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Effect of Irrigation Water Regimes on Yield of *Tetragonia Tetragonioides*

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Abstract: The main purpose of this experiment was to study the effect of several irrigation water regimes on *Tetragonia tetragonioides* (Pall) O. Kuntze in semi-arid regions. During the experiment period, it was measured that several irrigation regimes were affected in terms of growth, biomass production, total yield, mineral composition, and photosynthetic pigments. The experiment was conducted in the greenhouse at the University of Algarve (Portugal). The study lasted from February to April in 2010. Three irrigation treatments were based on replenishing the 0.25-m-deep pots to field capacity when the soil water level was dropped to 70% (T1, wet treatment), 50% (T2, medium treatment), and 30% (T3, dry treatment) of the available water capacity. The obtained results showed that the leaf mineral compositions of chloride and sodium, the main responsible ions for soil salinization and alkalization in arid and semi-arid regions, enhanced with the decrease in soil water content. However, the minimum amounts of chlorophyll, carotenoids, and soluble carbohydrates in the leaf content were obtained in the medium and driest treatments. On the other hand, growth differences among the several irrigation regimes were very low, and the crop yield increased in the dry treatment compared to the medium treatment; thus, the high capacity of salt-removing species suggested an advantage of its cultivation under dry conditions.

Keywords: irrigation water regimes; leaf mineral composition; semi-arid regions; available water capacity; biomass production; total yield

1. Introduction

In arid and semi-arid regions, such as the Mediterranean, supplies of good-quality water allocated to agriculture are expected to decrease because most available fresh/potable water resources were already mobilized [1]. According to the Food and Agriculture Organization (FAO) [2], due to the shortage of water, there is an enlargement of saline land in agricultural areas in some developing countries. As a result, yield is decreasing, provoking an increasing cost of agricultural products [3].

Soil salinization is recognized worldwide as being among the most important problems for crop production in arid and semi-arid regions [4]. Water deficit and salinity are the major limiting factors for plant productivity, affecting more than 10% of arable land on our planet, resulting in a yield reduction of more than 50% for most major crop plants [5]. The usually noted abiotic stresses that include a component of cellular water deficit are salinity and low temperature; stresses can also severely limit crop production [6]. Abiotic stresses, such as drought, salinity, extreme temperatures, chemical toxicity, and oxidative stress are serious threats to agriculture, and result in the deterioration of the environment. Abiotic stress is the primary cause of crop loss [7,8]. This problem is intensified in coastal areas due to sea-water intrusion. This results from reduced ground-water levels as the water demand exceeds the annual groundwater recharge [9]. As reported above, some of the emerging regions in risk of increasing levels of salinization of their soils are located in the Mediterranean Basin [10,11], Australia [12], Central Asia [13], and Northern Africa [14]. Salinity is one of the rising problems causing tremendous yield losses in many regions of the world, especially in arid and semi-arid regions. The use of halophytes can be an effective way of accumulating the salt in soil [15].

Intensive irrigation of agricultural crops with a high level of water mineralization causes salts to accumulate in the root zones, which adversely affects the crop productivity. In order to reduce such negative impacts, a regulated deficit irrigation (RDI) technique was adopted to combat salinization in arid and semi-arid environments by reducing the water application during certain growth stages of the crops [16].

When RDI is not feasible, halophyte crops might be a solution for the salinization of agricultural land. These crops can be irrigated by, for example, seawater, salt-contaminated phreatic sheets, brackish water, wastewater, or drainage water from other plantations [17,18].

Hence, our aims were to choose a salt-removing crop, tolerant to salinity, along with interest as a food crop, and to test its drought tolerance through its response to several water regimes. *Tetragonia tetragonioides* was the selected crop. In a previous experiment, its capability as a high biomass horticultural leaf crop was demonstrated, producing a plant dry weight of 40,000–50,000 dry mass (DM) $kg \cdot ha^{-1}$ if the plant population density is around 75,000 $plants \cdot ha^{-1}$ [19].

2. Materials and Methods

2.1. Experimental Procedure

The experimental work was conducted in the greenhouse of Horto at the University of Algarve, Faro, Portugal (37°2′37.1 N 7°58′30.8 W), from February to April in 2010. The salt-removing species *T. tetrogonioides* was selected. Plants were transplanted to 7-L pots when they had four leaves (10 February). The number of plants per pot was three, with four replications. The species were irrigated with tap water every three days until the beginning of the treatments (1 February–8 March). A nitrogen fertigation treatment was started on 8 March, with daily applied concentrations of 2 mM NO_3^- and 2 mM NH_4^+ as the cumulative amount of NO_3NH_4 ($g \cdot plant^{-1}$) to the end of the experimental studies (22 April). The electrical conductivity (EC_w) of irrigation water was 0.6 $dS \cdot m^{-1}$ and pH 7.

The treatments consisted of three irrigation regimes in a randomized complete block design with three replicated treatments based on replenishing the 0.25-m-deep pots to field capacity when the soil water level dropped to 70% (T1, wet treatment), 50% (T2, medium treatment), and 30% (T3, dry treatment) of the available water capacity (aw). This concept was developed by Reference [20], where "aw" is the range of available water that can be stored in soil and is available for growing crops. It was assumed by the same authors that the soil water content readily available to plants (θ_{aw}) is the difference between the volume of water content at field capacity (θ_{fc}) and at the permanent wilting point (θ_{wp}), calculated as follows:

$$\theta_{aw} = \theta_{fc} - \theta_{wp} \quad (1)$$

The watering volume was estimated to replenish the soil profile to field capacity at a depth of 0.25 m. The volumetric soil water content (m^3 water/m^3 soil; $m^3 \cdot m^{-3}$) was determined just before the water application.

To control soil water along the soil profile, the irrigation frequency, and the water amount, the pots were weighed every day. The soil water content was monitored periodically, gravimetrically measured for a depth of 0.00–0.25 m.

The plants were harvested destructively (26 April), washed in water, and dried with paper towels. Then, the fresh weight (FW) was measured. The fresh samples were dried in a forced drought oven at 70 °C for 48 h, and the dry weight (DW) was measured. Plant materials were collected for chemical analyses. The electrical conductivity (EC_s) and pH of soil were measured before and after the experiment.

2.2. Growth and Chemical Analysis

During the vegetation period, the stem length was measured, as well as the number of nodes and number of leaves of *T. tetragonioides* every seven days.

The plants' leaves were analyzed on total growth and mineral compositions (Na, Cl, N, K, P, Ca, and Mg). Dried leaves and stems were finally grounded and analyzed using the dry-ash method. The levels of Na and K were determined using a flame photometer, and the remaining cations (Na, K, Ca and Mg) were assessed by atomic absorption spectrometry. Chloride ions were determined in the aqueous extract by titration with silver nitrate according to the method of Reference [21]. Plant nitrogen (N) content was determined using the Kjeldhal method. Phosphorus was determined using the colorimetric method according to the vanadate–molybdate method. All mineral analyses were only performed on the leaves.

The analysis of pigments was done with a disc size of 0.66 cm and a total area of 1.37 cm^2. For sugars. there were ten discs, with a disc size of 0.66 cm and a total area of 3.42 cm^2. The amount of photosynthetic pigments (chlorophyll a (Chla), b (Chlb), total (ChlT), and carotenoids) was determined according to the method of Reference [22]. Shoot samples (0.25 g) were homogenized in acetone (80%). The extract was centrifuged at 3000× *g*, and absorbance was recorded at wavelengths of 646.8 and 663.2 nm for the chlorophyll assay and at 470 nm for the carotenoid assay using a Varian Cary 50 ultraviolet–visible light (UV–Vis) spectrophotometer. The levels of Chla, Chlb, ChlT, and carotenoids were calculated. Soluble sugars (glucose) in leaves were extracted as described by Reference [23]. The change in absorbance was continuously followed at 340 nm using an Anthos hat II microtiter-plate reader (AnthosLabtec Instrument, Hanau, Germany).

2.3. Statistical Analyses

Data (n = 4) were examined by one-way ANOVA. Multiple comparisons of the means of data between different salinity treatments within the plants were performed using Duncan's test at the $p < 0.05$ significance level (all tests were performed with the SPSS program version 17.0 for Windows).

2.4. Soil

Table 1 shows the soil texture and soil parameters before the experiment. According to the FAO, based on the United States Department of Agriculture (USDA) particle-size classification, the soil texture was sandy clay loam. The soil parameters show that the range in the soil's pH value was slightly alkaline and that the electrical conductivity (EC_s) was 1.1 dS·m^{-1} (non-saline soil) at 25 °C.

Table 1. Soil parameters before the experiment.

Soil Texture		Soil Parameters			
Sand (%)	58.9	Field capacity θ_{fc} ($m^3 \cdot m^{-3}$)	0.22	pH (H_2O)	7.7
Silt (%)	18	Wilting point θ_{wp} ($m^3 \cdot m^{-3}$)	0.12	EC_e* (dS·m^{-1})	1.1
Clay (%)	24.1	Available soil water θ_{aw} ($m^3 \cdot m^{-3}$)	0.12		
Classification: Sandy clay loam		Bulk density (g·cm^{-3})	1.41		

EC_e*—Electrical conductivity of the extract of a saturated soil paste (dS·m^{-1}).

Table 2 shows the volumetric soil water content (m^3 water/m^3 soil; $m^3 \cdot m^{-3}$) just before the water application. The volumetric soil water content in soil ranged between 0.20 and 0.15 $m^3 \cdot m^{-3}$.

Table 2. Volumetric soil water content (m^3 water/m^3 soil; $m^3 \cdot m^{-3}$) just before the water application.

Treatment	Determination	Θ ($m^3 \cdot m^{-3}$)
T1	$\theta_{wp} + 0.70 \times \theta_{aw}$	$\Theta_1 = 0.20$
T2	$\theta_{wp} + 0.50 \times \theta_{aw}$	$\Theta_2 = 0.17$
T3	$\theta_{wp} + 0.30 \times \theta_{aw}$	$\Theta_3 = 0.15$

2.5. Climate Condition in Greenhouse

The average climatic data during the experimental period in the greenhouse were as follows: maximal relative humidity, 88.4%; minimal relative humidity, 11.3%; maximal temperature, 45.8 °C; minimal temperature, 11.4 °C.

During the experimental period, the relative humidity of the greenhouse was increased, and the maximal temperature decreased.

3. Results and Discussion

3.1. Effect of Irrigation Water Regimes on Plant Growth

Table 3 shows the irrigation water regimes' effects on the *T. tetragonioides* growth (stem length, number of nodes, and number of leaves). A significant effect on the stem length can be seen. In the beginning of the experiment, the stem length of the crop showed very low variations between T1 and T2 treatments. During the last three weeks of the experimental period, the stem length increased showing equal differences between each treatment—T1 and T2, and T2 and T3 (Δ stem length ~0.5 cm). The number of nodes and number of leaves were also higher in treatment T1.

Table 3. Effect of irrigation water regimes on stem length, number nodes, and number of leaves of the species. Different letters within a column represent significant differences ($p \leq 0.05$).

Treatment	*Tetragonia tetragonioides*		
	Stem Length (cm)	**Number of Nodes**	**Number of Leaves**
T1	38.8 ± 1.9 a	22.5 ± 0.6 a	9.9 ± 0.58 a
T2	34.2 ± 0.5 b	18.1 ± 0.6 b	8.1 ± 0.37 b
T3	29.3 ± 1.5 c	19.2 ± 0.8 b	9.5 ± 0.22 b

3.2. Fresh (FW) and Dry (DW) Weight of Crop

The fresh weight (FW) of *T. tetragonioides* species showed low variation among treatments (Figure 1). There was a low increase of the fresh weight of stem, leaves, and seeds in treatment T1. Surprisingly, the obtained results in treatment T3 were slightly higher than in treatment T2.

The obtained results of dry matter show that the stem, leaves, and seeds of treatment T1 were slightly higher than other treatments. There was very low variation of dry matter between T2 and T3 treatments (Figure 2).

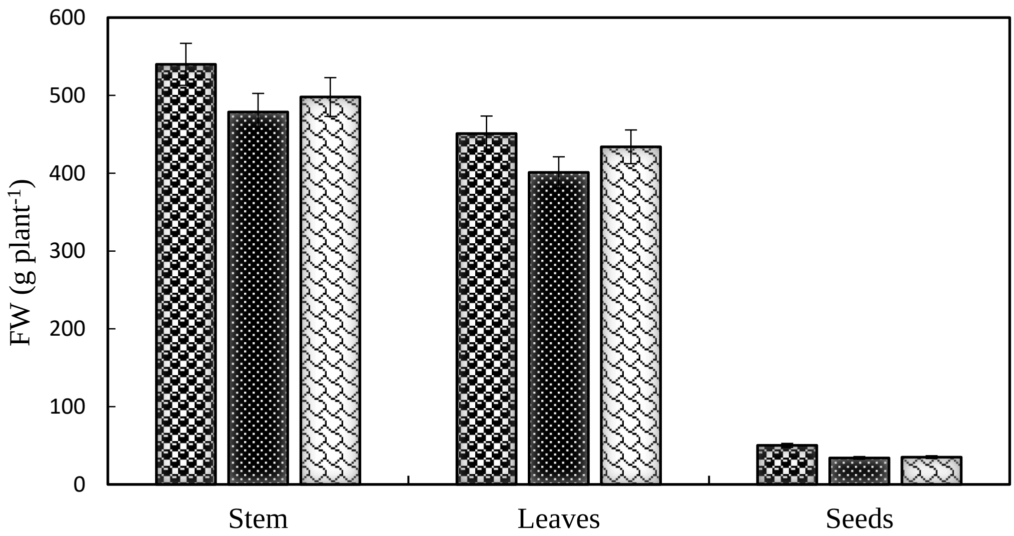

Figure 1. Fresh weight response of *Tetragonia tetragonioides* to the different irrigation treatments.

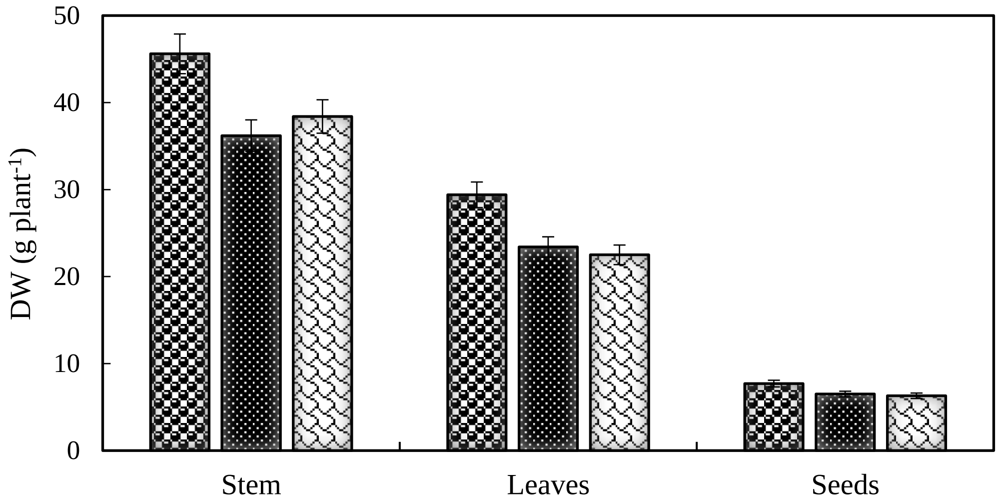

Figure 2. Dry weight response of *Tetragonia tetragonioides* to the different irrigation treatments.

3.3. Effect of Irrigation Water Regimes on Mineral Composition in Plant Leaves

Table 4 shows the effects of water application treatments on the mineral composition of *T. tetragonioides* leaves. In summary, the total nitrogen leaf content of the species showed low variation among treatments. There was an enhancement of chloride and sodium concentration with the decrease in water content. There was a general decrease in phosphorus, calcium, potassium, iron, and magnesium in the leaf content under drought conditions.

Table 4. Effect of irrigation water regimes on the leaf mineral composition.

Treatment	**Leaf Mineral Composition (%)**			
	Na	**Cl**	**Mg**	**Ca**
T1	3.4 ± 0.19	1.3 ± 0.07	0.43 ± 0.03	0.0006 ± 0.04
T2	4.3 ± 0.25	3.0 ± 0.22	0.36 ± 0.02	0.0004 ± 0.02
T3	4.4 ± 0.42	3.5 ± 0.09	0.35 ± 0.01	0.0004 ± 0.03
	N	K	P	Fe
T1	0.34 ± 0.02	4.2 ± 0.23	3.1 ± 0.02	0.0001 ± 0.02
T2	0.34 ± 0.01	3.7 ± 0.17	2.7 ± 0.04	0.0002 ± 0.03
T3	0.37 ± 0.01	4.1 ± 0.14	2.7 ± 0.02	0.0001 ± 0.01

3.4. Effect of Irrigation Water Regimes on Chlorophyll Content

The reaction of chlorophyll content of leaves of *T. tetragonioides* to the different water regimes is shown in Table 5. The results show that the chlorophyll content was higher in treatment T2 and lower in treatments T1 and T3. These results are in agreement with the findings obtained by Reference [24], where the minimum amounts of chlorophyll a, chlorophyll b, and total chlorophyll were obtained from the wettest and driest treatment in *Matricariachamomilla* L. potted plants. Similar results were obtained by References [25,26].

Table 5. Leaf chlorophyll content in leaf. DM—dry matter.

Treatment	**Chlorophyll Content**					
	C_a (mg·m^{-2})	C_b (mg·m^{-2})	C_{a+b} (mg·m^{-2})	C_a (mg·g^{-1}; DM)	C_b (mg·g^{-1}; DM)	C_{a+b} (mg·g^{-1}; DM)
T1	232 ± 12.1	81 ± 5.8	313 ± 17.6	27 ± 1.4	9.2 ± 0.7	36 ± 1.9
T2	289 ± 7.3	110 ± 4.5	400 ± 11.4	32 ± 1.3	12.2 ± 0.5	45 ± 1.7
T3	254 ± 13	92 ± 5.6	346 ± 18	30 ± 2	10.7 ± 0.6	41 ± 2.1

3.5. Effect of Irrigation Water Regimes on Carotenoid Content in Leaves

Carotenoids which exist in all higher plants are synthesized and located in the chloroplast along with the chlorophyll. Table 6 shows the carotenoid content of the leaves of *T. tetragonioides* under different irrigation water regimes. The maximum leaf carotenoid content was 8.44 mg·g^{-1} DW in treatment T2. In wetter and drier treatments (T1 and T3), the carotenoid content was lower, with values of 7.2 and 7.9 mg·g^{-1} DW, respectively. Lower carotenoid content was also obtained for stress water regimes of some fenugreek varieties [27]. Moreover, the leaf carotenoid levels of green beans decreased, which was attributed to water stress; the vegetation index (NDVI) then showed the highest correlations with the chlorophyll (a, b, and total) and carotene content of leaves [28].

Table 6. Carotenoid leaf content of the species.

Treatment	**Leaf Carotenoid Content**	
	Car (mg·m^{-2})	Car (mg·g^{-1}; DW)
T1	62.9 ± 3.2	7.2 ± 0.3
T2	75.8 ± 2.2	8.4 ± 0.4
T3	67.6 ± 4.1	7.9 ± 0.5

3.6. Effect of Irrigation Water Regimes on Soluble Carbohydrates Content

The irrigation water regimes had a slight effect on the soluble carbohydrate content on leaves of the species *T. tetragonioides*. The glucose and soluble carbohydrate content in leaves increased in the wet (T1) and dry (T3) treatments: glucose, 0.58 and 0.57 mg·mL^{-1}, respectively; soluble carbohydrates, 1.71 and 1.67 mg, respectively. These results are confirmed by Reference [29]. There was a decrease in glucose (0.54 mg·mL^{-1}) and soluble carbohydrate (1.59 g) content in leaves in the medium (T2) treatment (Table 7).

Table 7. Soluble carbohydrates content of leaves.

Treatment	**Soluble Carbohydrates**				
	Glucose (mg·mL^{-1})	Area (cm^2)	Soluble Carbohydrates (mg)	DW (cm^2)	Soluble Carbohydrates (g)
T1	0.57 ± 0.05	3.42	1.67 ± 0.14	0.001	1.9 ± 0.15
T2	0.54 ± 0.03	3.42	1.59 ± 0.08	0.001	1.8 ± 0.09
T3	0.58 ± 0.03	3.42	1.71 ± 0.09	0.001	2.0 ± 0.12

3.7. Yield of Species

T. tetragonioides produced significant amounts of dry matter, which ranged from 82.7 to 66.1 g·plant^{-1}. The partition of the plant dry matter to plant organs was changed by the effect of the irrigation water regimes (Table 8). The fact that the species was irrigated during the vegetation period T1 (70%, wet treatment) significantly increased the dry biomass of the species at the harvest time, averaging 6616 kg·ha^{-1}. The dry matter of the species decreased when the soil water decreased in treatments T2 (50%, medium treatment) and T3 (30%, dry treatment). There was no significant difference between treatments. The obtained results confirmed that the species *T. tetragonioides* is tolerant to drought conditions. The yield of the crop shows that the drought had less effect than the salinity (6616–5288 kg DM·ha^{-1}). These results are confirmed by the previous study of Reference [3].

Table 8. Effect of irrigation water regimes on yield of the species. FW—fresh weight; DW—dry weight; FM—fresh matter.

Treatment	*Tetragonia tetragonioides*				
	FW (g·plant^{-1})	DW (g·plant^{-1})	Yield (%)	FM (kg·ha^{-1})	DM (kg·ha^{-1})
T1	1041.2 ± 12	82.7 ± 4	7.8 ± 0.3	83,284 ± 967	6609 ± 329
T2	913.7 ± 23	66 ± 3	7.2 ± 0.5	73,094 ± 1805	5289 ± 248
T3	966.2 ± 22	67.3 ± 3	6.8 ± 0.3	77,300 ± 1787	5377 ± 242

4. Conclusions

The experimental results showed several effects of the water irrigation regimes on the growth, mineral composition, and photosynthetic pigments of *T. tetragonioides*, as listed below.

- Plant growth (stem, leaves, and seeds) increased slightly with an enhancement of the water level (near the field capacity), and the growth difference between the drier water regimes was very low. This increase was probably due to the increase of stomatal conductance and, consequently, transpiration and CO_2 fixation were higher. Hence, it is not surprising that experimental results, in which the only variable was water application, agree quite well with this supposed theory.

- Leaf mineral composition of chloride and sodium are the main responsible ions for soil salinization and alkalization, respectively, in arid and semi-arid regions, enhanced by the decrease in soil water content. The content was very high in relation to other plants, showing its high capacity as a salt-removing species.

- There was a generally low decrease in phosphorus, calcium, potassium, iron, and magnesium in leaf content under drought conditions, probably due to the chloride and potassium competition.

- The total nitrogen leaf content of species showed very low variation, probably due to the same fertigation for all irrigation treatments.

-The minimum carotenoid amounts of chlorophyll a, chlorophyll b, and total chlorophyll were obtained from the wettest and the driest treatment in *T. tetragonioides* plants, probably due to higher plant senescence provoked by these regimes.

- The glucose and soluble carbohydrate contents of leaves increased in the driest treatments and had enhanced tolerance to drought conditions.

- The yield of the species increased in the wettest and the driest treatments.

In conclusion, it can be suggested that *T. tetragonioides* is a species tolerant to drought conditions. Its capacity as a halophyte and salt-removing species when the soil water content decreases was shown, suggesting its use in arid and semi-arid regions. Moreover, growth and yield differences in the various irrigation regimes were very low, which suggests another important advantage of these species—its cultivation under dry conditions, when used as a leafy vegetable for human consumption or for animal feeding. Nevertheless, more research is needed in order to test plant development under drier conditions in arid and semi-arid climates.

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