundnut (Asha and Rao, 2002); *Oryza sativa* (Hsu and Kao, 2003); bel pepper (Nath and al., 2005); *Vigna subterranea* (Tsoata and al., 2015).The accumulation of TAA under stress is observed for all growth phase; it suggests that their accumulation helps probably plant to tolerate conditions of water restriction through osmotic adjustment (Yadav and al., 2005); for sorghum variations of osmotic potential are must ascribable with accumulation of free amino acids (AA) (Greenway and Munns, 1980;Yadav and al., 2005). Osmotic adjustment slows down damage due to dehydration in environments where water is scare, by maintaining cells turgidity and physiological processes (Taiz and Zeiger, 2006).The significant decline of TAA observed for unmycorrhizal control under severe water stress, could be correlated with the negative effects of strong water deficit on metabolism and in nitrogen metabolism particularly (Ruiz-Lozano, 2003).Under water stress TAA are accumulated in mycorrhizal plants more than in not unmycorrhizal ones. This result could be justified by the fact that mycorhization increases the level of tolerance of host plant (Rabie and Almadini, 2005), also by deficit of photosynthetic products diverted by fungi (Cooper, 1984; Plenchette, 1991; Thomson and al., 1994). Mycorhization improves more than 10 times the amount of TAA for severe stress compared to soft and average stress. This increment would allow plant to maintain its water status using less AA for osmotic adjustment for soft and average stress, but much more for severe water stress.

The unmycorrhizal plants accumulate more proline than mycorrhizal ones. This result could be correlated with the fact that mycorhization improves water status of plant and its tolerance to water stress (Abdelmoneim and al., 2014), but also with the fact that mycorrhizal symbiosis uses for its needs part of plant host organic compounds (Plenchette, 1991; Thomson and al., 1994). Leaves proline contents increase with water stress level for un-or mycorrhizal plants; this increase declines when level of stress rises for mycorrhizal plants. This stimulation was already observed in several former works: Tamura et al. (2003); Feng et al. (2003); Hireche (2006); Tsoata et al. (2015). This proline accumulation in plants is an adaptive strategy frequently observed in order to limit effects of water stress (Acevedo and al., 1989) and would be connected directly to the application of water stress (Cechin and al., 2006; Abdelmoneim and al., 2014). It would be due to hydrolysis of proteins or inhibition of proteins synthesis by oxidative stress, leading to accumulation of proline (Feng and al., 2003; Shao and al., 2005, 2006). Furthermore it could be due to activation of biosynthesis join to inactivation of proline degradation. Proline accumulated in plants under water stress would intervenes in several phenomena: osmotic adjustment, tolerance to water stress (Szabados and Savouré, 2010; Karimi and al., 2012; Tsoata and al., 2015). It could also intervene in the regulation of cytoplasm pH (Pesci and Beffagna, 1984). The low proline content observed in unmycorrhizal *T. vogelii*, shrubby leguminous plant under severe hydrous stress, does not corroborate that of Tsoata et al. (2015), on *V. subterranea* herbaceous leguminous plant. It suggests that *T. vogelii* would be less tolerant to water stress compared to *V. subterranea* (Ruiz-Lozano and Azcon, 1997); otherwise the two species would use different mechanisms to

tolerate water stress. Proline would be prior osmoticum in osmotic adjustment for *V. subterranea* compared to *T. vogelii*.

TSS content of mycorrhizal control plants is higher than that of unmycorrhizal control. Tsoata et al. (2015), under identical experimental conditions, obtained similar result on *V. subterranea*. This result suggests that mycorrhizal control plants manufacture more TSS than unmycorrhizal ones. This could be correlated to beneficial effects of mycorhization for host plant, in particular improvement of water and nutritional status, thus allowing mycorrhizal host to produce more photosynthetats than unmycorrhizal ones (Tian and al., 2010; Abdelmoneim and al., 2014). The TSS increase when water stress level increases but more for unmycorrhizal than mycorrhizal ones. This result corroborates that of Irogoyen et al. (1992) on alfalfa; Tsoata et al. (2015) on *V. subterranea* and would be correlated with the hydrolysis of polysaccharides in water deficit condition (Shao and al., 2009), in order to maintain cells turgidity. Unmycorrhizal plants would have needs of more TSS for osmotic adjustment, than mycorrhizal ones. Mycorrhizal plants accumulate less TSS than unmycorhizal control surely for osmotic adjustment (Fallon and Phillips, 1989), but also to satisfy requirements of organic carbon of mycorrhizal fungi (Bago et al.; 2000) thus reducing the accumulated quantity. Our results are not in agreement with those of Patakas et al. (2002) on grapevines, where no difference in sugar content between stressed and not stressed plants is observed.

Significant increase 35 % ($p < 0.05$) is observed in TSP content of mycorrhizal control compared to unmycorrhizal ones. This result is in agreement with those of Tsoata et al. (2015) and would be correlated with mycorhization which improves the mineral-water nutrition and tolerance to water stress of host plant. Under water stress for all plants and for soft and average stress, there is increase in leaves TSP, which corroborates the results of Sohrabi et al. (2012) on *Cicer arietinum* and Tsoata et al. (2015) on *Vigna subterranea*; but drops for the severe stress; decline already observed by Tsoata and al. (2015) on voandzou. The increase in foliar TSP could be correlated with an effective osmotic adjustment, making it possible for plant to maintain normal metabolic activity for soft and average stress or with increase in the degree of tolerance to water stress of *T. vogelii*. Indeed Ashraf and Mehmood (1990) associate higher proteins contents to degree of tolerance to water stress. But soluble protein concentration under water stress is modulated by the nature of plant species (Terri and al., 1986) and type of tissue (Irogoyen and al., 1992). However, at the beginning of water stress, plants react in a dynamic way to restore their water status, so that their metabolism isn't significantly touched (Tardieu, 2005). The severe water stress would be sufficient to cause metabolic changes lessened leaves soluble protein content (Keller and ludlow, 1993). This reduction was already observed for sunflower Rodriguez and al. (2002); Costa and al. (2011) on cowpea; it would be due to increase in proline content, hydrolysis of proteins or to inhibition of their synthesis by oxidative stress leading to accumulation of proline (Feng and al., 2003).

A number of rhizosphere and/or mycorrhizosphere enzymes, as acid phosphatase, are produced by plant roots (Li and Fan, 1993) and control biogeochemical cycle of nutrients as: N, P, S (Philippot and al., 2009; Richardson and al., 2009; Carminati and al., 2010). The results of this work emphasize higher activity of acid phosphatases for mycorrhizal seedlings compared to unmycorrhizal ones even for control. This result is in agreement with those several former works: Dodd et al.(1987); Khalil and al. (1994); Tsoata et al.(2015). It could be correlated with higher intracellular production of this enzyme by mycorrhizal fungi hyphae (Saito, 1995). Other works showed that, one of the initial answers of water stressed plants due to phosphorus deficiency, caused by restriction of water availability, is increase activity of acid phosphatases (Duff and al., 1994). But intense activity of these acid phosphatases could also be due to weak variation of content of roots cytokinin during water stress in mycorrhizal seedlings. Indeed, cytokinins stimulate synthesis of proteins and inhibit their degradation, thus leading to high proteic content and strong activity of roots ATPase (Kuiper and al., 1991). Activity of acid phosphatases under water stress increases when stress level rises. This result is in agreement with those of several former works: Vieira Da Silva (1969) on cotton; Barrett-Lennard et al. (1982) on corn; Tsoata et al. (2015) on *Vigna subterranea*. The intense activity of acid phosphatases in water stressed plants would allow hydrolysis of organic phosphorus of plant, thus facilitating its mobilization and translocation from senescence tissue to those which needs them (Duff and al., 1994). Moreover high activity of acid phosphatases in mycorrhizosphere which is correlated positively with soil water contents (Chethan Kumar and al., 2008; Sardans and al., 2008), could thus partially mitigate effects of water stress on plants, and thus improves its tolerance against this constraint (Wu and al., 2011; Sharma and al., 2005; Tsoata and al., 2015).

Pearson correlation results, suggested that root colonization is inversely proportional to both proline content and acid phosphatase activity and may not affect directly TAA and TSS. But root colonization by AMF could be directly implicated in the amelioration of proteins biosynthesis (Tsoata et al., 2015).

5. Conclusion

Results of this work show that, for mycorrhizal *T. vogelii*, at an early stage of growth and in condition of water restriction, all the studied biochemical parameters are significantly improved. The TAA, proline, TSS, and TSP, play an essential role in the osmotic adjustment of plant in water stress condition, enabling him to maintain its water status, physiological and metabolic processes. The intense activity of acid phosphatases in host plants enables them to solubilize and mobilize some nutrients, phosphorus in particular and to improve its resistance to this stress. The improvement of water absorption, nutritional status and tolerance to water stress by mycorhization, allow plant to

perform normal functioning and good yield where water is scare. On the basis of these results, one can advise mycorrhizal biofertilizer like sure means to fight against dryness and to increase agricultural yield on soils victim of dryness.

References

Abdelmoneim, T.S., Tarek Moussa, A.A., Almaghrabi, O.A., Hassan Alzahrani, S., Abdelbagi I. (2014). Increasing plant tolerance to drought stress by inoculation with arbuscular mycorrhizal fungi. *Life Science Journal,* 11(1), pp. 10-17.

Acevedo, E., Conesa, A., Monneveux, P., Srivastav, A. (1989). Physiology breeding of winter creats for stressed mediterranean environments. Colloque n° 55, July 3-6, Montpellier, France, pp. 449-458.

Akédrin, T.N., N'guessan, K., Aké-Assi, E., & Ake, S. (2010). Effet de Légumineuses herbacées ou subligneuses sur la productivité du maïs. *J. of Animal & Plant Sci*., 8(2), pp. 953-963.

Anonyme, (2013). Année international des légumineuses. Organisation des Nations Unies pour l'Alimentation et l'Agriculture, 4 p.

Asha, S., Rao, K.N. (2002). Effect of simulated water logging on the levels of amino acids in groundnut at the time of sowing. *Ind. J. Plant Physiol*, 7, pp. 288-291.

Ashraf, M., Mahmood, S. (1990). Response of four *Brassica* species to drought stress. *Environmental Experimental Biology,* 30, pp. 93-100.

Aspinall, D., Paleg, L.G. (1981). Proline accumulation: Physiological aspects: in L.G. Paleg, and D. Aspinall (Eds.), *Physiology and Biochemistry of drought resistance in plants*, Australia, Sydney Academic Press, pp. 215-228.

Azzouz, F. (2009). *Les réponses morphologiques et biochimiques chez le haricot (Phaseolus vulgaris L.) soumis à un stress hydrique. Mémoire* Diplôme de Magister Univer. D'Oran ESSENIA Fac. Sc. 82 p.

Bago, B., Pfeffer, P.E., Scharcha-Hill, Y. (2000). Carbon metabolism and transport in arbuscular mycorrhizas. *Plant Physiology,* 124, pp. 949-957.

Barrett-Lennard, E.G., Robson, A.D., and Greenway, H. (1982). Effect of phosphorus deficiency and water deficit on phosphatase activities from wheat leaves. *J. Exp. Bot.,* 33**,** pp. 682-693.

Carminati, A., Moradi, A.B., Vetterlein, D., Vontobel, P., Lehmann, E., Weller, V., Vogel, H., and Oswald S.E. (2010). Dynamics of soil water content in the rhizoshere. *Plant Soil,* 332, pp. 163-176.

Ceccarelli, S. (2010). Plant breeding and climate change: in Ceccarelli, S., and Grando, S. (eds) 2010. *Proceedings of the 10th International Barley Genetic Symposium 5-10April 2008,* pp.16-29, Alexandria, Egypt.

Cechin, I., Rossi, S.C., Oliveira, V.C., & Fumis, T.F. (2006). Photosynthetic responses and proline content of mature and young leaves of sunflower plants under water deficit. *Photosynthetica,* 44(1), pp. 143-146.

Chethan Kumar, K.V., Chandrashekar, K.R., and Lakshmipathy, R. (2008). Variation in Arbuscular Mycorrhizal Fungi and Phosphatase Activity Associated with *Sida cordifolia* in Karnataka. *World Journal of Agricultural Sciences,* 4(6), pp. 770-774.

Cooper, K.M. (1984). Physiology of VA mycorrhizal association. In: VA mycorrhizae, Eds Powell, C.L., and Bagyaraj, D.J., CRC Press, Inc, Bota Raton, Florida, pp. 155-186.

Costa, D.L.C.R., Lobato, S.D.K.A., Silvera, D.G.A.J., and Laughinghousevi, D.H. (2011). ABA mediated proline synthesis in cowpea leaves exposed to water deficiency and rehydration. *Turk. J. Agric. For.,* 35, pp. 309-317.

Dodd, J.C., Burton, C.C., Jeffries, P. (1987). Phosphatase activity associated with the roots and the rhizosphère of plants infected with vesicular mycorrhizal fungi. *New Phytologist,* pp. 107-172.

Duff, S.M.G., Sarath, G., Plaxton, W.C. (1994). The role of acids phosphatase in plant phosphorus metabolism. *Physiologia Plantarum,* 90, pp. 791-800.

Duke, J.A. (1981). *Handbook of legume of world economic importance*. Plenum, New York and London.

El Sohaimy, S.A. (2012). Functional foods and nutraceuticals-modern approach to food science. *World Appl. Sci. J.,* 20(5), pp. 691-708.

Fallon, K.M., and Phillips, R. (1989). Responses to water stress in adopted and unadopted carrot cell suspension cultures. *J. Exp. Bot.,* 40, pp. 681-687.

Feng, Z., Guo, A., Feng, Z. (2003). Amelioration of chilling stress by triadimefon in cucumber seedlings. *Plant Growth Regul,* 39, pp. 277-283.

Fischer, E.M., and Schär, C. (2010). Consistent geographical patterns of changes in high impact European heatwaves. *Nat. Geosci.,* 3, pp. 398-403.

Gao, X., Lu, X., Wu, M., Zhang, H., Pan, R. (2012). Co-Inoculation with *Rhizobia* and AMF inhibited soybean red crown rot: from field study to plant defense-related gene expression analysis. *PLOSONE,* 7(3), pp. 33-97.

Gaye, J.P., & Bloc, D. (1992). La tolérance au stress chez le maïs. *Perspective Agricole,* 175, pp. 100-106.

Gholamhoseini, M., Ghalavand, A., Dolatabadian, A., Jamshidi, E., Khodaei-Jorghan, A. (2013). Effects of arbuscular mycorrhizal inoculation on growth, yield, nutrient uptake and irrigation water productivity of sunflowers grown under drought stress. *Agricultural Water Management,* 117, pp. 106-114.

Greenway, H.R., Munns, R. (1980). Mechanism of salt tolerance in non-halophytes. *Ann. Rev. Plant Physiol.,* 31, pp. 149-190.

Hindumathi, A., Reddy, B.N. (2012). Synergistic effect of arbuscular mycorrhizal fungi and *Rhizobium* on the growth and charcoal root of soybean [*Glycine max* (L.) Merr.]. *World Journal of Science and Technolog,* 2(7), pp. 63-70.

Hireche , Y. (2006). *Réponse de la luzerne (Medicago sativa* L.) *au stress hydrique et à la profondeur du semis.*Thèse de Magistère en sciences agronomique, Université Al Hadj Lakhdar-Batna.

Hsu, Y.T., Kao, C.H. (2003). Changes in protein and amino acid contents in two cultivars of rice seedlings with different apparent tolerance to cadmium. *Plant Growth Regul.,* 40, pp. 147-155.

Irigoyen, J.J., Emerich, D.W., Sanchez-Diaz, M. (1992). Water stress induced changes in concentrations of proline and total soluble sugars in nodulated alfalfa (*Medicago sativa*) plants. *Physiologia Plantarum,* 84, pp. 55-60.

Jensen, E.S., Peoples, M.B., Boddey, R.M., Gresshoff, P.M., Hauggaard Nielsen, H., Bruno, J.R.A., Morrison, M.J. (2012). Legumes for mitigation of climate change and the provision of feedstock for biofuels and biorefineries. *Agron. Sustain. Dev.,* 32, pp. 329-364.

Karimi, S., Abbaspour, H., Sinaki, J.M., Makarian, H. (2012). Effects of water deficit and chitosan spraying on osmotic adjustment and soluble protein of cultivars castor bean (*Ricinus communis* L.). *Journal of Stress Physiology and Biochemistry,* 8(3), pp. 160-169.

Keller, F., Ludlow, M.M. (1993). Carbohydrate metabolism in drought-stressed leaves of pigeon pea (*Cajanus cajan*), *J. Exp. Bot.,* 44, pp. 1351-1359.

Khalil, S., Loynachan, T.E., Tabatabai, M.A. (1994). Mycorrhizal dependency and nutrient uptake by improved and unimproved corn and soybean cultivars. *Agronomy Journal,* 86, pp. 949-958.

Kuiper, D., Sommarin, M., Kylin, A. (1991). The effects of mineral nutrition and benzyladenine on the plasmalema ATPase activity from roots of wheat and *Plantago major* spp. *pleiosperma*. *Physiologia Plantarum,* 81, pp. 169-74.

Li, L.M., and Fan, X.H. (1993). The research overview of rhizosphere enzymes. *Soils,* 25, pp. 299-303.

Makoshi, M.S., & Arowolo, R.O.A. (2011). Therapeutic effects of *Tephrosia vogelii* ointement in the treatment of bovine dermatophilosis. *Journal of Veterinary Medicine and Animal Health*, 3(4), pp. 51-55.

Nath, A.K., Kumari, S., Sharma, D.R. (2005). In vitro selection and characterization of water stress tolerant culture of bell pepper. *Plant Physiol.,* 107, pp. 125-130.

National Academy of Sciences. (1979). *Tropical legumes: resources for future*. Washington D.C.

Orwa, C., Mutua, A., Kindt, R., Jamnadass, R., & Simons, A. (2009). Agroforestry Database: a tree reference and selection guide version 4.0, pp. 1-5.

Patakas, A., Nikolaou, N., Zioziou, E., Radoglou, K., Nortsakis, B. (2002). The role of organic solute and ion accumulation in osmotic adjustment in drought-stressed grapevines. *Plant Science,* 163, pp. 361-367.

Pennisi, E. (2008). The blue revolution drop by drop, gene by gene. *Science,* 32, pp. 171-173.

Pesci, P., & Beffagna, N. (1984). Inhibiting effect of fusicoccin on abscisic acid-induced proline accumulation in barley leaf segments. *Plant Science Letters,* 36, pp. 7-12.

Philippot, L., Hallin, S., Borjesoon, G., and Baggs, E.M. (2009). Biochemical cycling in the rhizosphere having an impact on global change. *Plant Soil,* 321, pp. 61-81.

Plenchette, C. (1991). Utilisation des mycorhizes en agriculture et horticulture. In : Strullu, D.G., Garbaye, J., Perrin, P., Plenchette, C. eds. *Les mycorhizes des arbres et plantes cultivées*. Paris: Lavoisier.

Rabie, G.H., Almadini, A.M. (2005). Role of bioinoculants in development of salt tolerance of *Vicia faba* plants under salinity stress. *Afr. J. Biotech.,* 4, pp. 210-220.

Ranieri, A., Bernardi, R., Lanese, P., and Soldatini, G.F. (1989). Changes in free amino acid content and protein pattern of maize seedlings under water stress. *Env. Exp. Bot.,* 29, pp. 351-357.

Richardson, A.E., Barea, J.M., McNeil, A.M., and Prigent-Combaret. (2009). Acquisition of phosphorus and nitrogen in rhizosphere and plant growth promotion by microorganisms, *Plant Soil,* 321, pp. 305-339.

Rillig, M.C. (2004). Arbuscular mycorrhizae, glomalin and soil aggregation. *Can. J. Soil Sci.,* 84, pp. 355–363.

Rodriguez, D.J., Romero-Garcia, J., Rodriguez-Garcia, R., Angulo-Sanchez, J.L. (2002). Characterization of protein from leaves and seeds: relationship of biomass and seed yield. In: *Trends in new crops and new uses,* Janick, J., Whipkey, A., Eds. ASHS Press, Alexandria, pp. 143-149.

Ruiz-Lozano, J.M. (2003). Arbuscular mycorrhizal symbiosis and alleviation of osmotic stress. New perspectives for molecular studies. *Mycorrhiza,* 13, pp. 309–317.

Ruiz-Lozano, J.M., and Azcón, R. (1997). Effect of calcium application on the tolerance of mycorrhizal lettuce plants to polyethylene glycol-induced water stress. *Symbiosis,* 23, pp. 9-21.

Saito, M. (1995). Enzyme activities of the internal hyphae and germinated spores of an arbuscular mycorrhizal fungus *Gigaspora margarita* Becker & Hall. New *Phytologist,* 129, pp. 425-431.

Sardans, J., Peñuelas, J., Ogaya R. (2008). Drought's impact on Ca, Fe, Mg, Mo and S concentration and accumulation patterns in plant sand soil of a Mediterranean ever green *Quercus ilex* forest. *Biogeochemistry,* 87, pp. 49-69.

Shao, H.B., Liang, Z.S., Shao, M.A., Sun, Q. (2005). Dynamic changes of antioxidative enzymes of ten wheat genotypes at soil water deficits. *Biointerfaces,* 42, pp. 187-195.

Shao, H.B., Liang, Z.S., Shao, M.A. (2006). Osmotic regulation of 10 wheat (*Triticum aestivum* L.) genotype at soil water deficits. *Biointerfaces,* 47, pp.132-139.

Shao, H.-B., Chu, L.-Y., Abdul Jaleel, C., Manivannan, P., Panneerselvam, R., and Shao, M.-A. (2009). Understanding water deficit stress- induced changes in the basic metabolism of higher plants-biotechnologically and sustainably improving agriculture and the ecoenvironment in arid regions of the globe. *Critical Reviews in Biotechnology,* 29(2), pp. 131-151.

Sharma, A.D., Singh, N., and Kang, J.K. (2005). Short-term water logging-induced changes in phosphatase activity in shoot and roots of sorghum seedlings: Role of phosphatases during water logging in relation to phosphorus during water logging in relation to phosphorus. *Gen. Appl. Plant Physilogy*, 31(2), 71-79.

Smith, M.D. (2011). The ecological role of climate extremes: current understanding and future prospects. *J. Ecol.*, 99, pp. 651-655.

Sohrabi, Y., Heidari, G., Weisany, W., Ghasemi_Golezani, K., and Mohammadi, K. (2012). Some Physiological Responses of Chickpea Cultivars to Arbuscular Mycorrhiza under Drought Stress1. *Russian Journal of Plant Physiology*, 59(6), pp. 708-716.

Szabados, L., and Savouré. (2010). Proline: a multifunctional amino-acid. Trends Plant Sci., 15, pp. 89-97.

Taiz, L., Zeiger, E. (2006). *Plant physiology*, 4th Ed., Sinauer Associates Inc. Publishers, Massachusetts.

Tamura, T., Hara, K., Yamaguchi, Y., Koizumi, N., Sano, H. (2003). Osmotic stress tolerance of transgenic tobacco expressing a gene encoding a membrane-located receptor-like protein from tobacco plants. *Plant Physiology*, 131, pp. 454-462.

Tardieu, F. (2005). Plant tolerance to water deficit: physical limits and possibilities for progress. *C. R. Geoscience*, 337, pp. 57–67.

Terri, J.A., Turner, M., Gurevitch, J. (1986). The response of leaf water potential and crassulacean acid metabolism to prolonged drought in *Sedum rubrotinctum*. *Plant Physiol.*, 81, pp. 678–680.

Thomson, B.D., Grove, T.S., Malajczuk, N., & Hardy Gest. (1994). The effectiveness of ectomycorrhizal fungi in increasing the growth of *Ecalyptus globules* Labill. In relation to root colonization and hyphal development in soil. *New Phytologist*, 126, pp. 517-524.

Tian, C., Kasiborski, B., Koul, R., Lammers, P.J., Bucking, H., Shachar-Hill, Y. (2010). Regulation of the nitrogen transfer pathway in the arbuscular mycorrhizal symbiosis: gene characterization and the coordination of expression with nitrogen flux. Plant Physiol., 153, pp. 1175-1187.

Tsoata, E., Njock, S.R., Youmbi, E., Nwaga, D. (2015). Early effects of water stress on some biochemical and mineral parameters of mycorrhizal *Vigna subterranea* (L.) cultivated in Cameroon. *Int. J. Agri. & Agric. R.*, 7(2), pp. 21-35.

UNDP BCPR. (2003). *Human development report. Millennium development goals: A compact among nations to end human poverty*. New York Oxford: UNDP.

Vance, C.P., Graham, P.H., Allan, D.L. (2000). *Biological nitrogen fixation. Phosphorous: a critical future need*. In Pedrosa, F.O., Hungria, M., Yates, M.G., Newton, W.E., eds, *nitrogen fixation: from molecules to crop productivity*. Kluwer Academic Publixhers, Dordrecht, The Netherlands, pp. 506-514.

Vieira-Da-Silva, J.B. (1969). Comparaison entre cinq espèces de *Gossypium* quant à l'activité de la phosphatase acide après un traitement osmotique. Etude de la vitesse de solubilisation et de formation de l'enzyme. *Z. Pflanzenphysiol.*, 60, pp. 385-387.

Visitpanich, T., Batterham, E.S., and Norton, B.W. (1985a). Nutritional value of chickpea (*Cicer arictinum*) and pigeon pea (*Cajanus cajan*) meals for growing pigs and rats. 1. Energy content and protein quality. *Austral. J. Agrie. Res.*, 36, pp. 327-335.

Westphal, E., Ferwerda, J.D., Van Gils-Meeus, H.A.E., Mustsaers, H.J.W., & Westphal-Stevels J.M.C. (1985). *Cultures vivrières tropicales avec référence spéciale au Cameroun*, Pudoc, Wageningen, Netherlands.

Wright, S.F. (2005). Management of arbuscular mycorrhizal fungi. In Zobel, R.W., and Wright, S.F., Eds., *Roots and Soil Management*: *Interactions between Roots and the Soil*. American Society of Agronomy, Crop Science Society of America, Soil Science Society of America, Madison, pp. 183-197.

Wu, C., Wang, Q., Xie, B., Wang, Z., Cui, J., and Hu, T. (2011). Effects of drought and salt stress on seed germination of three leguminous species. *Afri. J. of Biotech.*, 10(78), pp. 17954-17961.

Yadav, S.K., Lakshmi, N.J., Maheswari, M., Vanaja, M., Venkateswarlu, B. (2005). Influence of water deficit at vegetative anthesis and grain filling stages on water relation on water relation and grain yield in sorghum. *Ind. J. Plant Physiol.*, 10, pp. 20-24.