

# Phenotypic Diversity of Agronomical Traits and Nut Phenolic Compounds among Pistachio (*Pistacia vera* L.) Cultivars <sup>†</sup>

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**Abstract:** The phenotypic diversity of agronomical traits and nut phenolic compound content of four pistachio cultivars (Mateur, Elguetar, Kerman, and Ohadi), conducted under regulated deficit irrigation (RDI) during two growing seasons (2017/2018), were evaluated. The experimental orchard was located in the Regional Center of Agricultural Research of Sidi Bouzid (CRRRA), Tunisia. Three water treatments were applied from March to September; control (T0; 100% ETc during all the developmental stages), RDI treatment (T1; 50% ETc during stages I and II of fruit development followed by full irrigation 100% ETc during stage III), and stressed treatment (T2; 50% ETc during all the growing season). The results showed that treatments T0 and T1 no presented statistically significant difference in yield and nut biochemical traits, and 50% less irrigation water was used during stages I and II of nut development. The trees under treatment T2 were affected by water stress showing low values of yield and nut phenolic compounds.

**Keywords:** pistachio; drought stress; phenolic compounds



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## 1. Introduction

In semiarid conditions, water availability for crop irrigation has declined as a consequence of global climate change. Pistachio is a nut tree that has the reputation of being a drought tolerant and saline resistant species, cultivated under rainfed conditions in its region of origin [1]. It is well known that pistachio irrigation increases the yield, improves the nut quality, and dampens the normal alternate bearing pattern [2]. Regulated deficit irrigation (RDI) is a system of managing water supply by imposing some water deficits in specific phenological stages, which are found to be less sensitive, with no (or low) reduction in economic benefits [3]. Goldhamer and Beede [4] showed that the same reduction in the irrigation water during stages I (nut growth) and II (shell hardening) did not reduce the total amount of fruits and increased the percentage of shell splitting, although it also increased early splitting.

Pistachio nut has peculiar organoleptic characteristics including a high nutritional value and high unsaturated fatty acids content; it is a rich source of fat (about 50–70%) and a good source of proteins and minerals and bioactive components [5].

Our experiment was conducted to study the effectiveness of RDI irrigation on yield and nut phenolic compounds content in four pistachio cultivars conducted in a high-density orchard under semiarid conditions.

## 2. Materials and Methods

### 2.1. Plant Material

The trial was carried out in the Regional Center of Agriculture Research (CRRRA, Sidi Bouzid) in west central Tunisia (9°43' E, 35°01' N; altitude 354 m). Fifteen-year-old pistachio trees grafted onto *P. atlantica* rootstock were studied.

## 2.2. Treatments for Regulated Deficit Irrigation (RDI)

The phenological stages taken into account in the RDI treatments were those suggested by Goldhamer and Beede (2004): Stage I (from sprouting until the end of rapid nut growth), stage II (from maximum nut size until the beginning of kernel growth), and stage III (from the beginning of kernel growth until harvest). Three irrigations were applied during the two years of the study:

- Treatment (T0): trees received water to cover estimated evapotranspiration (ETc) losses.
- Treatment (T1): trees received 50% of the water received by the control trees during stages I and II and the same amount of water as the control trees during stage III.
- Treatment (T2): trees received water to replace 50% ETc calculated for the control treatment during all the steps.

Drip irrigation was applied 3 days per week and was controlled and adjusted weekly according to soil potential (measured by tensiometers located 25 cm from the drip head at depths of 30 and 60 cm). A drip line was utilized in each tree row, with four self-compensating drippers ( $4 \text{ L h}^{-1}$ ) per tree, 0.5 m apart. The amount of provided water was calculated on the basis of the crop evapotranspiration (ETc) and the crop coefficient (Kc) according to the FAO method [6]:  $\text{ETc} = \text{ETo Kc}$ .

## 2.3. Agronomical Traits

The trunk cross-sectional area (TCSA) was measured during the dormant season at 30 cm above the graft union. Yield (kg/tree) was determined per tree, and yield efficiency (YE) of each scion–stock was computed from the harvest data. At harvest, a representative nut sample (100 nuts) was taken for pomological evaluations.

## 2.4. Nut Biochemical Quality

Samples were grounded to powder by a mortar and pestle, separately. Twenty grams of each kernel were extracted by 200 mL of 95% methanol for 48 h at room temperature. Then, the extracts were filtered and evaporated at low pressure; samples were stored at  $-20^\circ\text{C}$  until analysis. The total anthocyanin content, flavonoid, total phenolics and antioxidant capacity were determined as described in [7].

## 2.5. Statistical Analysis

Mean values and mean standard error (SE), were calculated for each studied trait. Analysis of variance (ANOVA) was performed using SPSS 20.0 (SPSS Inc., Chicago, IL, USA). Means were compared using the Scheffe test at the 5% significance level.

# 3. Results and Discussion

## 3.1. Agronomical Traits

The results showed significant differences ( $p < 0.05$ ) among cultivars in yield, TCSA, YE, tree height, fruit weight, and production of blanks (Table 1). The irrigation regimes affected the yield and its components, and significant differences ( $p < 0.05$ ) were observed between the three treatments. The control and treatment T1 presented similar yield values, while treatment T2 showed a significantly ( $p < 0.05$ ) lower yield. These results are similar to those reported by [8], studying the impact of regulated deficit irrigation on pistachio trees, showing that the mean yield for three years of study was not reduced in T1 compared to the control treatment. The cv. Mateur presented the highest yield ( $7.1 \text{ Kg/tree}$ ) under the control treatment, whereas the lowest yield was observed in the cv. Elguetar under the treatment T2 ( $4.0 \text{ Kg/tree}$ ). The treatment T2 showed low values of split nuts and blanks as compared to the control and RDI treatment.

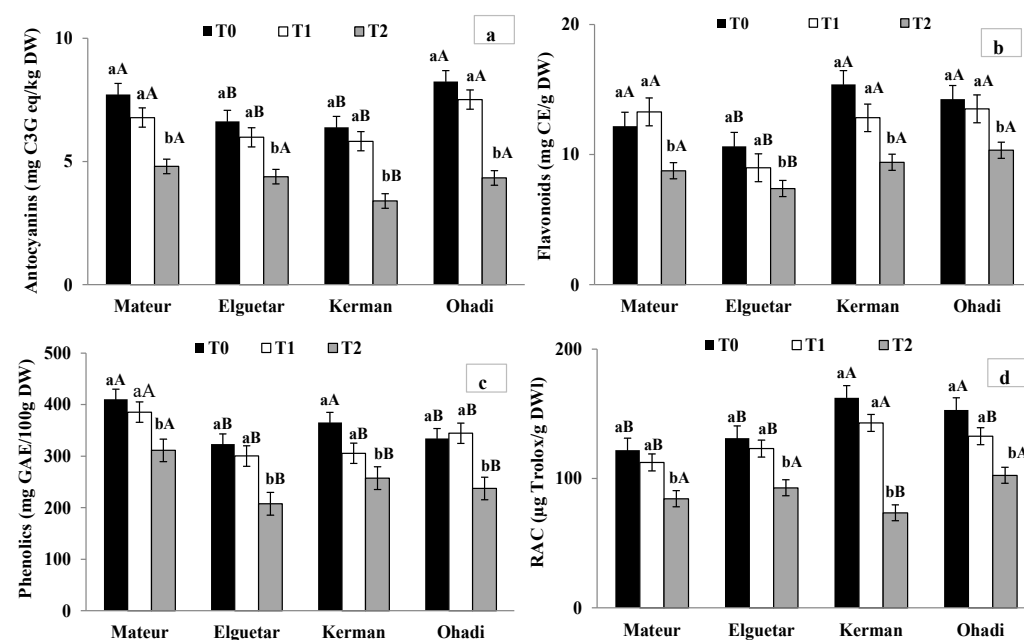
**Table 1.** Agronomical and pomological traits of pistachio cultivars. Values are means ( $n = 3$ )  $\pm$  SE. Different letters a, b, c, indicate differences ( $p < 0.05$ ) among three irrigation treatments (T0: control; T1: regulated deficit irrigation; and T2: stressed) in the same cultivar. a, b, c indicate differences ( $p < 0.05$ ) among cultivars.

	Cultivar	Yield	TCSA	YE	Height	FW	DW	L	Split	Blanks
T0	Mateur	7.1 $\pm$ 1 <sup>a</sup>	35.5 $\pm$ 5 <sup>a</sup>	0.20 <sup>a</sup>	2.5 $\pm$ 0.1 <sup>a</sup>	0.62 $\pm$ 0.1 <sup>b</sup>	0.50 $\pm$ 0.1 <sup>b</sup>	17.8 $\pm$ 2 <sup>a</sup>	76 $\pm$ 5 <sup>a</sup>	7 $\pm$ 2 <sup>b</sup>
	Elguetar	5.1 $\pm$ 1 <sup>b</sup>	31.8 $\pm$ 4 <sup>b</sup>	0.16 <sup>b</sup>	2.3 $\pm$ 0.1 <sup>a</sup>	0.44 $\pm$ 0.1 <sup>c</sup>	0.50 $\pm$ 0.1 <sup>b</sup>	15.8 $\pm$ 2 <sup>b</sup>	71 $\pm$ 5 <sup>b</sup>	9 $\pm$ 3 <sup>b</sup>
	Kerman	5.0 $\pm$ 1 <sup>b</sup>	32.5 $\pm$ 5 <sup>b</sup>	0.15 <sup>b</sup>	2.2 $\pm$ 0.1 <sup>a</sup>	0.58 $\pm$ 0.1 <sup>b</sup>	0.46 $\pm$ 0.1 <sup>c</sup>	17.2 $\pm$ 2 <sup>a</sup>	70 $\pm$ 2 <sup>b</sup>	10 $\pm$ 3 <sup>b</sup>
	Ohadi	3.1 $\pm$ 1 <sup>c</sup>	29.8 $\pm$ 2 <sup>c</sup>	0.10 <sup>c</sup>	2.0 $\pm$ 0.1 <sup>a</sup>	0.77 $\pm$ 0.1 <sup>a</sup>	0.64 $\pm$ 0.1 <sup>a</sup>	18.8 $\pm$ 2 <sup>a</sup>	55 $\pm$ 3 <sup>c</sup>	25 $\pm$ 5 <sup>a</sup>
T1	Mateur	6.4 $\pm$ 2 <sup>a</sup>	36.5 $\pm$ 4 <sup>a</sup>	0.17 <sup>a</sup>	2.3 $\pm$ 0.1 <sup>b</sup>	0.60 $\pm$ 0.5 <sup>b</sup>	0.51 $\pm$ 0.5 <sup>b</sup>	17.4 $\pm$ 1 <sup>a</sup>	70 $\pm$ 3 <sup>b</sup>	10 $\pm$ 2 <sup>b</sup>
	Elguetar	4.7 $\pm$ 3 <sup>b</sup>	33.2 $\pm$ 3 <sup>b</sup>	0.14 <sup>b</sup>	1.9 $\pm$ 0.1 <sup>b</sup>	0.46 $\pm$ 0.6 <sup>c</sup>	0.4 $\pm$ 0.7 <sup>c</sup>	15.4 $\pm$ 1 <sup>b</sup>	75 $\pm$ 2 <sup>a</sup>	13 $\pm$ 3 <sup>b</sup>
	Kerman	4.1 $\pm$ 1 <sup>b</sup>	28.2 $\pm$ 5 <sup>b</sup>	0.14 <sup>b</sup>	2.3 $\pm$ 0.1 <sup>a</sup>	0.52 $\pm$ 0.1 <sup>b</sup>	0.42 $\pm$ 0.1 <sup>c</sup>	17.0 $\pm$ 2 <sup>a</sup>	65 $\pm$ 2 <sup>b</sup>	12 $\pm$ 3 <sup>b</sup>
	Ohadi	2.1 $\pm$ 1 <sup>c</sup>	22.0 $\pm$ 4 <sup>c</sup>	0.10 <sup>c</sup>	1.8 $\pm$ 0.1 <sup>b</sup>	0.72 $\pm$ 0.1 <sup>a</sup>	0.62 $\pm$ 0.1 <sup>a</sup>	18.1 $\pm$ 2 <sup>a</sup>	48 $\pm$ 2 <sup>c</sup>	29 $\pm$ 2 <sup>a</sup>
T2	Mateur	5.1 $\pm$ 1 <sup>a</sup>	32.5 $\pm$ 3 <sup>b</sup>	0.15 <sup>a</sup>	2.2 $\pm$ 0.1 <sup>b</sup>	0.51 $\pm$ 0.5 <sup>b</sup>	0.48 $\pm$ 0.4 <sup>b</sup>	17.1 $\pm$ 1 <sup>a</sup>	68 $\pm$ 4 <sup>a</sup>	19 $\pm$ 1 <sup>b</sup>
	Elguetar	4.0 $\pm$ 2 <sup>b</sup>	38.6 $\pm$ 4 <sup>a</sup>	0.10 <sup>b</sup>	1.7 $\pm$ 0.1 <sup>b</sup>	0.42 $\pm$ 0.1 <sup>c</sup>	0.3 $\pm$ 0.1 <sup>c</sup>	15.1 $\pm$ 1 <sup>b</sup>	70 $\pm$ 3 <sup>a</sup>	16 $\pm$ 2 <sup>b</sup>
	Kerman	4.0 $\pm$ 1 <sup>b</sup>	30.0 $\pm$ 5 <sup>b</sup>	0.13 <sup>b</sup>	2.2 $\pm$ 0.1 <sup>a</sup>	0.50 $\pm$ 0.1 <sup>b</sup>	0.40 $\pm$ 0.1 <sup>b</sup>	17.0 $\pm$ 2 <sup>a</sup>	60 $\pm$ 3 <sup>b</sup>	14 $\pm$ 2 <sup>b</sup>
	Ohadi	2.0 $\pm$ 1 <sup>c</sup>	20.5 $\pm$ 2 <sup>c</sup>	0.10 <sup>c</sup>	1.7 $\pm$ 0.1 <sup>b</sup>	0.70 $\pm$ 0.1 <sup>a</sup>	0.60 $\pm$ 0.1 <sup>a</sup>	17.8 $\pm$ 2 <sup>a</sup>	41 $\pm$ 2 <sup>c</sup>	35 $\pm$ 2 <sup>a</sup>

Units and Abbreviations: Yield (kg/tree); TCSA = trunk cross-sectional area in cm<sup>2</sup>; FW = Fresh weight (g); DW = dry weight (g); YE = Yield Efficiency (kg/cm<sup>2</sup>); L = Length (mm).

### 3.2. Nut Phenolic Compounds Content

The anthocyanins, flavonoids, total phenolics, and relative antioxidant capacity of pistachio nuts under the three water regimes are shown in Figure 1.



**Figure 1.** Phenolic compounds in pistachio nuts under RDI. The anthocyanin content are shown in (a), flavonoids are shown in (b), total phenolic are shown in (c), relative antioxidant capacity are showed in (d). Values are means ( $n = 6$ )  $\pm$  SE. Letters (a, b, c), indicate difference among treatments in the same cultivars. Letters (A, B, C), indicate differences ( $p < 0.05$ ) among cultivars. Abbreviations: C3GE = Cyanidin-3-glucoside equivalents; CE = Catechin equivalents; GAE = Gallic acid equivalents; RAC = Relative Antioxidant Capacity; DW = dry weight.

The irrigation treatments applied significantly ( $p < 0.05$ ) affected the anthocyanins, flavonoids, total phenolics, and RAC values with the T2 treatment presenting lower values. The two treatments T0 and T1 presented similar behavior and showed statistically significant differences ( $p < 0.05$ ) with treatment T2. Our results are in accordance with those presented by [9] showing an anthocyanins content between 1.4 and 8.9 mg/Kg in pistachio

whole nut. The flavonoid values obtained in our study are in accordance with the range (3.5 to 7.2 mg CE g<sup>-1</sup> DW) of a previous study conducted by [10]. The antioxidant capacity values showed great variations among cultivars, but the values are in accordance with the range (45.7 to 122.6 µg TE g<sup>-1</sup> DW) reported by [10]. The irrigation treatments applied significantly affected the RAC values with the T2 treatment presenting lower values.

#### 4. Conclusions

Our results showed that the studied pistachio cultivars presented differential responses to RDI irrigation. However, treatment T1 led to a similar yield, nut weight, and phenolic compounds as the control treatment. The reduction in irrigation volumes by 50% during stage I and II of nut development over the control (100% ETc) could be a suitable irrigation strategy implemented in high density pistachio orchards in semiarid conditions.

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