

Highlights

1. Salinity decrease linearly water use of wild rocket
2. Reduction in yield of wild rocket by salinity occurs mainly for decline in leaf area and secondly leaf number
3. Salinity reduced specific leaf area and increased leaf succulence of wild rocket
4. Y_{WUE} of wild rocket was lowered from moderate to high salinity
5. Wild rocket ranks among moderately salt sensitive species

1 Water use and crop performance of two wild rocket genotypes under salinity conditions

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13 Abstract

14 In literature, the parameters of salinity tolerance of the main cultivated species are known, but are
15 missing for many minor species such as wild rocket, whose cultivation in many areas of southern
16 Italy affected by salinity is growing. Therefore, a research has been carried out i) to evaluate the
17 response to salinity in water use, water use efficiency, yield characteristics and morfological
18 features, and ii) identify the salinity tolerance parameters of two genotypes of wild rocket:
19 *Diplotaxis tenuifolia* (L.) DC and *D. muralis* (L.) DC. The study was carried out in the spring of
20 2007 and 2008 in Policoro (MT), southern Italy, under unheated plastic greenhouse conditions.
21 Wild rocket was sown in plastic pots containing 20 dm³ of soil. For each genotype, six soil salinity
22 levels were compared, obtained by accurately mixing before sowing, the soil with, 0.0, 0.5, 1.0, 2.0,
23 3.5 and 5.5 g dm⁻³ of NaCl + CaCl₂ 1:1 (on a weight basis). Irrigation was performed with fresh
24 water having electrical conductivity of 0.5 dS m⁻¹. In each year, 3 harvests were performed; water
25 use and the main production and plant growth parameters were recorded. *D. tenuifolia* provided a
26 yield 47.3% higher than *D. muralis*. By rising salinity, progressive decline in marketable yield and
27 growth of the leaves was recorded, while the dry matter content increased. The increase in salinity
28 has led to the progressive reduction of water use in both genotypes. From moderate salinity values
29 (about 5.5 dS m⁻¹), the reduction in yield water use efficiency as a result of increased salinity has
30 been observed. In addition, salinity reduced specific leaf area and increased leaf succulence. Both
31 genotypes rank among moderately salt sensitive species, according to Maas and Hoffman's model
32 (1977). However, *D. tenuifolia*, with a critical threshold of 1.98 dS m⁻¹ and a slope of 6.61% m dS⁻¹,
33 showed a slightly higher tolerance than *D. muralis* (threshold 1.34 dS m⁻¹ and slope 7.25% m dS⁻¹).

34 Reduction in yield due to salinity occurred mainly for the decrease in leaf size and, secondly,
35 number of leaves.

36

37 **Key words:** *Diploaxis tenuifolia*; *D. muralis*; critical threshold; slope; salinity tolerance; WUE

38

39 **Introduction**

40 The beginning of 21st century is marked by global scarcity of water resources, environmental
41 pollution and increased salinization of soil and water (Shahbaz and Ashraf, 2013).

42 It has been estimated that worldwide 20% of total cultivated and 33% of irrigated agricultural lands
43 are affected by high salinity. Furthermore, the salinized areas are increasing at a rate of 10%
44 annually for various reasons, including low precipitation, high surface evaporation, weathering of
45 native rocks, irrigation with saline water, and poor cultural practices. It has been estimated that
46 more than 50% of the arable land would be salinized by the year 2050 (Jamil et al., 2011).
47 Particularly, in the Mediterranean countries groundwater discharge increased over the second half
48 of the 20th century and, as a consequence, a great number of aquifers are currently overexploited
49 and at risk of seawater intrusion (Polemio, 2016). This overuse is concentrated in coastal areas,
50 where increasing population, growth of urban areas, and increases of irrigation and industrial
51 demands and tourism are occurring (Polemio et al., 2013; Taniguchi et al., 2009; Tulipano et al.,
52 2005). These trends were also observed in Italy, where seawater intrusion is the main cause of
53 groundwater quality degradation in coastal karst aquifers, the largest of which are located in the
54 Apulia region (Polemio et al., 2011).

55 Salinity is one of the most serious factors limiting productivity of agricultural crops, which causes
56 major reductions in cultivated land area, crop productivity and quality (Flowers, 2004; Munns and
57 Tester, 2008; Shahbaz and Ashraf, 2013; Yamaguchi and Blumwald, 2005). Salinity inhibits plant
58 growth i) for osmotic effect which reduces the plant ability to take up water, affects a wide variety
59 of metabolic activities, and causes an oxidative stress because of the formation of reactive oxygen
60 species such as superoxides and hydroxy and peroxy radicals (Munns, 2002; Munns, 2005; Sergio
61 et al., 2012), ii) by specific ion toxicity (*e.g.*, Na⁺ and Cl⁻) (Munns, 2002; Munns, 2005; Yeo et al.,
62 1991) and iii) by ionic imbalances acting on biophysical and/or metabolic components of plant
63 growth (Grattan and Grieve, 1999).

64 On the whole, the above effects lead to a reduction of net photosynthesis (Cantore et al., 2007;
65 Munns et al., 2006; Munns and Tester, 2008), the rate of leaf surface expansion (Wang and Nil,
66 2000), the fresh and dry weights of leaves, stems, and roots (Chartzoulakis and Klapaki, 2000;
67 Hernandez et al., 1995). The listed adverse effects result in a reduction in yield that, for a given

68 level of salinity, may vary depending on the genotype, salt type, climatic conditions and agronomic
69 techniques (Cucci et al., 2000; Flagella et al., 2002; Maas, 1986).

70 In many coastal areas of Southern Italy (as Apulia and Basilicata regions) where the problem of
71 irrigation water's salinity is increasing, the cultivation of wild rocket (i.e. *Diplotaxis tenuifolia* L.
72 DC., *D. muralis* L. DC.) is widespread and in further expansion. Indeed, the last decades wild
73 rocket has become popular and widely cultivated in greenhouses and open field. In Italy, several
74 species of the genus *Diplotaxis* are consumed as vegetables since ancient times. The leaves,
75 characterized by a unique aroma and piquant flavor, can be eaten raw in salads or cooked in many
76 recipes. Compared to other leafy vegetables, wild rocket has high content of fiber and iron, ascorbic
77 acid, phenols, carotenoids and glucosinolates (Barillari et al., 2005; Cavaiuolo and Ferrante, 2014;
78 D'Antuono et al., 2009; Di Venere et al., 2000), to which important bioactive properties (e.g.,
79 antioxidant, antitumour, etc.) are often ascribed (Ramos-Bueno et al., 2016).

80 Experimental evidence on the behaviour of wild rocket in the presence of salinity are scarce and
81 conflicting. In particular, de Vos et al. (2013) ranked *D. tenuifolia* as a salt tolerant species, having
82 found that yield reduction occurs with the salinity of nutrient solution greater than 100 mM NaCl.
83 These authors claim that the species can be considered among the new halophytes. However,
84 opposite results were obtained by Bonasia et al. (2017) that, for the same species, reported
85 significant reductions in yield (about 20%) passing from the salinity of 2.5 dS m⁻¹ to 3.5 dS⁻¹. The
86 latter authors also observed a positive effect of moderate salinity (3.5 dS m⁻¹) on different
87 qualitative parameters. In fact, they report a reduction in the content of nitrates, which is widely
88 recognized as being harmful to health (Buttaro et al., 2016), and an improvement in certain
89 qualitative features, health beneficial, such as vitamin C, polyphenols, carotenoids and antioxidant
90 activity. With an higher salinity, however, they did not find any further qualitative improvements.
91 Also Hamilton and Fonseca (2010) found an increase in the phenols with the salinity increase from
92 1.5 to 9.5 dS m⁻¹ only in one of the two experiments conducted, while they did not observe any
93 effect on vitamin C content.

94 Considering the growing economic importance of wild rocket cultivation in many salt-affected
95 areas, as the case of Mediterranean countries, and the conflicting literature data on the salt tolerance
96 of this vegetable, this work is proposed to provide further insights on the crop performance of two
97 genotypes of wild rocket (*D. tenuifolia* and *D. muralis*) in response to the soil salinity levels. The
98 information obtained from the research aim to provide useful information for the optimal crop
99 management of wild rocket under salinity conditions.

100

101

102 **Material and methods**

103 *Experimental site characteristics*

104 The research was carried out in the spring 2007 and 2008 at experimental farm 'E. Pantanelli' of the
105 University 'Aldo Moro' of Bari, Policoro (MT), Southern Italy (40°10' NL, 16°39' EL, altitude 15
106 m a.s.l.). This site is characterized by sub-humid climate according to the De Martonne
107 classification (Cantore et al., 1987).

108 The experiment was performed under unheated plastic greenhouse conditions (covered by an EVA
109 200 µm thick film), using cylindrical pots (0.34 m diameter and 0.3 m height) adequately equipped
110 with flowerpot saucers, each containing 20 dm³ of soil, collected in the same location. The soil was
111 a fine, mixed, subactive, thermic Chromic Haploxererts (Cassi et al., 2006), with the following
112 physical and chemical characteristics: sand ($2 > \text{Ø} > 0.02$ mm) 29.5%, silt 37.5%, clay ($\text{Ø} < 2$ µ)
113 33.0%; pH 7.6; total N (Kjeldahl method) 1.48 g kg⁻¹, available P₂O₅ (Olsen method) 25.9 mg kg⁻¹,
114 exchangeable K₂O (ammonium acetate method) 249 mg kg⁻¹, organic matter (Walkley–
115 Blackmethod) 33.7 g kg⁻¹, total limestone 14 g kg⁻¹, active limestone 4.5 g kg⁻¹; saturated paste
116 extract electrical conductivity (ECe) 0.90 dS m⁻¹; ESP 2.0%; bulk density 1.24 kg dm⁻³; soil
117 moisture at field capacity (measured in situ) 31.8% and at wilting point (−1.5 MPa) 15.3% (w/w) of
118 soil dry weight.

119 Weather data were measured in the greenhouse by an automatic weather station including a
120 pyranometer (model CM 4, Kipp and Zonen, Delft, The Netherlands), thermistor (model E001,
121 Tecno.El, Rome, Italy), hygrometer (C-83_N Rotronic, Zurich, Switzerland) and anemometer
122 (model VT 0805B, SIAP Bologna, Villanova di Castelnaso-BO, Italy), for measuring solar
123 radiation, air temperature, relative humidity and wind speed, respectively. Data were collected by
124 the electronic system operated through a data-logger (model Kampus, Tecno.El, Rome, Italy)
125 connected via modem to a PC. The trend of global radiation (R_g), minimum and maximum air
126 temperature (T_{\min} , T_{\max}) was increasing from sowing time until the end of the experiment according
127 to the typical one of the area concerned by the experiment. R_g ranged between about 5.0 and 19.5
128 MJ m⁻² d⁻¹, and about 4.5 and 19.0 MJ m⁻² d⁻¹, respectively in the first and second year. T_{\max} , of
129 about 14 °C in the period of sowing, increased gradually until the period of last harvest, reaching 28
130 and 29.5 °C in the first and second year, respectively. T_{\min} ranged between 4 °C to about 16 °C for
131 both years.

132

133 *Experimental design and crop management*

134 The following treatments were compared: two genotypes of wild rocket (*D. tenuifolia* and *D.*
135 *muralis*) and six soil salinity levels, obtained by accurately mixing to the soil, before sowing, 0.0,
136 0.5, 1.0, 2.0, 3.5 and 5.5 g dm⁻³ of NaCl + CaCl₂ 1:1 (on a weight basis), indicated with S₁, S₂, S₃,

137 S₄, S₅ and S₆, respectively. A completely randomized block experimental design with 4 replicates
138 was adopted. Each plot consisted of tree pots.

139 Before sowing, the soil of each pot was fertilised with 1.78 and 2.26 g of diammonium phosphate
140 and urea, respectively. The wild rocket was sown on February 1st 2007 and 2008. Fifteen days after
141 sowing (DAS), thinning was performed by leaving five plants per pot, arranged in an homogeneous
142 manner with respect to the pot surface. Each year three growing cycles were performed by
143 exploiting the ability of this species to re-growth after harvest. The harvests (on March 20th, April
144 23th, May 25th 2007, and on March 25th, April 28th, May 30th 2008) were performed cutting off
145 leaves 1 cm above the collar with a knife. Evapotranspiration (ET) was estimated by the water
146 balance method (Moazed et al., 2014) by weighing every days the pots that were considered as a
147 weight lysimeter.

148 In order to satisfy water requirements of the rocket, fresh water having an electrical conductivity of
149 0.5 dS m⁻¹ was supplied manually from the top of the pots. Water was applied when the allowable
150 water depletion (p) was reached in the S₁ treatment of each genotype. The threshold was assumed to
151 be 0.45 of total available water (p = 0.45) during the whole growing cycle, in accordance with the
152 values of *Brassicaceae* vegetable species (Allen et al., 1998). In all treatments the whole amount of
153 water consumed was restored at each irrigation event. The water that eventually leached in the
154 flowerpot saucers, was collected and used for the subsequent watering in the same pots.

155 Throughout the growing cycle, water was supplied 28 times in the first year and 27 times in the
156 second one. The seasonal irrigation volume was higher for *D. tenuifolia*, and decreased from the
157 non-saline treatment to the more saline one, according to the differences in ET measured.

158

159 *Yield and plant biometric characteristics*

160 At each harvest, shoots (leaves and stems) collected in each pot were utilized to determine yield
161 (total, marketable and unmarketable), number of leaves per plant, leaf area (LA) per plant and leaf
162 area index (LAI). LA was measured using a leaf area meter (Li-COR, 3100, Lincoln Nebraska,
163 USA). Specific leaf area (SLA) was calculated as the ratio between LA and leaf dry weight. Leaf
164 succulence (LS) was calculated as the ratio between leaf fresh weight and LA.

165 Leaves that were yellow, necrotic or damaged by pests or fungi were considered unmarketable.

166 The leaf dry matter (DM) content was assessed on the marketable product. To determine DM, a
167 sample of about 80 g of marketable product was dried in a ventilated oven at 60 °C, until a constant
168 weight was reached (about 48 h).

169

170 *Water use efficiency, yield response factor*

171 The water use efficiency (WUE) was calculated for each harvest as the ratio between marketable
172 yield and ET, i.e. yield WUE (Y_WUE).

173 Yield response factor to water (K_y), to predict wild rocket yield under saline conditions, was
174 calculated as angular coefficient of the following linear equation (Stewart et al., 1977):

175

$$176 \quad 1 - Y_a/Y_m = K_y (1 - ET_a/ET_m)$$

177

178 where Y_a (kg ha^{-1}) and ET_a (mm) are actual marketable yield and evapotranspiration for saline
179 treatments, respectively; Y_m (kg ha^{-1}) and ET_m (mm) are maximum marketable yield and
180 evapotranspiration for non-saline treatment; K_y is yield response factor.

181

182 *Soil salinity*

183 At the beginning of the growing cycle and at each harvest, soil samples were taken along the profile
184 of each pot through a cylindrical probe (\varnothing 2.5 cm), and tested for the electrical conductivity of the
185 saturation extract (ECe).

186

187 *Statistical analysis*

188 The data collected were elaborated by analysis of variance (ANOVA) procedure; mean values were
189 separated by Student-Newman-Keuls (SNK) test at $P = 0.05$. The SPSS 17 software was used for
190 the analysis.

191

192 **Results and Discussion**

193 *Soil salinity*

194 Soil salinity, as planned, did not show any considerable difference during the two cycles of
195 cultivation (Tab. 1). In fact, ECe has undergone only a slight increase during the growing cycles of
196 wild rocket, both as a result of the low salts content of irrigation water and for the correct
197 calibration of the irrigation volumes that avoided percolation and, consequently, the leaching of the
198 salts. In the few cases in which the percolation of excess water occurred, this was collected and
199 redistributed in the same pot in the successive irrigation. The described trend was not differentiated
200 between the two genotypes of wild rocket.

201 Therefore, as average of genotypes and years, ECe was growing in relation to the saline treatments
202 from 0.9 dS m^{-1} of S_1 to 12.3 dS m^{-1} of S_6 at sowing time, and from 1.6 dS m^{-1} of S_1 to 13.0 dS m^{-1}
203 of S_6 at the last harvest (Tab. 1). The salinity response models that will be reported below are

204 related to the average salinity of the crop cycle obtained from the average ECe measured at sowing
205 time and at each harvest.

206

207 *Yield and morphological features of plants*

208 There were no significant differences in growth and yield behaviour of *D. muralis* between the two
209 years, while *D. tenuifolia* provided higher yield in the second year, mainly for the largest number of
210 leaves per plant and, secondly, for the largest leaf medium surface (Tab. 2).

211 *D. tenuifolia*, with 45.8 g plant⁻¹ of total yield and 42.8 g plant⁻¹ (as a total of the three harvests) of
212 marketable yield, was more productive than *D. muralis* which, however, provided a total and
213 marketable average yield respectively of 32.0 and 29.1 g plant⁻¹.

214 The differences in yield between genotypes were mainly determined by the different number of
215 leaves per plant (11.9 and 16.3 respectively for *D. muralis* and *D. tenuifolia*) and, secondly, by the
216 different average surface area of the leaves (16.4 and 17.6 cm² leaf⁻¹, respectively for *D. muralis*
217 and *D. tenuifolia* respectively). Also Cantore et al. (2000) observed a greater yield of *D. tenuifolia*
218 than *D. muralis*, regardless of the cultivation period.

219 For both genotypes, yield was declining between the first and the third harvest. In fact, as the
220 average of the two years, the marketable yield has gone from 13.1 and 16.6 g plant⁻¹ of the first
221 harvest to 7.4 and 13.4 g plant⁻¹ of the third one, respectively for *D. muralis* and *D. tenuifolia* (Fig.
222 1).

223 Also other authors (Bianco and Boari, 1997; Boari et al., 1998) reported a considerable yield
224 decline after the 1st growing cycle of wild rocket cultivated during spring-summer period. Contrary,
225 the wild rocket cultivated in autumn-spring period showed an increasing trend of yield between the
226 first to the last harvest, proving that the different period of the crop cycle is the main factor
227 affecting the re-growth capacity of wild rocket (personal communication).

228 The production behaviour of the two genotypes in relation to saline treatments was very similar. In
229 general, the increase in salinity level was shown to progressively reduce the plant vegetative growth
230 and, consequently, yield (Tab. 2). For both genotypes, total yield and marketable one began to
231 decrease significantly in correspondence with the S₃ treatment. Total yield ranged from 43.2 and
232 58.8 g plant⁻¹ of S₁ to 14.5 and 23.5 g plant⁻¹ of S₆, respectively for *D. muralis* and *D. tenuifolia*;
233 instead, marketable yield ranged from 42.4 and 57.2 g plant⁻¹ of S₁ to 8.9 and 17 g plant⁻¹ of S₆,
234 respectively for the two genotypes. It should be noted that the differences between S₁ and S₆ for
235 marketable yield are higher than those found for total yield due to the greater incidence of the waste
236 that occurred with the increase in salinity. The adverse effects of osmotic stress and toxic stress for
237 the accumulation of toxic elements (ie: Na⁺, Cl⁻) induced by salinity, in fact, are more pronounced

238 on old leaves with early phenomenon of chlorosis and senescence, contributing to the increase of
239 waste, as observed also on other species (Munns and Tester, 2008; Wang et al., 2012; Yasar et al.,
240 2006; Negrão et al., 2017). It should be stressed that the wild rocket is among those vegetables
241 belonging to the salad category, whose commercial product requires the complete absence of
242 yellowed or necrotic leaves (Charfeddine and Gonnella, 2009). Reduction in yield caused from
243 increased salinity was mainly determined by the reduction in leaf area and, secondly, by decrease in
244 number of leaves (Tab. 2). Specifically, for both genotypes the leaf area per plant began to decrease
245 significantly in S₃, while the number of leaves per plant began to decrease in S₄. With the highest
246 salinity level, compared to control (S₁), the leaf area was reduced by about 79% and 70%,
247 respectively for *D. muralis* and *D. tenuifolia*, while the reduction in the leaf number was 44.5 and
248 40.8% respectively. On different species have been found a similar behaviour, with a adverse effect
249 of salinity which is initially manifested on the morphology of the leaves such as the leaf surface and,
250 second, on the number of leaves (Ünlükara et al., 2008; Ziaf et al., 2009). On the other hand, in
251 species such as spinach, increasing salinity would lead to the progressive reduction of the leaf
252 surface without affecting the leaf number (Ünlükara et al., 2017). Increasing salinity led to the
253 progressive increase in leaf dry matter content, which increased from 8.8 and 9.0 g 100 g⁻¹ FW in S₁,
254 to 11.0 and 10.9 g 100 g⁻¹ FW in S₆, respectively for *D. muralis* and *D. tenuifolia*. In addition, rising
255 salinity caused a progressive reduction in SLA and increase in LS. In fact, from S₁ to S₆, SLA
256 decreased by 35.0% for *D. muralis* and 33.8% for *S. tenuifolia*. LS, on the other hand, increased by 23.9%
257 for *D. muralis* and 23.8% for *D. tenuifolia*. SLA, an indicator of leaf thickness, is an important variable in
258 crop growth models, as it relates dry matter production to leaf area expansion and consequently to light
259 interception and photosynthesis (Gary et al., 1993). Generally, evidence shows that salinity
260 increases the leaf lamina thickness, due to an increase in mesophyll cell size or number of layers
261 (Kozłowski, 1997; Longstreth and Nobel, 1979). The increase in SLA or, conversely, the increased
262 leaf thickness or succulence with increased salinity results in conservation of internal water,
263 efficient water storage and dilution of accumulated salts (Flowers and Yeo, 1986; Munns and Tester,
264 2008). Increase in LS or decrease in SLA with rising salinity has also been reported for *D.*
265 *tenuifolia* (Bonasia et al., 2017; de Vos et al., 2013) and others *Brassicaceae* species as *Cochlearia*
266 *officinalis* (de Vos et al., 2013), *Cakile maritima* (Debez et al., 2004), *Crambe maritima* (de Vos et
267 al., 2010) and *Thellungiella salsuginea* (M'rah et al., 2006) and indeed appears to be a common
268 adaptation to salinity among species of the *Brassicaceae*. Probably, changes in leaf traits are linked
269 to osmotic effect of NaCl (which resembles a water-stress effect) rather than the ionic effect
270 (Munns, 1993; Munns and Tester, 2008).

272 *Effect of salinity on ET, WUE and yield response factor*

273 The ET of the wild rocket did not change significantly between years (Tab. 2). In agreement with
274 the leaf surface differences observed between the two genotypes, *D. tenuifolia* had a higher water
275 consumption than *D. muralis*. In fact, the leaf surface is one of the main factors influencing ET
276 (Allen et al., 1998).

277 Y_WUE was not different between the years for *D. muralis*, while for *D. tenuifolia* in 2008 there
278 was a 18.7% higher Y_WUE than in 2007, probably due to the higher yield obtained, to which
279 corresponded an higher LAI (Tab. 2).

280 Salinity influenced ET of the wild rocket. Specifically, as occurred for leaf production and leaf
281 surface, this parameter began to decrease in S₃ until reaching the 44.3 and 36.0% reduction in S₆,
282 respectively for *D. muralis* and *D. tenuifolia* (Tab. 2; Fig. 2). The reduction of ET caused by the
283 increase in soil salinity is due to the combined effect of salts on the soil and on the plant. In
284 particular, the soil evaporative component is influenced because soil salts result in increased soil
285 osmotic potential (Caruso, 1993), which in turn causes water activity to drop and consequently
286 reduces evaporation. Moreover, the formation of a crust due to salt precipitation decreases porosity
287 and increases tortuosity, which further contribute hindering evaporation (Gran et al., 2011).

288 The transpiration component was influenced by the concomitant reduction of leaf surface (Wang
289 and Nil, 2000), the variation of the morphological characteristics of the leaves (i.e. thickening,
290 reduction of stomatal density and of pore size), reduction of xylematic potential and of stomatal
291 conductance (Boari et al., 2014; Brugnoli and Lauteri 1991; Omamt et al., 2006; Sharma et al.,
292 2005). Decreases in plant water use due to salinity should be taken into account in irrigation
293 scheduling in order to prevent excess of water applications and excess of leaching, which in turn
294 can lead to excessive consumption of resources, waterlogging and radical asphyxia.

295 Although salinity has led to a reduction in water use, it had a growing adverse effect on Y_WUE
296 from moderate salinity conditions due to the concomitant greater reduction in yield. The reduction
297 of Y_WUE recorded with highest salinity, compared to control, was of 62.2 and 52.2%,
298 respectively for *D. muralis* and *D. tenuifolia* (Tab. 2). Probably this is attributable to the ionic
299 (toxic) effect of salinity. At low salinity, however, where the osmotic effect prevails (physiological
300 drought), the Y_WUE has not undergone any significant changes, as observed also on different
301 species subject to moderate water shortage (Boutraa et al., 2010; Chen et al., 2013; Favati et al.,
302 2009;). The reduction in Y_WUE with increasing salinity was found both on leafy and fruit
303 vegetable crops such as lettuce (Ünlükara et al., 2008), eggplant (Ünlükara et al., 2010) and tomato
304 (Zhang et al., 2016). A contrasting effect, however, was observed for species with high salinity
305 tolerance as amaranth (Omamt et al., 2006) and some *Brassicaceae* (Ashraf, 2001).

306 Stewart and Hagan (1973) proposed a model to predict crop yield from ET. The relation between
307 relative ET and relative yield decreases for water stress with yield response factor (K_y) has been
308 used to evaluate plant tolerance to water stress (Doorenbos and Kassam, 1979). If $K_y \leq 1$, the plant
309 is tolerant and if $K_y \geq 1$, the plant is sensitive to water stress. According to some authors that have
310 also used this method for salinity (Katerji et al., 1998; Shalhevet, 1994; Stewart et al., 1977;
311 Ünlükara et al., 2010; Ünlükara et al., 2017), the model was used to predict wild rocket yield under
312 saline conditions.

313 According to the Stewart and Hagan (1973) model tested in this study on wild rocket, the
314 relationship between relative yield and relative ET showed a slope (K_y) of 1.80 and 1.78, for *D.*
315 *muralis* and *D. tenuifolia*, respectively (Fig. 3). This high K_y value indicates that wild rocket is
316 highly sensitive to water stress caused by salinity.

317

318 *Salinity tolerance model application*

319 To identify the typical parameters of Maas and Hoffman (1977) model of response to salinity, the
320 linear regression of relative yield was calculated as a function of the average E_{Ce} of the growing
321 cycle.

322 The data of the two years were plotted together since there were not differences about the effects of
323 salinity level on the yield and related parameters between the two years.

324 From the linear regression we pointed out the critical threshold values of E_{Ce} above which the
325 relative yield starts to decrease, the slope, namely the relative yield reduction for each additional
326 increase of E_{Ce} above the critical threshold, E_{Ce50} or the E_{Ce} value corresponding to 50%
327 reduction in relative yield.

328 The above mentioned salinity tolerance parameters was similar between the two genotypes of wild
329 rocket, but shown a slight trend to the better salinity tolerance of *D. tenuifolia* (threshold 1.98 dS m⁻¹;
330 slope 6.61% m dS⁻¹; E_{Ce50} 9.54 dS m⁻¹) in respect to the *D. muralis* (threshold 1.34 dS m⁻¹; slope
331 7.25% m dS⁻¹; E_{Ce50} 8.24 dS m⁻¹) (Fig. 4). Despite these differences, the results demonstrate the
332 ranking of both genotypes among moderately salt sensitive species, among which we find several
333 vegetable species such as fennel (Cucci et al., 2014; Semiz et al., 2012), broccoli, cabbage, turnip,
334 carrots, lettuce, spinach (Flagella et al., 2002). More specifically, applying the Maas and Hoffman
335 (1977) model to the relative yield obtained in each harvest, a decreasing trend of salinity tolerance
336 between the first and the last harvest can be observed as can be seen from the trends of the
337 characteristic parameters of the model (Fig. 5). This is probably attributable to two factors: i) the
338 progressive accumulation of harmful salts in the parts of the plant that are not removed by the
339 harvest (roots and collar) and ii) the increase in air temperature between the sowing and the last

340 harvest. As it is well known, in fact, many authors report the reduction in salinity tolerance with the
341 increase in the duration of exposure to saline stress and the increase in the air temperature and the
342 evaporative demand of the atmosphere. In fact, the application of saline water in the presence of
343 high temperature conditions exacerbated the process of salt accumulation and plant growth
344 reduction (Helal and Mengel, 1981; Li et al., 2001; Meiri et al., 1982). From Maas and Hoffman's
345 model (1977) applied to the relative number of leaves and to the relative leaf surface, it is confirmed
346 that the reduced yield in relation to salinity was mainly due to the decrease in leaf surface and, to a
347 lesser extent, to number of leaves (Fig. 6, 7). Contrasting results, compared to those obtained in this
348 research, are reported by de Vos et al. (2013). Indeed, these authors observed that *D. tenuifolia*
349 grown in soilless conditions has begun the reduction in total fresh weight with the salinity of nutrient
350 solution greater than 100 mM NaCl (about 10 dS m⁻¹). Unlike these last results, Bonasia et al.
351 (2017) observed about 20% reduction in yield by increasing the salinity from 2.5 to 3.5 dS m⁻¹.
352 Differences in the results of the literature and those obtained in our research can be attributed to the
353 different growing conditions. In particular, in our research the direct sowing of wild rocket in saline
354 soil was carried out. In this way seedlings experienced salt stress since the germination phase. de
355 Vos et al. (2013), instead, used a 15-days seedlings that had been subjected to saline stress 6 days
356 after transplantation (when they had already spend beyond 50% of the vegetative cycle). Bonasia et
357 al. (2017), on the other hand, expose the wild rocket seedling to salt stress immediately after
358 emergence. We believe that direct sowing is the most appropriate method for assessing the salinity
359 tolerance of this species, as direct sowing represents the most common technique for the production
360 of wild rocket for baby leaf.

361 **Conclusions**

362 *D. tenuifolia* has been more productive than *D. muralis*. The increase in soil salinity has led to a
363 progressive reduction in water use. This aspect is to be considered in irrigation scheduling to save
364 water and to avoid the risk of water excesses with negative effects on the environment and yield.
365 Until moderate salinity levels, the Y_WUE has not undergone variations while it has decreased with
366 high salinity. *D. muralis* and *D. tenuifolia* rank among the species moderately sensitive to salinity
367 according to the Maas and Hoffman (1977) model. *D. tenuifolia*, compared to *D. muralis*, has a
368 milder salinity sensitivity. The information obtained in this research may be useful to farmers,
369 operating in salty soils or forced to irrigate with brackish water, in order to apply appropriate
370 strategies to avoid significant yield decline or even crop failure. Specifically, taking into account
371 also the results obtained by de Vos et al. (2013), in the presence of high salinity, the transplant
372 technique could be used, though much more expensive.

373

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377

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Table 1. Values (\pm SD) of electrical conductivity of saturated paste extract of soil (ECe, dS m⁻¹) at sowing time and at each harvest in the two years. The values represent the average of the two genotypes.

Salinity treatments	2007				2008			
	Sowing	1 st harvest	2 nd harvest	3 rd harvest	Sowing	1 st harvest	2 nd harvest	3 rd harvest
S ₁	1.0 \pm 0.17	1.2 \pm 0.15	1.4 \pm 0.19	1.6 \pm 0.18	0.9 \pm 0.10	1.1 \pm 0.12	1.3 \pm 0.18	1.5 \pm 0.19
S ₂	2.1 \pm 0.18	2.2 \pm 0.14	2.4 \pm 0.19	2.5 \pm 0.20	2.0 \pm 0.16	2.1 \pm 0.18	2.3 \pm 0.20	2.4 \pm 0.21
S ₃	3.4 \pm 0.25	3.4 \pm 0.30	3.6 \pm 0.38	3.6 \pm 0.35	3.2 \pm 0.31	3.4 \pm 0.36	3.4 \pm 0.28	3.6 \pm 0.39
S ₄	5.3 \pm 0.37	5.5 \pm 0.29	5.8 \pm 0.35	6.1 \pm 0.44	5.4 \pm 0.42	5.4 \pm 0.33	5.5 \pm 0.46	5.7 \pm 0.46
S ₅	8.5 \pm 0.43	8.6 \pm 0.42	9.1 \pm 0.31	9.3 \pm 0.47	8.6 \pm 0.48	8.7 \pm 0.40	8.8 \pm 0.43	8.9 \pm 0.39
S ₆	12.3 \pm 0.40	12.6 \pm 0.53	13.0 \pm 0.60	13.1 \pm 0.61	12.4 \pm 0.58	12.4 \pm 0.55	12.6 \pm 0.41	12.8 \pm 0.63

Table 2. Effect of year and salinity on total and marketable yield, leaves number per plant, leaf area per plant, leaf dry matter, specific leaf area (SLA), leaf succulence (LS), evapotranspiration (ET) and water use efficiency (Y_WUE), of *D. muralis* and *D. tenuifolia*.

Treatments	Total yield (1) (g plant ⁻¹)	Marketable yield (1) (g plant ⁻¹)	Leaves number (2) (n. plant ⁻¹)	Leaf area (2) (cm ² plant ⁻¹)	Leaf dry matter (2) (g 100 g ⁻¹ FW)	SLA (2) (m ² kg ⁻¹ DM)	LS (2) (g FW cm ⁻²)	ET (1) (L pot ⁻¹)	Y_WUE (2) (kg m ⁻³)
<i>Diplotaxis muralis</i>									
Year (Y)	ns	ns	ns	ns	ns	ns	ns	ns	ns
2007	31.3	28.8	11.6	192.9	9.7	21.4	0.049	13.3	10.3

2008	32.7	29.3	12.2	197.6	9.5	20.6	0.052	14.3	9.8
Salinity Levels (SL)	**	**	**	**	**	*	*	**	**
S ₁	43.2 a	42.4 a	13.7 a	285.7 a	8.8 c	24.6 a	0.046 d	16.7 a	12.7 a
S ₂	42.2 a	41.2 a	13.8 a	274.5 a	8.9 bc	24.0 a	0.047 d	16.0 a	12.9 a
S ₃	39.1 b	36.9 b	13.6 a	248.7 b	9.2 b	22.4 ab	0.049 cd	15.1 b	12.2 a
S ₄	30.0 c	26.2 c	12.6 b	175.5 c	9.5 ab	20.7 b	0.051 c	13.7 c	9.6 b
S ₅	23.0 d	18.9 d	10.4 c	127.8 d	10.1 a	18.4 c	0.054 b	12.0 d	7.9 c
S ₆	14.5 e	8.9 e	7.6 d	59.2 e	11.0 a	16.0 d	0.057 a	9.3 e	4.8 d
Y x SL	ns	ns	ns	ns	ns	ns	ns	ns	ns

Diplotaxis tenuifolia

Year (Y)	**	**	*	*	ns	ns	ns	ns	*
2007	40.2	38.6	15.2	259.2	9.6	23.3	0.046	15.2	12.3
2008	51.4	47.0	17.5	314.0	9.8	22.3	0.047	15.6	14.6
Salinity Levels (SL)	**	**	**	**	*	**	*	**	**
S ₁	58.8 a	57.2 a	18.4 a	381.7 a	9.0 c	26.6 a	0.042 d	17.8 a	15.7 a
S ₂	57.7 a	56.0 a	18.3 a	374.8 ab	9.0 c	26.1 a	0.043 cd	17.5 a	16.0 a
S ₃	55.5 ab	53.4 b	18.9 a	356.7 b	9.3 bc	24.2 ab	0.044 cd	16.6 b	16.1 a
S ₄	45.9 b	41.4 c	17.2 b	277.7 c	9.7 b	22.4 b	0.046 c	15.2 c	13.7 b
S ₅	33.6 c	31.8 d	14.2 c	215.3 d	10.2 ab	20.0 c	0.049 b	13.7 d	11.6 c
S ₆	23.5 d	17.0 e	10.9 d	113.5 e	10.9 a	17.6 d	0.052 a	11.4 e	7.5 d
Y x SL	ns	ns	ns	ns	ns	ns	ns	ns	ns

ns, *, ** indicate *F* test not significant or significant at $P \leq 0.05$ and $P \leq 0.01$, respectively. Mean separation within columns by SNK test ($P \leq 0.05$).

(1) The values are the sum of three harvests.

(2) The values are the average of three harvests.

Figures captions

- Fig. 1. Effect of salinity on marketable yield obtained at each harvest in 2007 and 2008 for *D. muralis* and *D. tenuifolia*. Vertical bars indicate \pm SD (n = 4).
- Fig. 2. Effect of salinity on relative evapotranspiration of *D. muralis* and *D. tenuifolia*. Pooled data of the two years.
- Fig. 3. Relationships of relative marketable yield decrease [$1-(Y_a/Y_m)$] vs relative ET decrease [$1-(ET_a/ET_m)$], for *D. muralis* and *D. tenuifolia*. Pooled data of the two years.
- Fig. 4. Maas and Hoffman (1977) model applied to the relative marketable yield (sum of three harvests) of the two crop cycles for *D. muralis* and *D. tenuifolia*.
- Fig. 5. Trend of threshold and slope derived from Maas and Hoffman (1977) model applied to the relative marketable yield for each harvest for *D. muralis* and *D. tenuifolia*. Pooled data of the two years.
- Fig. 6. Maas and Hoffman (1977) model applied to the relative leaf area per plant (average of three harvests) of the two crop cycles for *D. muralis* and *D. tenuifolia*.
- Fig. 7. Maas and Hoffman (1977) model applied to the relative leaves number per plant (average of three harvests) of the two crop cycles for *D. muralis* and *D. tenuifolia*.

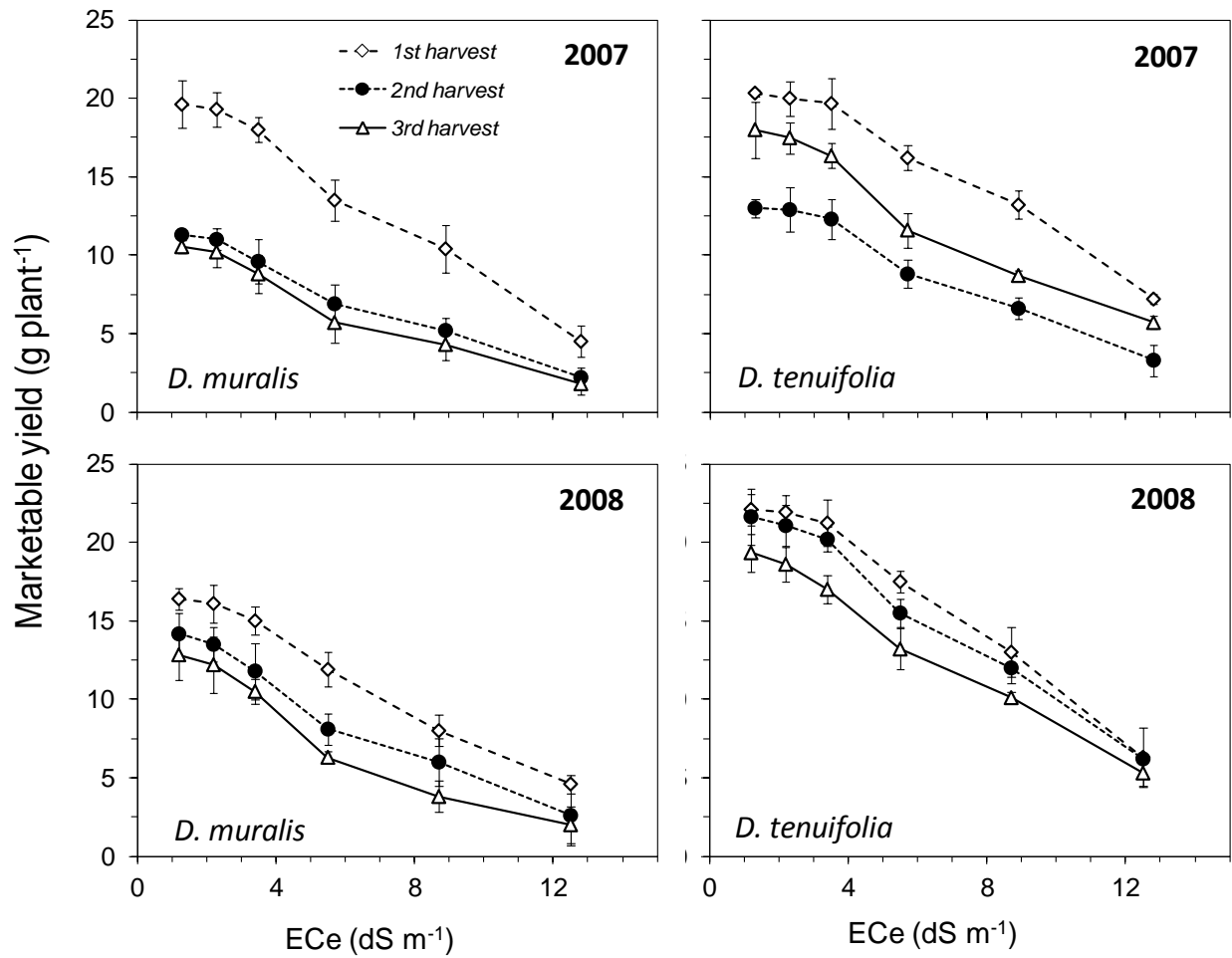


Fig. 1. Effect of salinity on marketable yield obtained at each harvest in 2007 and 2008 for *D. muralis* and *D. tenuifolia*. Vertical bars indicate \pm SD (n = 4).

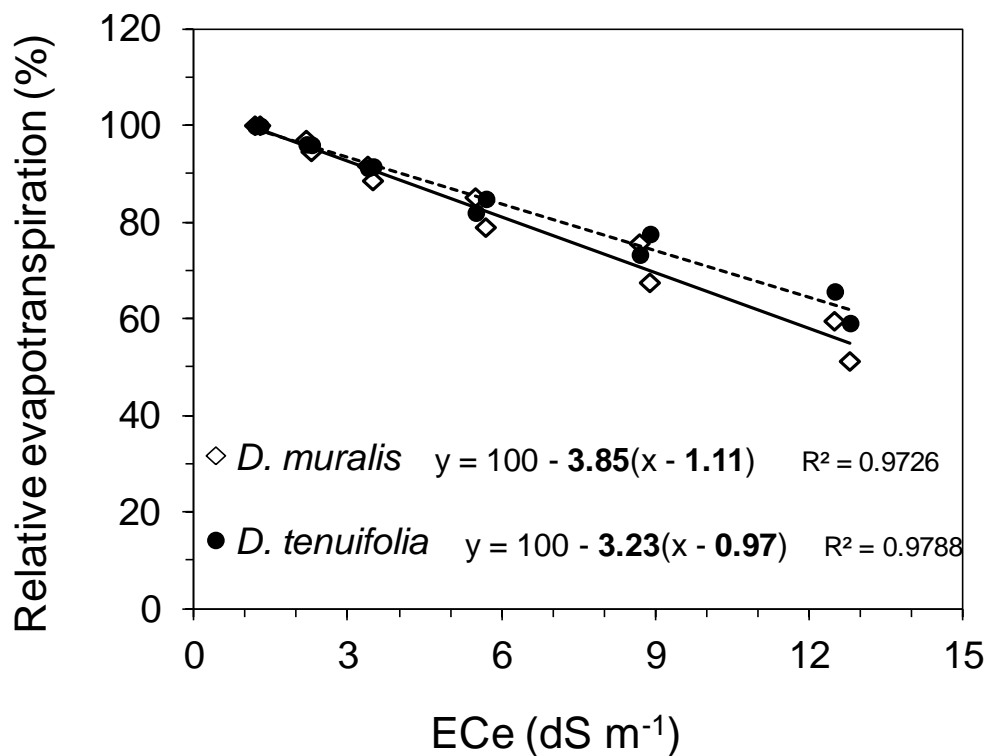


Fig. 2. Effect of salinity on relative evapotranspiration of *D. muralis* and *D. tenuifolia*. Pooled data of the two years.

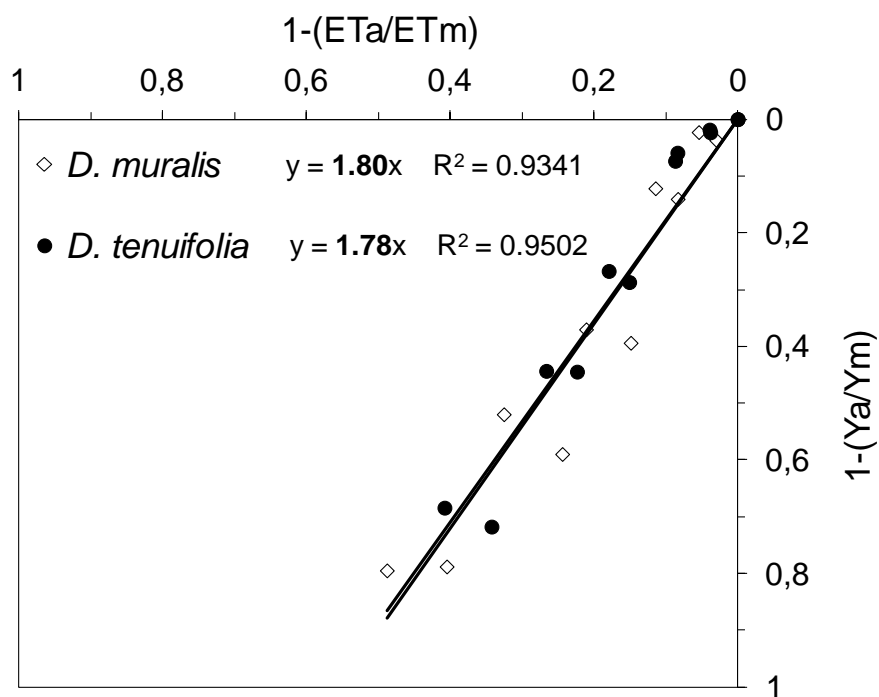


Fig. 3. Relationships of relative marketable yield decrease [1-(Ya/Ym)] vs relative evapotranspiration decrease [1-(ETa/ETm)], for *D. muralis* and *D. tenuifolia*. Pooled data of the two years.

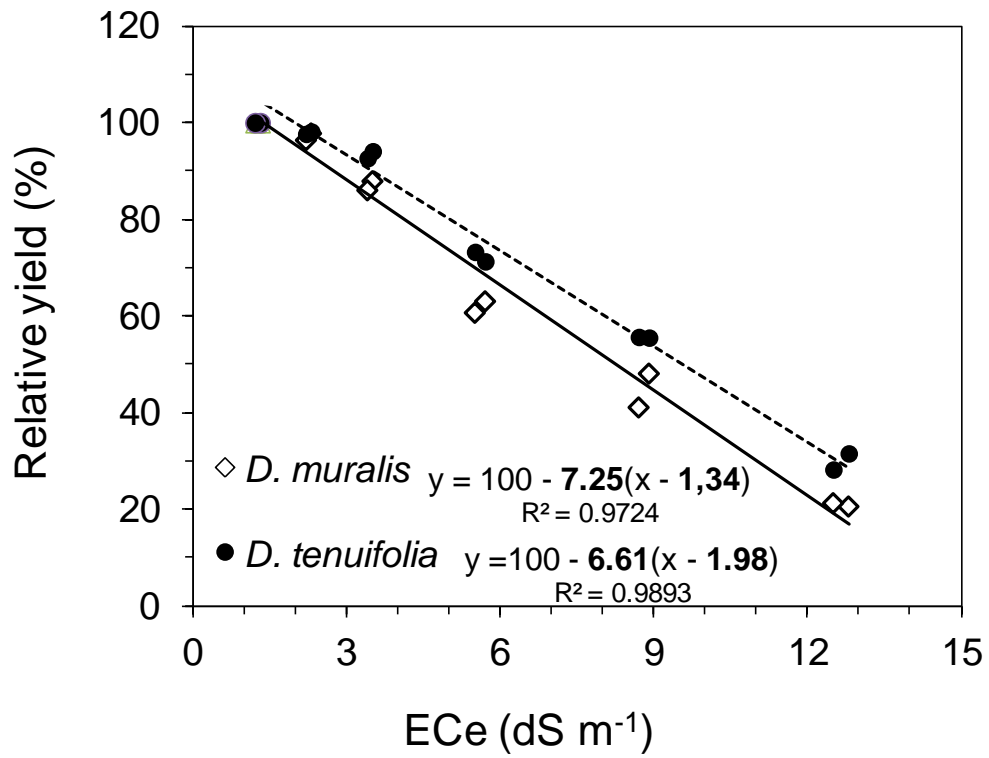


Fig. 4. Maas and Hoffman (1977) model applied to the relative marketable yield (sum of three harvests) of the two crop cycles for *D. muralis* and *D. tenuifolia*.

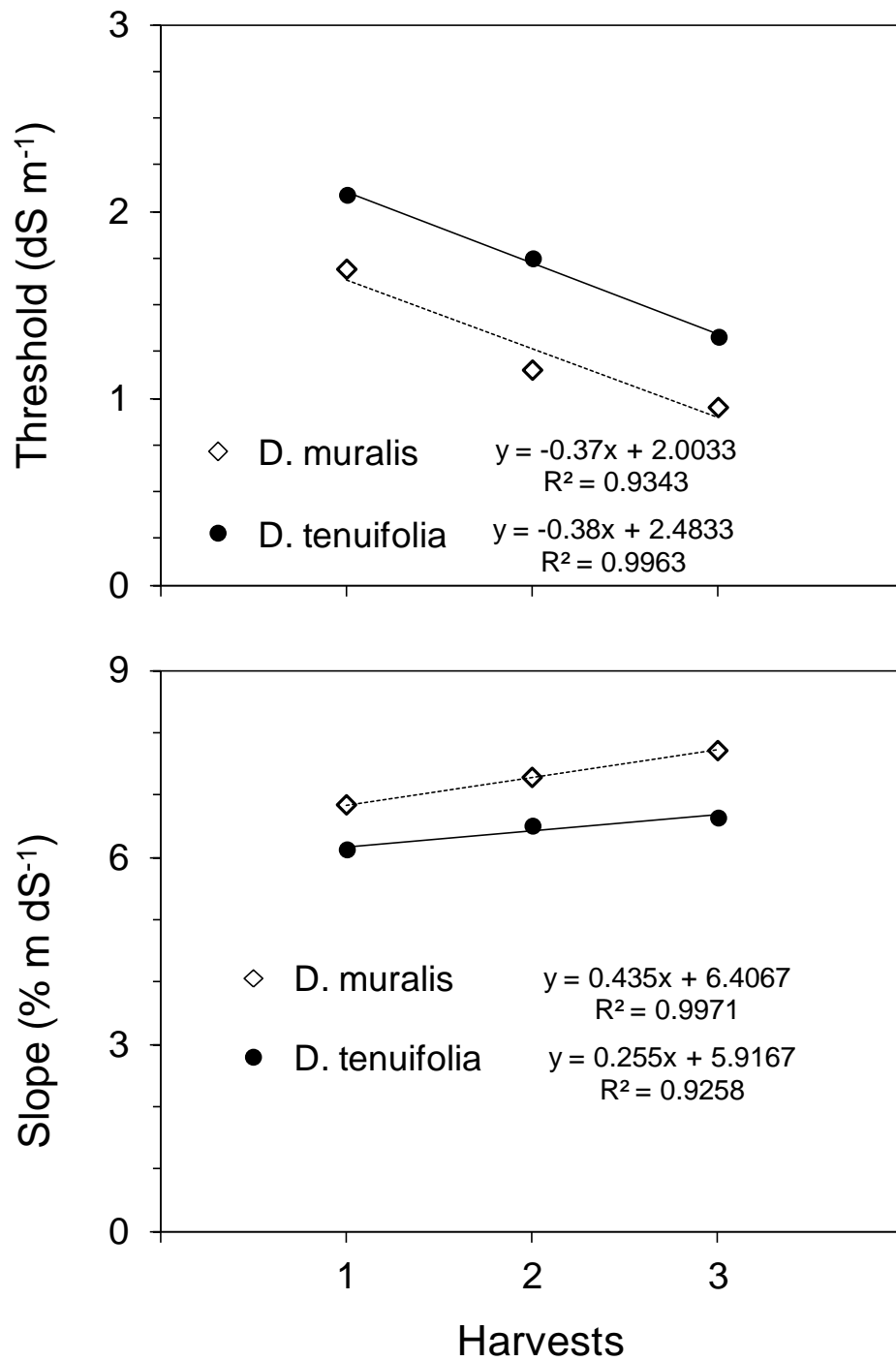


Fig. 5. Trend of threshold and slope derived from Maas and Hoffman (1977) model applied to the relative marketable yield for each harvest for *D. muralis* and *D. tenuifolia*. Pooled data of the two years.

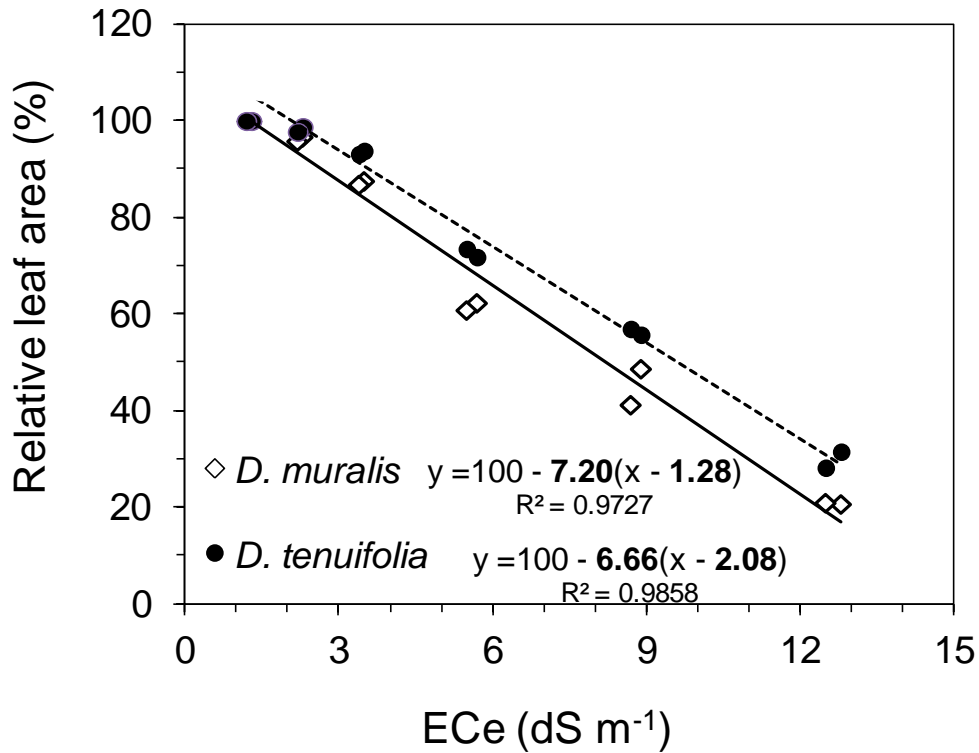


Fig. 6. Maas and Hoffman (1977) model applied to the relative leaf area per plant (average of three harvests) of the two crop cycles for *D. muralis* and *D. tenuifolia*.

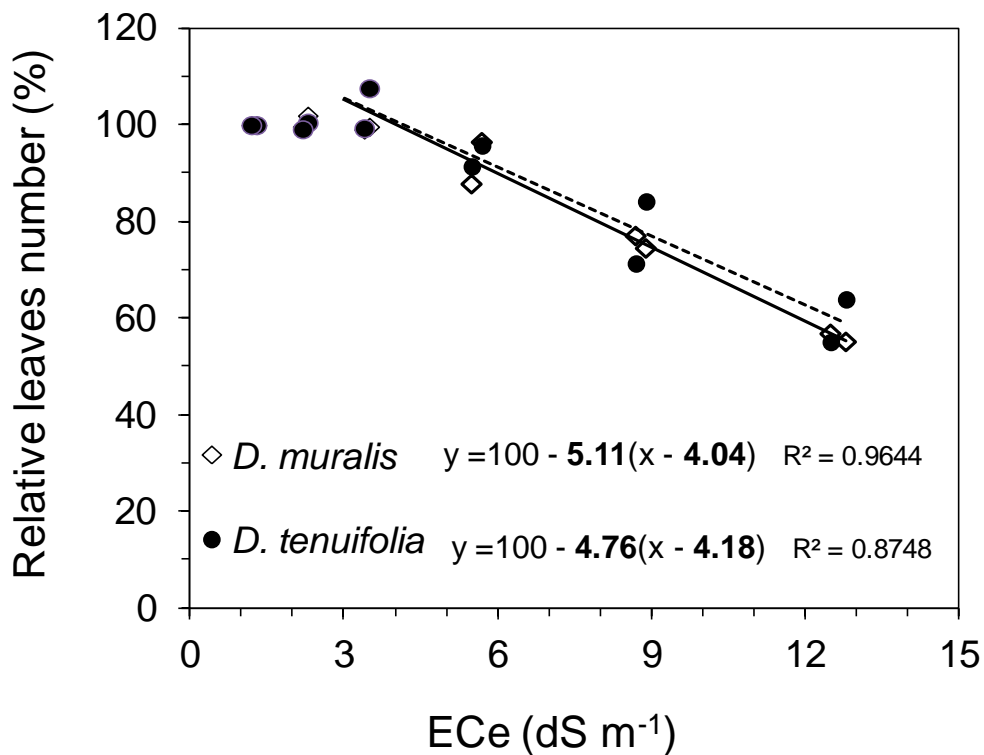


Fig. 7. Maas and Hoffman (1977) model applied to the relative leaves number per plant (average of three harvests) of the two crop cycles for *D. muralis* and *D. tenuifolia*.

