CONTRIBUTION TO STUDY OF *ACACIA TORTILIS SUBSP RADDIANA* SEED GERMINATION, AND GROWTH SEEDLING UNDER DIFFERENT OSMOTIC AND DROUGHT CONSTRAINTS

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Abstract. *Acacia raddiana* or *Acacia tortilis* (Forsk) Hyane subsp. raddiana (Savi) Bernan, (Fabaceae) is characterized by a great ecological plasticity. It is largely used by the local populations as medicinal plants, fodder, wood energy, coal because it high calorific value. This plant thus plays a big role in rural economy because it is also appreciated in craft industry and for the clothes, tools and kitchen utensil manufacture. The study of the intensity osmotic stress effect on the germination of Acacia raddiana seeds reveals that there is a very strong negative correlation between the increase of the concentration of PEG and the germination rate (r = -0.9251). The fact remains that in spite of very small negative potential, Acacia raddiana seeds are very resistant since it coming to sprout at potential matching -6.0 MPa. In otherwise, the foliar RWC of seedling remained stable during the 25 days of treatment at about 81.61 ± 7.61% (control) and dropped from 17th to 25th day at 32% (stressed). The growth was also affected by stress. Thus, both leaves number and stem height is reduced in stressed plants compared to controls.

Keywords: *Acacia raddian;* Drought stress; Osmotic stress; Germination; Growth.

INTRODUCTION

The desertification represents a major cause of the loss of the vegetable cover in the meadow-saharan areas which are dry ones with a very weak and irregular pluviometry coinciding with very high rates of evaporation [1]. It is in the context of analysis of fight against the desertification means that we were interested in a particularly resistant desert species *Acacia tortilis subsp raddiana,* also named *Acacia raddiana.*

This species is the more abundant and widespread leguminous woody in Algerian desert belt [2]. It is a woody species well adapted to desert zone which the deep rooting allows fixation of sandy dune and promotes water infiltration and the redistribution of nutritive elements in soil. It permits to create and to keep a microclimate and ecological niches favourable to the herbaceous strata and telluric micro flora by its windbreak effect, its litter, its shadow and its root exudations [3],[4].

It is well adapted to disturbances such as drought, fire, browsing and pollarding. In spite of its ecological importance and wide distribution, knowledge about its long-term dynamics, including essential life historical characteristics such as longevity and growth is poor.

The study of seed germination and monitoring of growth parameters of young seedling plants under stress and non stress condition able us to understand this species ability to regenerate plants, particularly in case of *Acacia raddiana* which represents a keystone species growing across arid ecosystems in Africa and the Middle East, from moist savannas to hyper-arid deserts.

MATERIAL AND METHOD

Acacia raddiana seeds were brought from The National Institute of Forrest Research (INRF) - Station of Tamanrasset, Algeria.

Germination studies: Scarified *Acacia raddiana* seeds were germinated in 9 cm Petri dishes fitted with two layers of Whatman No.1 filter paper moistened initially with 4mL distilled water or treatment solution. Osmotic stress was applied using different PEG 6000 solution with an osmotic potential solution of 0, -2,-4,-6,-8 and -10 Bars. Each treatment consisted of three replicates with 20 seeds in each. Seeds were incubated under constant dark. The Petri dishes, for constant dark conditions, were kept in a lightproof box at 29±2°C. Germination (2 mm protrusion of radicle) was scored daily during eight days according to [5].

RWC and growth parameters, Experiments were performed in a greenhouse under controlled conditions. (photoperiod:16 h; mean temperature and relative humidity: 29±1°C. day and 16±1°C. night; 45% respectively).Drought stress was applied by stop watering after six weeks culture. The whole of the plants was divided into two batches, a batch of pilot seedlings which was regularly sprinkled every two days and a batch of stressed seedlings. The plants were harvested at the 1st, 7th, 12th, 17th, 22nd, and 25th of stop watering, on which measurements were carried out.

In addition to fresh weight (FW), dry weight (DW) and turgescent weight (TW) the relative water content (RWC) was estimated using the equation $RWC\% = [(FW - DW)/(TW - DW)] \times 100$.[6]

The height of stems and leave numbers were measured for each bach of plants (controls and stressed). The measures were carried on ten (10) plants for each bach at the 1st, 7th, 12th, 17th, 22nd and 25th day of stop watering.

Statistical analysis, One way ANOVA and Post Hoc Tukey's test for significant differences at $\alpha<0.05$ were performed using STATISTICA® Software 6.0

RESULTS AND DISCUSSION

After studying the *Acacia raddiana* kinetic seeds germination (Fig. 1A) under normal and osmotic stress conditions we noticed that seeds treated with distilled water presented three phases: latency, exponential, and stationary when seeds reach the maximum of germination. In normal watered conditions seeds reached maximum germination after three days (81.66%), this rate remains unchanged until the 8th day.

Seeds treated sprayed with PEG solutions -2 and -4 Bars, respectively, the beginning of germination was very slow with a lag phase of 2 days and reached 81.66% and 60% germination rate respectively. For -6 Bars PEG solution, the seeds have started their germination at the third day of incubation and reached a maximum of 28.33%. For PEG -8 and -10 treatments, we noticed a very low germination rate for the first, which reached 8.33% at the end of eight days with 5 days of lag phase and we recorded no germinated seed for the second.

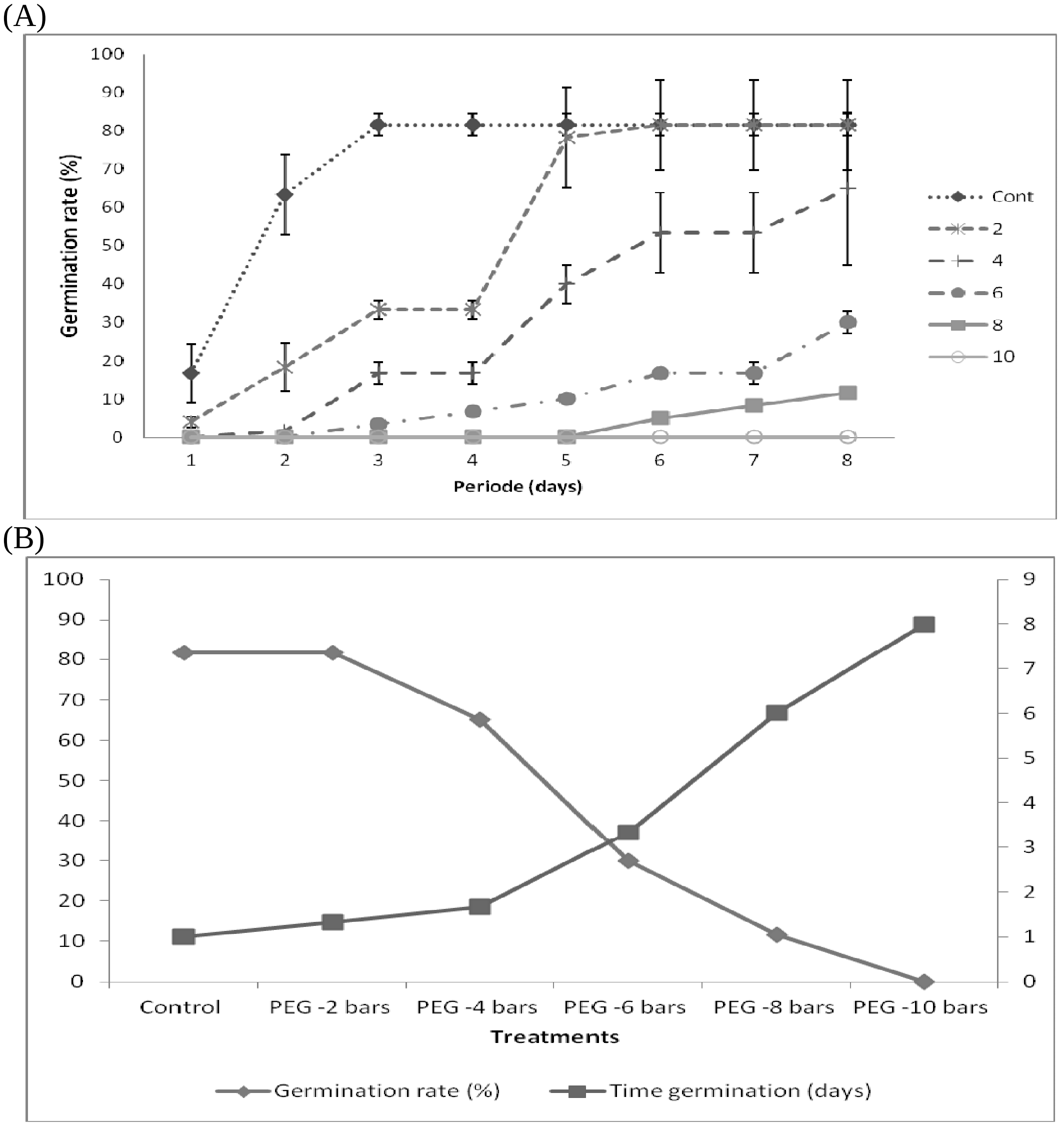

Fig. 1. (A) *Acacia raddiana* kinetic seed germination under normal and osmotic stress conditions; Distilled water (♦), PEG -2 (Δ)-, PEG -4(□),-6(●),PEG -8(■) and PEG -10(○) Bars (values are means ±SE of three repetitions) (B) Acacia raddiana seeds time and rate germination)

The study of the effect of the intensity of osmotic stress on the germination of seeds of *Acacia raddiana* reveals that there is a very strong negative correlation between the increase of the concentration of PEG and the germination rate (r = -0.9251). It remains that in spite of very low negative potential seeds of *Acacia raddiana* are very strong since it is still able to sprout in potential matching -6.0 Bars.

Table 1.
Acacia raddiana germination rates and average time germination under different osmotic treatments.

Treatements PEG en bars	Germination rate (%)	Statistical similtude	Time germination (latency phase) (Days)	Statistical similtude
Control	81,67	A	1	D
PEG -2	81,67	A	1,33	D
PEG -4	65,00	A	1,67	D
PEG -6	30,00	BC	3,33	C
PEG -8	11,67	C	6	B
PEG -10	0,00	C	8	A

Note: Different letters denote significant differences (Tukey test, $p < 0.05$)

Table 1 and Fig. 1.B show that osmotic stress intensity had affected both maximum rate germination that decrease among the increase of solution pressure and inversely time germination that increases as well as the pressure increases. Our results are close to those reported by [5].

Acacia tortilis ssp. *raddiana* is a crucial element in his original group in the balance and maintenance of many arid and desert ecosystems. The introduction of this species in reforestation programs offer a solution for sustainable reforestation in arid and semi-arid areas but also those affected by and would therefore diversified operations[5]. However, the success of phases of germination and growth of this kind inevitably involves a good knowledge of its germ and developmental characteristics and its behaviour against environmental conditions. [5]

The *Acacia raddiana* germinative capacity was widely affected by drought (here simulated by PEG). In fact, both germination capacity and germination were affected. Time germination was longer as well as the stress intensity increases. These results show that *Acacia raddiana* seeds are moderately tolerant to osmotic stress (rate and germination time varied significantly as a result of osmotic stress). The germination capacity at water potential of -8 bars, is practically zero. Our results confirm the results of [5] and [7] who noticed that in *Acacia raddiana* germination stop almost -7 bars. In contrast, [8]reported that when the seeds of *Acacia tortilis* and *Acacia senegal* are subjected to simulated water stress by adding polyethylene glycol (PEG) to water imbibition, germination is significantly lower than controls (germination in absence of osmoticum) for less than -1.8 MPa (-18 Bars) water potential. If the pressure equal to -2.1 MPa (-21 Bars), seeds still germinate about quarter. The potential limit value to which all of seed cannot germ is not beyond -6 bars, the maximum germination rate decreases and the average germination time increases over the osmotic pressure. These results are consistent with what has reported by [9]

According to [10], delayed germination can also be explained by the time necessary for the seed to put in place mechanisms that allow it to adjust its internal osmotic potential relative to the environment. Our study as well as [5] one showed that *Acacia raddiana* is moderately demanding water in germination stage, but this doesn’t mean that tolerant species to water stress during germination are those that are more adapted to drought at the adult stage [11]. In this sense [8] confirm that *Acacia tortilis* subsp *raddiana* is an advantage to the adult stage for its tolerance to water stress because of branching and the rapid roots development at the stage of growth.

Indeed, although it is one of the important factors in the establishing of species [12],[13], tolerance water stress at the time of germination that is the first phase of the growth cycle, according to the following conditions can be an advantage or a disadvantage [14]. Results research related to the effects of water stress on germination show that it is difficult to link the tolerance to water stress, at germination time to the ecology of the same species [15]. This result also noted by [14] for some Sahelian species, lets say that the resistance to water deficit in germination phase is not the predominant criterion of the ecological distribution of taxa. [16] joining [17] and [18]argue that the ability to germinate under conditions of water and salt stress is not necessarily representative of the ecology of the adult plant. [19]

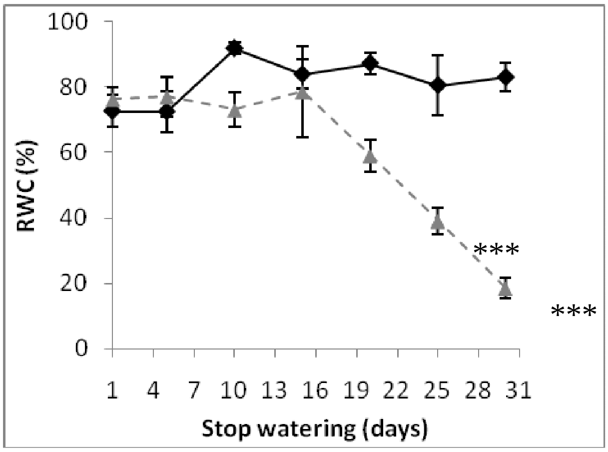

Fig. 2. *Acacia raddiana* Relative water content (RWC) under normal (♦) and stress condition(▲) among 25 days of drought (values are means ±SE of three repetitions)
* Significant difference with control at * P<0.05, ** P<0.01 or *** P<0.001 by Tukey multiple test

In otherwise, the RWC of young Acacia raddiana seedling remains relatively stable during 17 days of treatment as well as in stressed plants then in irrigated ones and start decreasing at the 22^{th} day of treatment to reach 38.05±1.63 % at the 25^{th} day (Fig. 2).

Stem height in control plants reached 45.55 ± 8.45cm after 9 weeks of growth. Stressed plants in turn reached 34.4 ± 6.0 cm in the same period of growth with 25 days of stress. In stressed plants, we see the installation of a plate from the 17th to the 25th day stop watering. So between the 1st and 17th days there was a steady increase in growth (0.44 cm / day). In control plants growth remained constant and continuous with an increase in the height of 1.09cm/d for 25 days. (Fig.3A).

Regarding the leaves number, they reached 34.1 ± 6.52 units in controls and 23.4 ± 2.63 units in stressed plants after 25 days of stress. In control plants among 25 days, the speed leave number apparition increase reached 0.58 unit / day and continued to grow. For stressed lot, the leave apparition was lower (0.28 units / day in 17 days) and stops from the 17th day were we noticed a decrease due to wilting and total drying of the leaves (Fig.3B).

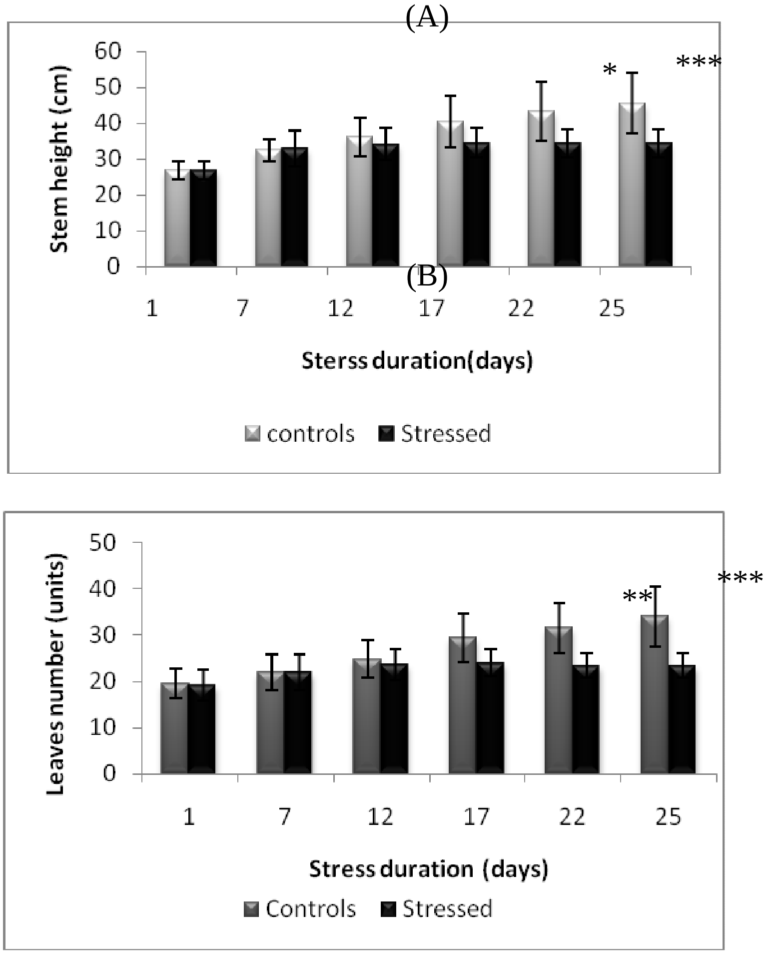

Fig. 3. *Acacia raddiana* growth parameters (A) Stem height (B) Leaves number under normal (□)and stress condition(■) among 25 days of drought (values are means ±SE of ten repetitions)

* Significant difference with control at * P<0.05, ** P<0.01 or *** P<0.001 by Tukey multiple test

In fact, the study of *Acacia raddiana* young seedling growth parameters in relation with plant water status able us to show that stressed plants can resist to drought among 17 days treatment. They were able to maintain turgid and growth was not affected as well as in stem height and number of leaves appearing comparatively to well watered plants. Among this period we have noticed an accumulation of some compatibles solutes that able plant to maintain its turgor (results not shown). According to [20] osmotic adjustment is beneficial for increasing crop yields under water-deficit conditions, experimental data offer little supporting evidence. This is the case of wheat [21],[22],[23]and Sorghum [24]. After 17 d. a significant difference appeared between controls and stressed plants where results shows a reduction in growth and a decrease in RWC till the wilting at the 25th d of treatment. One of the first responses to the abiotic stress is a reduction in plant growth. As the drought period is getting longer, most of plants reduce their shoot growth, while the elongation of roots is usually stimulated, which permits deeper soil penetration in search for water [25].Generally, when water is not restricting growth, plants invest a considerable fraction of photoassimilates in the expansion of photosynthetic tissues, maximising light interception and, as a consequence, growth [26] and [27]. Photosynthesis and growth (biomass production) are the primary processes to be affected by drought [28]. Here, *Acacia raddiana*, young seedling seems to adopt the same way to cope with drought stress by reducing its growth. Our results are close to those reported by [29] where growth reduction was observed under moderate and severe stress. This suggests that even at reduced soil water availability *Acacia raddiana* plants are able to grow. This agree with the findings of [30] who have observed that water withhold would arrest growth but maintaining plants at low soil water content (40%) would allow them to continue growing, although at a slower rate than fully irrigated plants.

CONCLUSIONS

In spite to be affected by osmotic stress, *Acacia raddiana* seeds were able to germinate in stress conditions when water potential of the solution was near to -6 bars. These results suggest that, *A. raddiana* could easily germinate under drastic conditions in arid or semi-arid areas. In otherwise, after analysis of tolerance of young seedling to drought, we noticed that *Acacia raddiana* is able to maintain a normal turgor and growth during the two first weeks of stress. These preliminary results deserve to be complete by a larger study on the physiological behaviour of *Acacia raddiana* in stress conditions including osmotic adjustment and biochemical responses that permit this plant to cope with water stress. The comprehension of those mechanisms will able us to determine if *Acacia raddiana* could be a real advantageous element in the rehabilitation of the ecosystems particularly in deprived areas.

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