Annals of West University of Timişoara, ser. Biology, 2014, vol XVII (1), pp.1-8

WATER DEFICIT EFFECT ON YIELD AND FORAGE QUALITY OF *MEDICAGO SATIVA* POPULATIONS UNDER FIELD CONDITIONS IN MARRAKESH AREA (MOROCCO)

Mohamed FARISSI[1,2]*, Cherki GHOULAM[1], Abdelaziz BOUIZGAREN[2]

[1]Unit of Plant Biotechnology and Agro-physiology of Symbiosis, Faculty of Sciences and Techniques, PO. Box 549, Gueliz 40000 Marrakesh, Morocco
[2]Unit of Plant Breeding, National Institute for Agronomic Research (INRA), PO. Box 533, Gueliz 40000, Marrakesh, Morocco.
**Corresponding author's email address: farissimohamed@gmail.com*
Received 29 December 2013; accepted 28 February 2014

ABSTRACT
The present study focused the effect of water deficit on agronomic potential and some traits related to forage quality in plants of Moroccan Alfalfa (Medicago sativa L.) populations (Taf 1, Taf 2, Dem and Tata) originated from Oasis and High Atlas of Morocco and an introduced variety from Australia (Siriver). The experiment was conducted under field conditions in experimental station of INRA-Marrakech and under two irrigation treatments. The first treatment was normal irrigation, providing an amount of water corresponding to the potential evapo-transpiration of the crop, and the second treatment was water deficit stress (one irrigation per cut). For each treatment, the experiment was conducted as a split plot based on a randomized complete block design with four replications. The plants were measured and analyzed over three cuts. Some agronomic traits as, plant height, fresh and dry forage yields were measured. The forage quality was evaluated by leaf:stem ratio and the contents of plants in proteins and nitrogen. The results indicated that the water deficit has negatively affected the plant height and forage yield. The decrease in leaf:stem ratio was observed under water deficit conditions. However, the proteins and nitrogen contents were unaffected. The behavior of tested alfalfa genotypes was significantly different. The Moroccan alfalfa populations were more adapted to water deficit conditions comparatively to Siriver variety and the Tata population was the most adapted one.
KEY WORDS: *Alfalfa, water deficit, forage yield, forage quality*.

INTRODUCTION

Alfalfa (*Medicago sativa* L.) is the main cultivated forage crop in Morocco covering 100,000 ha (22% of the total cultivated area of forage crops), because of its suitability to low-input conditions, its positive effects on soil fertility, and the high protein content and quality of its forage (Campiglia *et al*, 1999; Huyghe, 2003; Bouizgaren *et al*, 2011; Latrach *et al*, 2014; Farissi *et al*, 2014). The local cultivars of this species are widely used in the Moroccan traditional agro-ecosystems, oasis and mountain. They strongly contribute to socio-economic development of local families

(Farissi *et al*, 2011; Farissi *et al*, 2013a; 2013b; 2013c) as a basic nutrition for livestock. However, the successive of drought years induced by climate change and population growth in Morocco, reduced increasingly the amount of agricultural water reserve (Farissi *et al*., 2013a). Consequently, the growth and the productivity of alfalfa and many vegetal species were found limited under water unavailability conditions. Indeed, drought stress is one of the major abiotic stresses that limit plant production and growth of many *Medicago* species (Chebouti *et al*, 2001; Bouizgaren *et al*, 2011). This constraint causes a many agro-physiological and biochemical changes that can be observed at the whole plant level as the death of plant and / or decreases in its productivity. Almost of all changes are associated with activation of some physiological and biochemical processes allowing an adaptation to this constraint (Farissi *et al,* 2013a).

To maximize agricultural productivity in areas known for water scarcity, genotypes with high-level tolerance must be selected and recommended for these areas. In this context, the present work aims to evaluate the effect of water deficit on growth and productivity of some alfalfa cultivars. Some agronomical parameters as plant height and forage yield were evaluated. The forage quality, commonly sought in the forage species, was assessed by measuring leaf: stem ratio and contents of plants in proteins and nitrogen.

MATERIALS AND METHODS

Site description

The experiment was carried out at Saâda Experimental Station of National Institute for Agronomic Research-Marrakesh (INRA-Marrakesh) located at 7 km west of Marrakesh city. The site is an agro-ecological zone having an altitude of about 468 m above sea level. The experiment was conducted during 2011 on a loamy clay soil with annual average maximum temperature of 28.7°C, annual average minimum temperature of 13.5 °C and total annual precipitation of 226.54 mm. The evaluation was done during April to July at the site and this period was characterized by the scarcity of rainfall associated with high temperatures.

Plant Material and field conditions

This study concerned Four Moroccan alfalfa populations (*Tafilalet 1 -Taf1, Tafilalet 2-Taf 2, Demnate -Dem* and *Tata*) and an introduced variety from Australia "*Siriver*". These populations are originated from, South-east and ouest oasis and High Atlas Mountains of Morocco where they have been cultivated for many centuries and are still widely used by farmers in their traditional agro-ecosystems (Farissi *et al,* 2011; 2013a; 2013b). The trial was carried out in Saâda Experimental Station of National Institute for Agronomic Research-Marrakesh (INRA-Marrakech) and under two irrigation treatments. The first treatment was normal irrigation, providing an amount of water corresponding to the potential evapotranspiration of the crop, and the second treatment was water deficit stress (one irrigation per cut). For each treatment,

the experiment was conducted as a split plot based on a randomized complete block design with four replications. To avoid water going to the neighbouring subplots, we grouped within one plot all subplots subjected to the same treatment. The interplot distances were 10 m. The subplot area was 2 m^2. All cultivars were sown on the same date in autumn for both treatments with a density of about 200 plants per m^2. Gravity irrigation was adopted in both treatments.

Plant height

Plant heights were measured during each cut (three consecutive cuts) and at four times of plant growth, 7^{th} day, 14^{th}, 21^{st} and 28^{th} day.

Forage yield

Fresh forage yield was assessed by whole-plot harvest. After weighing harvested forage, 500g of fresh material were dried in an oven at 70°C during 48 h for dry forage yield measurement. The same cutting regimes were applied for all cultivars tested.

Forage quality

The forage quality was assessed by leaf: steem ratio, proteins contents and nitrogen contents:

Leaf: stem ratio

The leaves and stems were separated and weighed using a precision weighing balance.

Proteins contents

The contents of proteins were determined according to Bradford (1976) method. The samples were ground with a mortar and pestle using Tris-HCl buffer (0,1M pH 7,5) and centrifuged at 16000 g for 15 min. Diluted extract (2 ml) was added to tubes containing 2 ml of Bradford reagent. After stirring, the optical density was measured at 595 nm. The amounts of proteins were determined using a standard range established by using reference solutions of bovine serum albumin (BSA).

Nitrogen contents

The plant nitrogen contents were determined according to Khjeldal method (Faghire *et al,* 2011; Bargaz *et al*, 2012). 0.5 g of dried matter was finely ground and placed in Buchi tubes with a pinch of catalyst (potassium sulphate (100 g) , copper sulfate (20 g) and selenium powder (2g) and 10 ml of concentrated H_2SO_4. Then, the tubes were placed in the digestion block and closed with a lid. The digestion of the organic matter was held for 2 hours at temperature of 400°C. The tubes were placed and cooled at the distiller programmed to add NaOH (10 N) and distilled water. Distillation during 4 min to recover a volume of about 100 ml in a flask containing 50 ml of boric acid H_3BO_3 (20 g / L) and 5 drops of red indicator. The distillate collected was titrated with a solution of hydrochloric acid (N/100) to turn greenish coloration to the original red color. The volume of hydrochloric acid of titration was used to

calculate the percentage of nitrogen. Results were expressed in mg of nitrogen per g dry weight.

RESULTS AND DISCUSSIONS

Effect on plant height

Figure 1 showed the effect of irrigation water regime on plant heights of four Moroccan alfalfa populations and *Siriver* variety at different times of growth. Under water deficit conditions, the plant heights were significantly reduced ($P<0.001$) in all cultivars tested and at all different times of plant growth. ANOVA test showed that the behavior of the tested alfalfa material was not significantly different ($P>0.05$). However, in terms of the average values recorded, the plants of *Siriver* variety showed the lowest averages at all times assessed of growth and the plants of *Tata* have maintained the most height in all times of growth. The other populations showed the intermediate values.

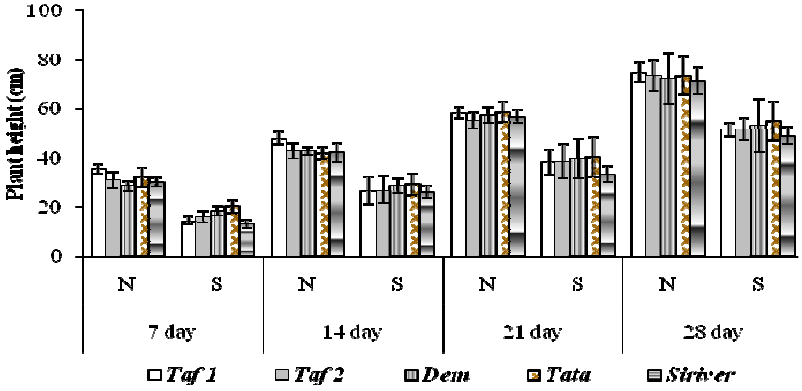

Fig.1. Effect of irrigation water regime (N: normal irrigation; S: water stress) on plant heights of four Moroccan alfalfa populations (*Taf 1*, *Taf 2*, *Dem* and *Tata*) and *Siriver* variety at four times of plant growth (7th day, 14th, 21st and 28th day). Values are means of three consecutive cuts with 12 replicates for each. Bars represent standard errors.

Effect on forage yield

The effect of water deficit on fresh and dry forage yields is showed in figure 2. Results indicated that the fresh and dry forage yields were significantly reduced ($P<0.001$) by water deficit. The greatest reductions were recorded in *Siriver* variety (17.33 and 20.14 % for fresh yield and dry yield respectively). The *Tata* population developed the lowest reductions, comparatively to the other cultivars tested, showing the reductions of 11.22 and 16.34% in fresh yield and dry yield respectively. The remaining populations showed the intermediate reductions with the preference for *Dem* population.

Effect on forage quality

The effect of water deficit on forage quality is indicated in table 1. Under this constraint the leaf: stem ratio was significantly reduced (P<0.001) with the significant differences (P<0.001) between the alfalfa cultivars studied. Indeed, the highest values were noted in plants of *Tata* population (0.72). The lowest values were mentioned by *Siriver* variety (0.59) while *Taf 1*, *Taf 2* and *Dem* have appeared respectively 0.61, 0.63 and 0.66. For the proteins and the nitrogen contents, same table shows that both parameters are unaffected by water deficit comparatively to optimal irrigation (P>0.05). However, the average values recorded for the nitrogen contents varied from 43.88 in *Siriver* variety to 46.85 mg.g DW^{-1} in *Tata* population. For protein contents, the values ranged between 17.65 to 18.72 mg. g FW^{-1} for *Siriver* and *Tata* respectively.

Table 1. Effect of water deficit on some parameters related to forage quality (Leaf: Stem ratio and the contents in proteins and nitrogen) in plants of four Moroccan alfalfa populations (*Taf 1, Taf 2, Dem* and *Tata*) and *Siriver* variety. Values are means of three consecutive cuts with four replicates for each.
*: Significance at 0.05 probability level; ***: significance at 0.001 probability level; NS: not significant at 0.05.

Parameter	POP	Normal irrigation	Water deficit	Irrigation	POP	Interaction
Leaf : Stem ratio	**Taf 1**	0.68	0.61	63.42***	18.90***	6.17***
	Taf 2	0.82	0.63			
	Dem	0.79	0.66			
	Tata	0.89	0.72			
	Siriver	0.76	0.59			
Nitrogen (mg.g DW^{-1})	**Taf 1**	44.91	45.15	0.23 NS	1.02 NS	2.45 NS
	Taf 2	44.77	45.57			
	Dem	44.58	46.28			
	Tata	45.30	46.85			
	Siriver	45.87	43.88			
Proteins (mg.g FW^{-1})	**Taf 1**	17.42	17.70	1.05 NS	3.01*	1.84 NS
	Taf 2	17.68	17.90			
	Dem	17.91	18.50			
	Tata	18.16	18.72			
	Siriver	18.38	17.65			

In the arid and semiarid areas, the availability and the management of irrigation water have become priorities of great importance. In Morocco, the successive years of drought, induced by climate change and population growth, increasingly reduced the amount of agricultural water reserve. Consequently, a rational and efficient use of irrigation water will be helpful to solve this problem (Farissi *et al,* 2013a). The most important strategies employed in the last few years to reduce the

effects of water scarcity on plant production have focused on a selection of host genotypes that are tolerant to water deficit conditions. In the present study, we have noted that the water deficit has significantly reduced the plant height and the forage yield. The behavior of alfalfa cultivars tested was significantly different. *Tata* population appeared to be the most tolerant population and *Siriver* variety were found the least tolerant one. However, the other populations have developed an intermediate tolerance. The reduction in plant height is one of the plant strategies for tolerance to environmental constraints. Similar results were noted in *Medicago falcata* L. (Wang *et al*, 2012) and *Zea mays* L. (Reddy *et al*, 2013). Also, the reduction in growth and yield by water deficit has been documented in many species of *Medicago* as *M. sativa* L. (Wang *et al,* 2009; Bouizgaren *et al,* 2011), *M. truncatula* (Chebouti *et al*, 2001) and *M. intertexta* (Laouar *et al*, 2001).

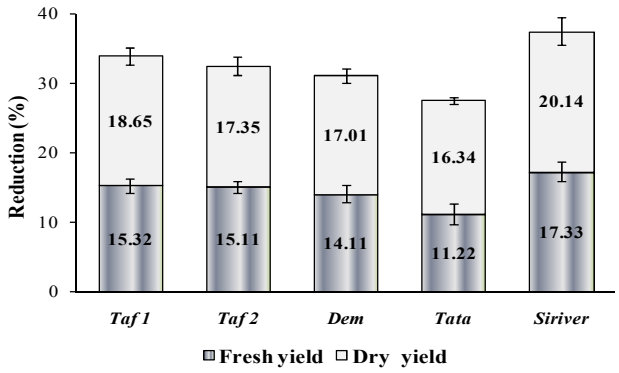

Fig.2. Effect of water deficit on fresh and dry forage yields of four Moroccan alfalfa populations (*Taf 1*, *Taf 2*, *Dem* and *Tata*) and *Siriver* variety. Results are expressed as reduction percentage of optimal irrigation. Values are means of three consecutive cuts with four replicates for each. Bars represent standard errors.

As well as forage yield, a major challenge faced alfalfa growers during the past 30 years has been the increased emphasis on forage quality. The need to produce high-quality hay affects marketing and price, as well as yield and stand life. Demands for high-quality alfalfa by the marketplace have been relentless. Although crop yield is still the primary economic factor determining forage crop value per unit on land area, forage quality has become a close second (Orloff and Puntam, 2007). Our results showed the decrease in leaf: stem ratio under water defcit. Reduced leaf to stem ratio is a major cause of the decline in forage quality with maturity, and the loss in quality that occurs under adverse hay curing conditions. The proportion of stems increases and quality decreases. This ratio is crucial to the alfalfa digestible quality. Thus, food quality forage decreases with decreasing the ratio of leaves and stems (Volenec and Cherney, 1990). Drought invariably decreases plant quality through reduced leaf:stem

ratio, accelerated flowering and lignin accumulation (Barker and Caradus, 2001). In this study, the forage quality was also assessed by the proteins and the nitrogen contents. We have noted that the water deficit have unaffected these parameters in all cultivars studied ($P>0.05$).

CONCLUSION

We conclude that the water deficit has negatively affected the plant height and forage yield. The decrease in leaf:stem ratio was observed under this constraint. However, the proteins and nitrogen contents were unaffected. The Moroccan alfalfa populations were more adapted to water deficit conditions comparatively to *Siriver* variety and *Tata* population was the most adapted one.

REFERENCES

- Bargaz A., Faghire M., Abdi N., Farissi M., Sifi B., Drevon JJ., Ikbal M.C., Ghoulam C. 2012. Low Soil Phosphorus Availability Increases Acid Phosphatases Activities and Affects P Partitioning in Nodules, Seeds and Rhizosphere of *Phaseolus vulgaris*. *Agriculture* 2:139-153.
- Barker D.J., Caradus J.R. 2001. Adaptation of forage species to drought. In: Proceedings of XIX International Grassland Congress, Brazil, 241-246.
- Bouizgaren A., Farissi M., Khalida R., Ghoulam C., Barakate M., Al Feddy M.N. 2011. Assessment of summer drought tolerance variability in Mediterranean alfalfa (*Medicago sativa* L.) cultivars under Moroccan fields conditions. *Arch of Agro Soil Sci* 1-14.
- Bradford M.M.1976. A rapid and sensitive method for the quantification of microgram quantities of protein utilizing the principal of protein dye binding. *Anal Biochem* 72:248-257.
- Campiglia E., Caporali F. Barberi R., Mancinelli R. 1999. Influence of 2-, 3-, 4- and 5-year stands of alfalfa on winter wheat yield. In: Olesen JE, Eltun R, Goodlimg MJ, Jensen ES, Kopke U, editors. Proc. Int. Workshop 'Designing and Testing Crop Rotations for Organic Farming'. DARCOF, Tjele, DK. p. 165–171.
- Chebouti A., Mefti M., Abdelguerfi A. 2001. Effet du stress hydrique sur le rendement en gousses et en graines chez trois espèces de luzernes annuelles: *Medicago aculeteata*, *M. orbicularis M. truncatula*. *Cah Opt Médit* 45: 163-166.
- Faghire M., Bargaz A., Farissi M., Palma F., Mandri B., Lluch C., Tejera García N.A., Herrera-Cervera J.A., Oufdou K., Ghoulam C. 2011. Effect of salinity on nodulation, nitrogen fixa tion and growth of common bean (*Phaseolus vulgaris* L.) inoculated with rhizobial strains isolated from the Haouz region of Morocco. *Symbiosis,* 55:69-75.
- Farissi M., Bouizgaren A., Faghire M., Bargaz A., Ghoulam C. 2011. Agro-physiological responses of Moroccan alfalfa (*Medicago sativa* L.) populations to salt stress during germination and early seedling stages. *Seed Sci Technol* 39: 389-401.
- Farissi M., Bouizgaren A., Faghire M., Bargaz A., Ghoulam C. 2013a. Agro-physiological and biochemical properties associated with adaptation of *Medicago sativa* populations to water deficit. *Turk J Bot* 37:1166-1175.
- Farissi M., Faghire M, Bouizgaren A., Bargaz A., Makoudi B., Ghoulam C. 2014. Growth, nutrients concentrations and enzymes involved in plants nutrition of alfalfa populations under saline conditions. *J Agr Sci Tech* 16:301-314.
- Farissi M., Ghoulam C., Bouizgaren A. 2013b. Changes in water deficit saturation and photosynthetic pigments of Alfalfa populations under salinity and assessment of proline role in salt tolerance. *Agr Sci Res J* 3:29-35.
- Farissi M., Ghoulam C., Bouizgaren A. 2013c. Variabilité de la tolérance à la salinité de la luzerne : évaluation au stade germination de populations issues de différents agro-écosystèmes marocains. *Fourrages* 216:329-332
- Huyghe C. 2003. Les fourrages et la production de protéines. In: Acte des Journées de l'Association France, aise pour la Production Fourragère (AFPF). Paris: AFPF. p. 17–32.
- Laouar M., Kiès N., Abdellaoui K., Bennour A., Bettahar N., Kadi S., Bouzza L. 2001. Effet du stress hydrique sur le comportement physiologique de dix populations de *Medicago intertexta*. *Cah Opt Médit* 45:167-171.

- Latrach L, Farissi M, Mouradi M, Makoudi B, Bouizgaren A., Ghoulam C. 2014. Growth and nodulation in alfalfa-rhizobia symbiosis under salinity: effect on electrolyte leakage, stomatal conductance and chlorophyll fluorescence. *Turk J Agric For* doi: 10.3906/tar-1305-52.
- Orloff S.B., Puntam D.H.2007. Forage quality and testing. In C.G. Summers and D.H. Putna,eds., Irrigated alfalfa management in Mediterranean and desertzones. Chapter 16. Oakland: University of California Agriculture and Natural Resources Publication 8302.
- Reddy K. R., Brien Henry W., Seepaul R., Lokhande S., Gajanayake B., Brand D. 2013. Exogenous Application of Glycinebetaine Facilitates Maize (*Zea mays* L.) Growth under Water Deficit Conditions. *Am J Exp Agric* 3: 1-13.
- Volenec JJ., Cherney JH. 1990. Yield components, morphology, and forage quality of multifoliolate alfalfa phenotypes. *Crop Sci.* 30:1234–1238.
- Wang H., Liu L., Zhou D. 2012. Effects of soil water deficit on seedlings of different *Medicago falcata* L. populations. *Afr J Agric Resh* 7:3228-3236.
- Wang W.B., Kim Y.H., Lee H.S., Kim K.Y., Deng X.P., Kwak S.S. 2009. Analysis of antioxidant enzyme activity during germination of alfalfa under salt and drought stresses. *Plant Physiol Biochem* 4:570-577.