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ORIGINAL PAPER

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EVALUATION OF DROUGHT RESISTANCE IN *Nitraria schoberi* AS A NATIVE PLANT BY IRRIGATION INTERVALS FOR APPLYING IN ARID URBAN LANDSCAPE

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ABSTRACT

Climate change as a global phenomenon manifested mainly through global warming and water scarcity in the world. Using of native drought resistant plants in the urban landscape of arid and semi-arid regions can help water resources management. We conducted an experiment to investigate drought resistance of a native plant (*Nitraria schoberi*) in Iran by three irrigation intervals (each 2, 4 and 6 days). Data collection was performed in three stages. Our results indicated that irrigation intervals did not affect significantly the chlorophyll a and b content and ratio of chlorophyll a/b. Generally with increasing in irrigation intervals, carotenoid, proline and sugar content and also RWC were increased in during experiment. It is concluded that due to increase of proline and sugar content with increasing of irrigation interval, *Nitraria schoberi* is a drought tolerant plant which can be used in urban landscape of arid and semi-arid regions.

Key words: drought tolerance, irrigation, native plant, osmo-protectants

INTRODUCTION

Water scarcity is a critical issue that affects more than 40 per cent of the global population and is projected to rise. The world's most water stressed region is the Middle East with averages of 1200 cubic meters of water per person [Tropp and Jägerskog 2006]. Desertification, urban growth, and economic restructuring are making water increasingly scarce and water access increasingly inequitable in cities around the world.

Landscape is a vital component of the urban environment that can provide environmental, human health, psycho-social and economic benefits [Frank 2003]; it is also a large user of water [Lazarova et al. 2005]. In arid and semi-arid areas to reduce water consumption in urban landscape and also for developing it, some researchers have suggested the use of native ornamental plants in urban planting design [Simmons et al. 2011, Baltzoi et al. 2015].

Drought is a major abiotic stress affecting the majority of the world's plants. Drought-resistant plants can survive extended periods with little precipitation or irrigation, but this does not mean they can provide acceptable landscape function and performance with little water. Plants respond to survive under water-deficit conditions via a series of physiological, cellular, and molecular processes culminat-

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ing in stress tolerance [Shinozaki and Yamaghuchi--Shinozaki 2007].

During water deficit, many physiological and biochemical processes are disturbed Drought-tolerant plants in comparison with the drought-sensitive plants often accumulate the high proline and malondialdehyde levels [Sun et al. 2013, Chaitanya et al. 2009] and the low chlorophyll and sugar contents [Sofo et al. 2004, Parida et al. 2007, Romero et al. 2013]. Relative water content (RWC) is an important indicator of water status in plants; it reflects the balance between water supply to the leaf tissue and transpiration rate [Lugojan and Ciulca 2011]. Water stress can induce significant changes in relative water content [Zlatko Stoyanov 2005, Ganji et al. 2012].

Using native plants can be a smart option in landscape design because native plants are adapted to local soils and climate conditions, they generally require less watering and fertilizing than non-natives. Natives are often more resistant to insects and disease as well, and so are less likely to need pesticides. Using native plants helps preserve the balance and beauty of natural ecosystems [Britt et al. 2003]. Due to exposure to scarce and saline water resources, native plants in arid and desert areas are mostly drought resistant. Some studies show that in the regions, native plants compare with the non-natives requires minimal watering [García-Navarro et al. 2004, Simmons et al. 2011].

Recent trends in landscaping and gardening under semi-arid conditions include the use of drought-adapted trees and shrubs in low water-use landscape designs [Franco et al. 2006]. To manage of drought stress in landscape plants, the adoption of water-wise cultivation such as deficit irrigation or reduced irrigation water augmented with drought-tolerant crops to cope with escalating water scarcity problems [Álvarez and Sánchez-Blanco 2011, 2013].

Nitraria schoberi L. is an evergreen shrub that called Ghar-e-Dagh in Persian that belongs to family Nitrariaceae. *Nitraria schoberi* is widely distributed all over Middle East like arid and semi-arid areas of Iran. As *Nitraria schoberi* grows in arid areas it seems to be a drought-resistant shrub [Naseri 2014]. The purpose of this work was to study the drought resistant of *Nitraria schober*i as a native plant in Iran under different irrigation intervals. Relative water content, proline level, sugar and chlorophyll content were measured to evaluate the changes that take place in plants exposed to drought condition and whether these changes lead to drought resistance in the plant.

METHODS AND MATERIALS

Plant material and experimental conditions

The plant seeds were provided from Pakan Bazr that is a reputable seed company in the field of collection and production of native and pasture seeds in Iran-Isfahan. Before sowing, the seeds were stratified (4 weeks in 4°C). Seedlings about 40 days old (5 cm height), were transplanted into the fields. Plant spacing was 1.5 m between rows and between plants in the row. This study was carried out at the Research Center for Plant Sciences, Ferdowsi University of Mashhad, during 2015 summer season.

Experimental design and treatments

The plants were regularly irrigated with tap water for 10 days. Then the following three irrigation intervals were applied during summer (three months): every 3 days, every 6 days and every 9 days (with the same water quantity 2 liters per plant). The experiment was conducted in completely randomized design (3 treatments × 5 replications).

Recorded data

The data were recorded three times (three stages) with one month interval (except RWC which was only measured during the third month at the end of the experiment). The following parameters measured and calculated were relative water content (RWC), proline and chlorophyll content and also total carbohydrates. In order to calculate RWC, leaf fresh weight (FW) samples were weighed, then they placed in distilled water at 4°C in the dark for 24 h to rehydrate and the turgid weight (TW) measured. Finally were direct at 75°C for 24 h and the dry weight (DW) determined. The RWC was calculated using the following formula [Silveira et al. 2003]:

$$RWC = (FW – DW)/(TW – DW) \times 100.$$

To calculate of chlorophyll and carotenoid, 50 mg of fresh leaf was extracted in 10 ml 80% acetone. Absorbance values were determined on a spectrophotometer (UV-VIS, Optima SP-3000 Plus, Bratislava, Slovakia) at 663, 645 and 470 nm, for chlorophyll a, b and carotenoid [Arnon 1946]. Chlorophylls and carotenoid were calculated using the following equations:

$$\text{Chlorophyll a } (\mu g \cdot ml^{-1}) = 12.7\ (A_{663}) - 2.69\ (A_{645})$$

$$\text{Chlorophyll b } (\mu g \cdot ml^{-1}) = 22.9\ (A_{645}) - 4.68\ (A_{663})$$

$$\text{Carotenoide } (\mu g \cdot ml^{-1}) = \\ = 1000\ A470\text{-}1.82\ \text{chl a}\text{-}85.02\ \text{chl b}/198$$

To determine the proline content was used the Troll and Lindsley [1955] technique, modified by Dreier and Goring [1974]. 100 mg of fresh leaf material was homogenized in 2 ml of 40% methanol, and then heated in a water bath at 85°C for 60 min. Absorbance values at wavelength 528 nm using a spectrophotometer (UV-VIS, Optima SP-3000 Plus, Bratislava, Slovakia). Total carbohydrates were measured in 100 mg of powder of dried leaves, which were soaked in 80% (v/v) ethanol for 24 h was determined by the method of Shields and Burnet [1960]. The absorbance was measured at 585 nm in a spectrophotometer (UV-VIS, Optima SP-3000 Plus, Bratislava, Slovakia). Concentration of Chlorophyll, carotenoid, proline and sugar were given as mg per gram dry weights (mg/g DW). The data were subjected to Analysis of variance (ANOVA-One Way) and mean values were compared with Duncan's multiple range test ($p < 0.05$) using Minitab 17 software.

RESULTS AND DISCUSSION

Chlorophyll content

As can be seen in the Figure 1, the trend of chlorophyll a and b changes in irrigation intervals (2, 4 and 6 days) and during the three stages was inconsistent. The analysis of chlorophyll (a) in the leaves showed averagely no significant differences among irrigation intervals during three stages (three months). In the first stage (first month), irrigation interval of every 4 day and in the third stage (third month) irrigation interval of every 6 day were significantly lower, while in the second stage (second month) irrigation interval of every 4 day was significantly higher. In the first and third months and also in the average of stages, chlorophyll (b) was significantly lower in irrigation interval of every 4 day compared to every 2 and 6 days. In the first month, ratio of chlorophyll a/b was not significantly different among of irrigation intervals. Chlorophyll a/b ratio, in irrigation interval of every 4 day showed a significant increase than irrigation interval of every 2 and 6 days in the second and third stages and also in the average of stages.

Chlorophyll a, b and a/b ratio of leaves are widely used to determine the general state of the photosynthetic system [Dai et al. 2009]. fbgSome studies indicated that chlorophyll content was decreased in the drought stress conditions [Kuroda et al. 1990]. In our study irrigation intervals had not a strong effect on the chlorophyll content. This may indicate that the intervals between irrigation (minimum 2 days and maximum 4 days) have not been adequate to change the chlorophyll content.

In the second stage and also in the average of stages, the carotenoide value significantly was increased in irrigation intervals of every 4 days than every 2 and 6 days. In the first and second stages, the significant decrease in carotenoide was obtained in irrigation interval of every 2 day while in the third stage it was observed in irrigation interval of every 6 day (fig. 2). Carotenoids as pigments in plants play several roles in photosynthesis and also in the mechanisms of stress tolerance [Gill and Tuteja 2010]. The high level of carotenoids in drought condition has been reported by Abbasi et al. [2014], Deng et al. [2003]. The findings of the present study indicated that almost with increasing in the irrigation interval, carotenoid level was increased.

Proline content

The results indicated that in the second and third stages and also in the average of stages, with increasing of irrigation interval, proline content was significantly increased. In the first stage, no significant differences were found between irrigation interval of every 2 and 4 day while the highest proline content

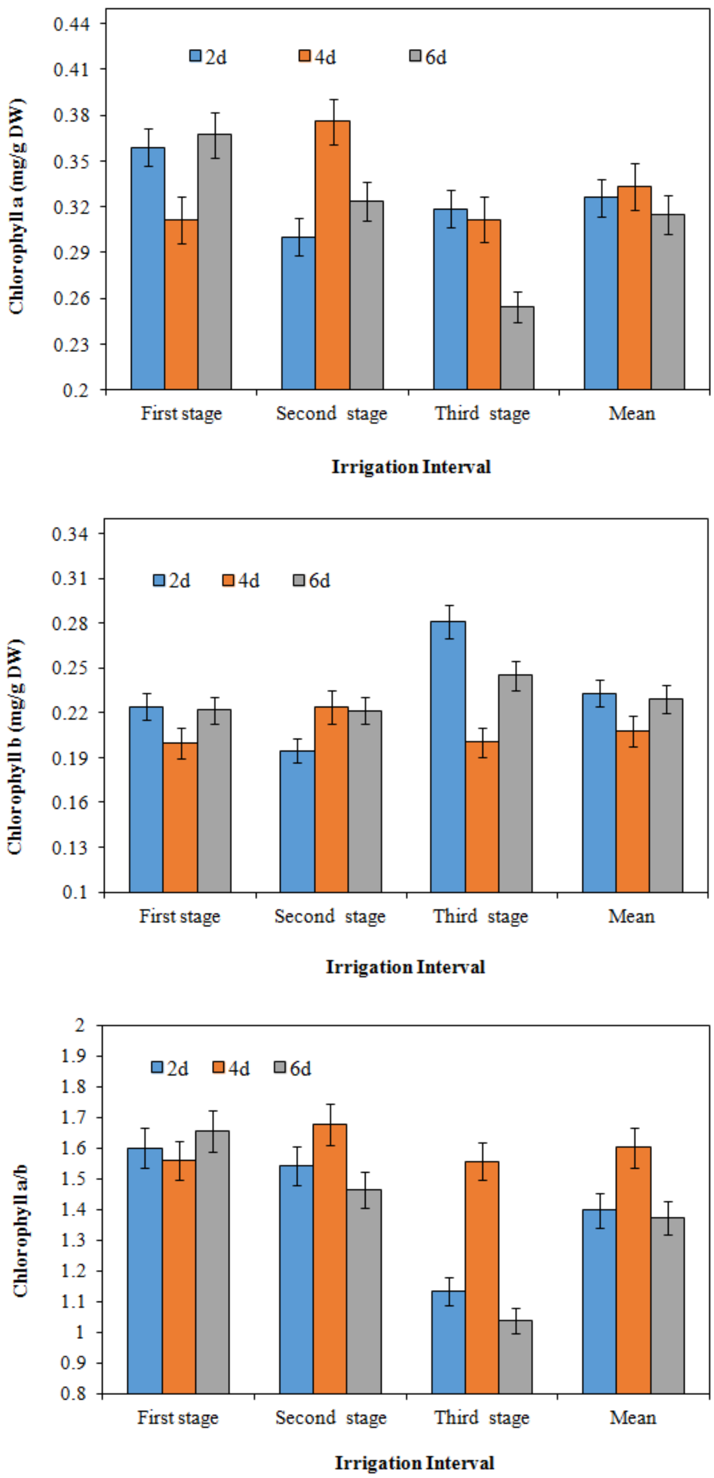

Fig. 1. The effects of irrigation intervals (each 2, 4 and 6 days) on the chlorophyll a, b and a/b during three stage

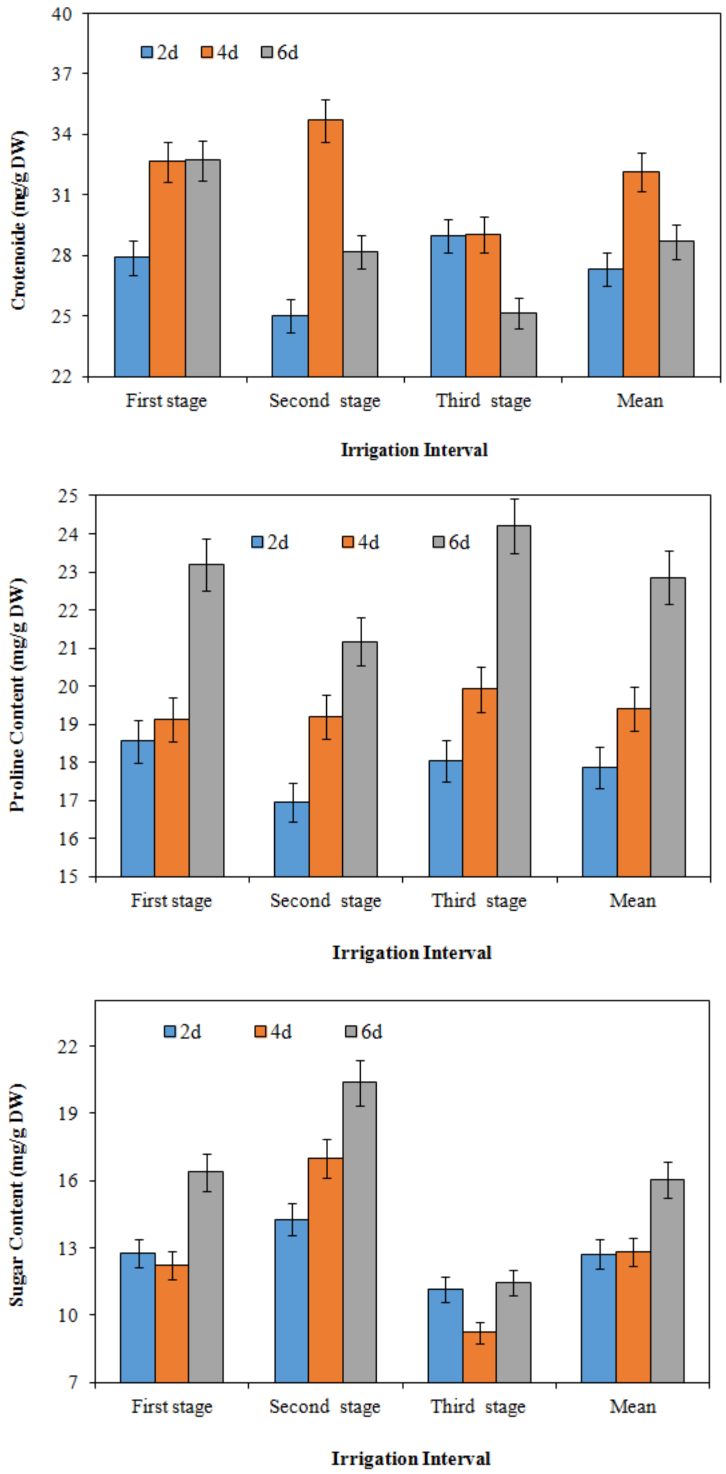

Fig. 2. The effects of irrigation intervals (each 2, 4 and 6 days) on the carotenoid, proline and sugar content during three stages

was obtained in irrigation interval of every 6 day (fig. 2). Some studies show that drought-tolerant plants than the drought-sensitive, accumulate the more level of proline [Chaitanya et al. 2009, Sun et al. 2013]. In this study, increase of irrigation interval had a major effect in increasing the proline content.The accumulation of proline plays an adaptive role [Verbruggen and Hermans 2008] and the main strategy of plants for avoiding detrimental effects. It is the key adaptations for successful growth under acute water stress [Akram et al. 2007]. Proline can also protect cells damage due to low water potential of cells [Krazsenky and Jonak 2012]. In the present study we found that *Nitraria schoberi* accumulated high proline levels in response to water stress.

Sugar content

As depicted in Figure 2, in all stages the highest sugar content was obtained in irrigation intervals of every 6 day. In the second stage with increasing of irrigation interval, the sugar content was significantly raised but it was affected only by irrigation intervals of every 6 day in the first stage. In the third stage, sugar content was significantly decreased in irrigation intervals of every 6 day.

One of the mechanisms for tolerance to water-deficit stress in plants is the accumulation of osmoprotectants such as sugar. The drought tolerance in plants is strongly correlated to the accumulation of soluble sugars [Parida et al. 2007, Romero et al. 2013]. In our study, the increase of irrigation intervals was nearly associated with the increase of sugar content. Therefore, it seems that the sugar accumulation was correlated with drought tolerance and our results were consistent with the mentioned studies.

Relative water content (RWC)

Increasing irrigation interval from each 2 and 4 day to each 6 day had significant increase on RWC. The relative water content (RWC) parameter can be used to screen of plants drought tolerance (fig. 3).

RWC as an important indicator of the degree of cell and tissue hydration plays a key role for optimum physiological functioning and growth processes. There are conflicting results regarding the effect of water stress on the RWC. Some studies indicated that drought-tolerant plant species keep high RWC compared with drought-sensitive species [Marcelo et al. 2007]. In the present study, in the longest irrigation interval (each 6 days) RWC was increased that the results almost are in agreement with those reported by Colom and Vazzana [2003] and also by Marcelo et al. [2007].

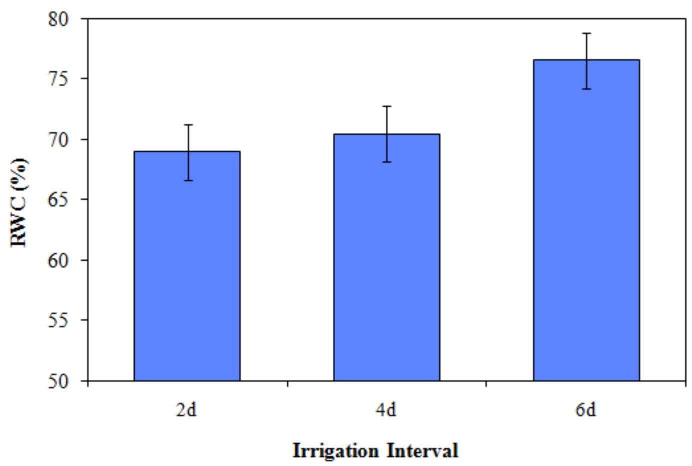

Fig. 3. The effects of irrigation intervals (each 2, 4 and 6 days) on RWC (%) during third stage

Drought stress triggers various plant reactions, ranging from cellular metabolism to changes in growth rates and the biochemical and physiological responses. Increase in proline and sugar level and relative water content and decrease of chlorophyll concentration are considered as drought tolerance mechanisms [Colom and Vazzana 2003, Parida et al. 2007, Krazsenky and Jonak 2012]. In the present study due to increase of proline and sugar content with increasing of irrigation interval, it can be said that *Nitraria schoberi* is a drought tolerant plant.

CONCLUSION

In the present study that the irrigation intervals was investigated during the early growth stages of *Nitraria schoberi*, can be said that nearly with increasing in irrigation interval, carotenoid, proline and sugar content and also RWC were increased. It show that increase in value of proline and sugar in response to increasing irrigation intervals can be considered as a mechanism of drought resistance and *Nitraria schoberi* is a drought tolerant plant. The plant was able to control the water deficit stress by stimulating proline production and sugar accumulation. Since the chlorophyll content averagely was not affected by irrigation intervals and also RWC was increased (third stage) during the experiment, may be claimed that with increasing irrigation interval to 6 days in the early growth stages of the plant, greenery and freshness of the plant has not diminished. It is concluded that *Nitraria schoberi* can apply in the landscape of arid and semi-arid regions such as Mashhad and most cities of Iran.

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