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Title: IRRIGATION SCHEDULING OF OLIVE ORCHARD BASED ON MIDDAY STEM WATER POTENTIAL

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Abstract: Irrigation scheduling of fruit trees according to water balance provided significant differences between locations. In recent years, water status measurements such as water potential have been suggested as irrigation tools in different fruit trees. The aim of this study was to adjust water potential threshold values previously studied and water application approaches that permit the irrigation scheduling of olive trees based on midday stem water potential. The experiments were performed during three seasons (from 2005 to 2007) in two different locations (Badajoz and Ciudad Real) with different weather and cultural conditions. In both locations, the olive orchards were seven years old at the beginning of the experiment but had significantly different canopy development. In Ciudad Real the canopy shaded area at the beginning of the experiment was 15% and the first crop was harvested in 2003. On the other hand, canopy shaded area of the olive orchard in Badajoz experiment was 40% and the first crop was harvested in 2001. Therefore, we assimilated Ciudad Real orchard as young, while Badajoz was mature. Three different irrigation treatments were compared in both locations: Control treatment with traditional water balance as irrigation scheduling and two treatments in which midday stem water potential (SWP) provided the information about water management. In the midday water stem potential irrigation (WI) the threshold value of SWP was -1.2 MPa before the beginning of the massive pit hardening period and -1.4 after this date. Finally, in the deficit treatment (DI) the threshold value of SWP was -2.0 MPa throughout the season. In WI and DI treatment irrigation was applied when SWP reached the threshold value. No significant differences were found between Control and WI in any of the seasons and locations when water potential, leaf conductance, shoot and fruit growth and yield (fruit and oil) were considered. In both locations, the same SWP value in WI treatment produced similar water application as the Control treatment. In DI treatment, shoot growth was significantly reduced in both locations in all the seasons. The SWP in DI trees was clearly affected in both locations, while leaf conductance was only reduced in the Badajoz experiment. In the Ciudad Real experiment no significant differences were found in fruit growth, whereas differences were found in Badajoz. However, yield was significantly reduced in Ciudad Real, but not in Badajoz. WI treatment was successful for no water stress conditions. On the other hand, DI treatment was a mild water stress treatment which reduced yield only in low covert orchard, but not in the ones with almost maximum canopy shaded area.

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Dear Dr.Clothier:

We should be grateful if you would consider the attached manuscript entitled

"IRRIGATION SCHEDULING OF OLIVE ORCHARD BASED ON MIDDAY STEM WATER

POTENTIAL" for publication in the Journal Agricultural Water Management.

Our work presents an approach for using midday stem water potential in the irrigation scheduling of olive trees. The experiments were performed in two different locations and during three years, in order to establish the usefulness of this approach. The results support that the threshold values suggested for no water stress conditions are the same though olive orchard were very different (as ours). In addition we discuss the use in deficit conditions.

All the authors have read the manuscript and approved it for publication.

Sincerely yours

Alfonso Moriana

<u>Highlights</u>

Irrigation was successfully scheduling only with midday stem water potential (SWP).

Control and no water stress SWP was similar in physiology measurements and yield.

No water stress SWP threshold was valid for different locations during three years.

Water applied in no water stress SWP treatment was similar to Control.

1	IRRIGATION SCHEDULING OF OLIVE ORCHARD BASED ON MIDDAY
2	STEM WATER POTENTIAL
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25 Abstract

26 Irrigation scheduling of fruit trees according to water balance provided significant 27 differences between locations. In recent years, water status measurements such as water 28 potential have been suggested as irrigation tools in different fruit trees. The aim of this 29 study was to adjust water potential threshold values previously studied and water 30 application approaches that permit the irrigation scheduling of olive trees based on 31 midday stem water potential. The experiments were performed during three seasons 32 (from 2005 to 2007) in two different locations (Badajoz and Ciudad Real) with different 33 weather and cultural conditions. In both locations, the olive orchards were seven years 34 old at the beginning of the experiment but had significantly different canopy 35 development. In Ciudad Real the canopy shaded area at the beginning of the experiment 36 was 15% and the first crop was harvested in 2003. On the other hand, canopy shaded 37 area of the olive orchard in Badajoz experiment was 40% and the first crop was 38 harvested in 2001. Therefore, we assimilated Ciudad Real orchard as young, while 39 Badajoz was mature. Three different irrigation treatments were compared in both 40 locations: Control treatment with traditional water balance as irrigation scheduling and 41 two treatments in which midday stem water potential (SWP) provided the information 42 about water management. In the midday water stem potential irrigation (WI) the 43 threshold value of SWP was -1.2 MPa before the beginning of the massive pit hardening 44 period and -1.4 after this date. Finally, in the deficit treatment (DI) the threshold value 45 of SWP was -2.0 MPa throughout the season. In WI and DI treatment irrigation was 46 applied when SWP reached the threshold value. No significant differences were found 47 between Control and WI in any of the seasons and locations when water potential, leaf 48 conductance, shoot and fruit growth and yield (fruit and oil) were considered. In both

49	locations, the same SWP value in WI treatment produced similar water application as
50	the Control treatment. In DI treatment, shoot growth was significantly reduced in both
51	locations in all the seasons. The SWP in DI trees was clearly affected in both locations,
52	while leaf conductance was only reduced in the Badajoz experiment. In the Ciudad Real
53	experiment no significant differences were found in fruit growth, whereas differences
54	were found in Badajoz. However, yield was significantly reduced in Ciudad Real, but
55	not in Badajoz. WI treatment was successful for no water stress conditions. On the other
56	hand, DI treatment was a mild water stress treatment which reduced yield only in low
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59	Keywords: Deficit irrigation, olive oil, water relations, water status.
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73 **1. Introduction**

74 Water is a scarce natural resource which is very important in agricultural practices. 75 Although irrigated lands are around 17% of the total agricultural surface, they provide 76 more than 40% of the total production (Fereres and Evans, 2006). However, the increase 77 of water scarcity in arid and semi-arid zones, the competition with other social uses 78 (such as sanitary, landscape uses) and the general feeling that irrigated agriculture is an 79 over-exploited system, are producing a decrease in the availability of water resources 80 for agricultural use. Regulated deficit irrigation (RDI) is a practice which was suggested 81 around the early 80's in peach trees (Chalmer et al., 1981) and consists of a reduction of 82 water applied during the most drought resistant phenological stages without a yield 83 penalty. From the first work in peach orchards, RDI has been a common research line in 84 most fruit trees (Bebohudian and Mills, 1997). Therefore, in most of the species the 85 drought sensitivity to water stress has been well described (Bebohudian and and Mills, 86 1997). Traditionally, RDI-scheduled irrigiation has been suggested in each phenological 87 stage as a fraction of the crop evapotranspiration (ET_c). But, when studies in different 88 locations are compared the results are very different (i.e., peaches, Girona, 2002).

89 This lack of results when different locations or/and cultivars are used, is 90 probably related to different agronomical conditions - mainly soil and/or phenological 91 development response. Because drought conditions are based on a percentage of ET_c 92 and not on physiological measurements, the same reduction in applied water produces 93 different water stress conditions. In the 1990's several authors suggested plant water 94 status measurements as an efficient tool for irrigation scheduling (Turner, 1990; Fereres 95 and Goldhamer, 1990). Huguet et al. (1992) and Shackel et al (1997) are probably the 96 first studies that suggested an approach for using the plant water status measurements 97 (trunk diameter fluctuations and water potential respectively) as a tool for irrigation
98 scheduling. At the beginning of the XXI century, different approaches with continuous
99 water status measurements were also suggested (sap flow, Nadezhdina and Cermaj,
100 1997; trunk diameter fluctutaions, Goldhamer and Fereres, 2001).

101 There are two main problems with using water status measurements as an 102 irrigation tool: the relationship of the values with environmental conditions (Hsiao, 103 1990) and the estimation of the amount of water to be applied. The great relationship 104 with environment means that the absolute value of the measurements are, in fact, the 105 sum of the effect of environmental and water stress conditions. Most of the approaches 106 suggest reference equations that link the indicator used with, usually, evaporative 107 demand (Shackel et al., 1997; Goldhamer and Fereres, 2001, Fernández et al. 2008). 108 Although other authors assume that when the influence of the environment is low a 109 unique threshold value could be used (i.e. in plum, Lampinen et al., 2001; in vineyards, 110 Girona et al. 2006) or a parameter which is not related with evaporative demand (i.e. in olive with predawn water potential, Gucci et al., 2007). 111

112 The second limitation is the estimation of the irrigation water amount. Most of 113 these measurements show the water stress level, but they do not provide any 114 information about water applied. Most of the approaches suggested using plant water 115 status measurements, in fact, as a secondary tool. They irrigated with an estimation 116 based on a percentage of ET_c and adjusted water applied only when the indicator is at 117 the threshold value (Lampinen et al 2001, Gucci et al., 2007). These approaches 118 suppose a small improvement compared to traditional water balance. On the other hand, 119 other studies are based on plant water status measurements and restricting the water 120 applied in order to establish a steady water stress level. Girona et al. (2006) suggested

irrigating with a great amount of water (4 to 6 mm day⁻¹) when midday leaf water potential is lower than a threshold value. The studies of Goldhamer with trunk diameter fluctuations (Goldhamer and Fereres, 2001; Goldhamer and Fereres, 2004) suggest a small increase in the amount of water is linked to the plant measurements. However, although the results of Goldhamer and Fereres (2004) in almonds were very satisfactory, Conejero et al (2011) reported a significant delay in peaches when a fast change in plant water status is scheduled.

128 In the last decades several plant and soil sensors have been suggested as 129 irrigation tools. Trunk diameter fluctuations (TDF), sap flow and water potential are, 130 nowadays, the most used in scientific studies. Several have reported that TDF is more 131 sensitive to water stress conditions than water potential (peaches, Goldhamer et al., 132 1999; olives, Moriana and Fereres, 2002) and sap flow (lemon, Ortuño et al., 2005). 133 However, water potential (WP) is a traditional technique in irrigation and water 134 relationship studies that is considered more reliable than TDF in some papers (olive, 135 Moriana et al., 2003; plum, Intrigliolo and Castel, 2006). Although, WP is a non-136 continuous and non-automatic measurement, the lower variability, lower cost and the 137 greater amount of data in the literature (compared to sap flow or trunk diameter 138 fluctuations measurements) make it more practical for commercial uses (Naor and 139 Cohen, 2003; Bonet et al., 2010; Moriana et al., 2010)

The aim of this study is to evaluate irrigation scheduling in olive trees based on midday stem water potential considering the situation of "non stress" and its use as a guideline for the application of controlled water deficit. We compare the results in water status, applied water and yield with the standard method of water balance. We hypothesized that the effect of evaporative demand and different cultivar and locations on the value of SWP is low. Therefore, the same SWP threshold will be used for
different orchards (difference in location and cultivar) and no reference equation will be
needed.

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149 **2. Materials and Methods**

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151 **2.1 Site description and experimental design**

152 The experiments were performed in two different locations: Ciudad Real and Badajoz 153 from 2005 to 2007. The cultivars were different, in Ciudad Real cv "Cornicabra" and in 154 Badajoz cv "Morisca" but both of them were for oil production. In both locations the 155 experimental design was a randomized complete blocks design with 4 blocks in Ciudad 156 Real and 3 in Badajoz. Each experimental plot was formed by two border lines with a 157 central line where measurements were performed. The measurements were performed 158 on Ciudad Real 2 trees per treatment and block and in Badajoz on 4 trees per treatment 159 and block.

160 In Ciudad Real, the experiment was performed in an olive orchard near Ciudad 161 Real, Spain (3° 56'W, 39° N; altitude 640 m). The trees, planted in the field in 1998, 162 were seven years old in 2005 with a canopy shaded area of 15% and the first crop (more 163 than 5 Kg per tree) in 2003. The climate of the study area is Mediterranean with an 164 average annual rainfall of 397 mm, mostly distributed outside a four-month summer 165 drought period. The soil is a shallow clay-loam (Alfisol Xeralf Petrocalcic Palexeralfs) 166 with a 0.75 m depth and a discontinuous petrocalcic horizon between 0.75-0.85 m. The volumetric water content for the first 0.3 m. (m m⁻³) was 22.8 % at field capacity (soil 167 168 matric potential of -0.03 MPa) and 12.1 % at wilting point (soil matric potential of -1.5 MPa) and 43.0 % and 21.1 %, respectively, from 0.3 to 0.75 m. Tree spacing was 7 m x
4.76 m (300 trees ha⁻¹). Drip irrigation (four emitters per tree providing 8 L·h⁻¹) was
provided daily.

172 In Badajoz, the experiment was performed in an olive orchard on La Orden 173 experimental farm near Badajoz (6° 40' W; 38° 51' N,; altitude 200 m.). The trees, 174 planted in the field in 1998, were seven years old in 2005 with a canopy shaded area of 175 40%. The first crop (more than 5 Kg per tree) was harvested in 2001. There was no crop 176 during the 2005 season which produced the beginning of an alternate bearing cycle from 177 2006. The climate of the study area is Mediterranean with an average annual rainfall of 178 463 mm, mostly distributed outside a four-month summer drought period. The soil is a 179 deep clay-loam (Alfisol Xeralf Tipic Haploxeralf) with a 1.5 m depth. The volumetric water content for the first 0.3 m. (m m⁻³) was 21.0 % at field capacity (soil matric 180 181 potential of -0.03 MPa) and 9.0 % at wilting point (soil matric potential of -1.5 MPa). Tree spacing was 6 m x 4 m (417 trees ha⁻¹). Drip irrigation (four emitters per tree 182 providing $4 Lh^{-1}$) was provided daily. 183

184 Meteorological data were measured in nearby automatic weather stations in each 185 location. The amount of rain (Table 1 and Fig. 1) was below the historical average in 186 2005 but greater in 2006 and 2007. During the end of the 2006 and the beginning of the 187 2007 seasons, rains were uncommonly higher. The rain value and distribution is 188 common in the Mediterranean climate with hot and dry summers (no rains) and cold 189 winters. The distribution and amount of rain in Ciudad Real and Badajoz were similar. 190 The maximum monthly temperatures were similar in both locations with very hot 191 summers (Fig. 1). Minimum temperatures were lower in Ciudad Real than in Badajoz. 192 In Ciudad Real, minimum temperatures of around -10°C were measured, especially 193 during 2005 season, while the monthly minimum in Badajoz was always higher than -194 8.0 °C (Fig. 1). The reference evapotranspiration, ET_o, was estimated using the Penman-195 Monteith equation employing daily data from the nearby automatic weather stations. 196 The seasonal ET_o values varied from 1160 mm to almost 1300 mm in Ciudad Real, 197 while in Badajoz, a warmer location, they were from 1263 mm to 1420 mm (Table 1). 198 The main difference in both locations is related to the more severe winters in Ciudad Real than in Badajoz, which clearly reduces the ET_0 and the growth season of the olive 199 200 orchards. The crop evapotranspiration (ETc) was estimated using the FAO method 201 (Doorenbos and Pruitt 1974), employing the crop coefficient (Kc) suggested for olive 202 trees (Orgaz and Fereres, 1997), with correction for the canopy size (Fereres and 203 Goldhamer, 1990). The seasonal ET_c was again clearly different but in this case more 204 related to the canopy shaded area. The values of ET_c in Ciudad Real increased with time 205 for the crown growth, while in Badajoz they were almost constant.

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207 2.2 Irrigation treatments

In all the treatments irrigation was daily and was scheduled twice a week. Threeirrigation treatments were performed in both locations:

Control Treatment. Trees were irrigated with 100% ET_c, estimated as described
 above.

Midday stem water potential irrigation (WI). Trees were irrigated according to
 the midday stem water potential (SWP) measured, with the same threshold value
 for each location (the description of SWP measured is below). In the first year
 (2005) irrigation was applied when SWP was lower than -1.2 MPa in all the
 season. However, in mid-summer SWP values were lower than -1.2 MPa and it

was impossible to increase even though water applied was extremely great
(Table 1). The threshold values were changed in 2006 and 2007 seasons. Before
the beginning of the massive pit hardening SWP threshold value was -1.2 MPa
and after the beginning of this period was -1.4 MPa.

Deficit irrigation (DI). Trees were irrigated according to the midday stem water
 potential (SWP) measured, with the same threshold value for each location.
 Irrigation was applied when SWP was lower than -2.0 MPa.

224 In the Control treatment, irrigation started when we estimated that around 50% of 0.75m 225 depth water profile was consumed. In WI and DI treatment, the irrigation was scheduled 226 twice a week with the SWP measurements of 2 trees per treatment in three blocks in 227 Ciudad Real and 4 trees per treatment in one block in Badajoz of the experimental 228 orchards. In both treatments irrigation started when SWP was statistically lower than the 229 threshold (T-test for comparison). The approach for water applied was to apply the first 230 irrigation event at 1 mm and then change according to the deviation of the SWP from 231 the threshold:

- When deviations were lower than 10%, the variation in the irrigation was 0.25
 mm day⁻¹
- When deviations were between 10-20%, the variation in the irrigation was 0.5 mm day⁻¹
- When deviations were between 20-30%, the variation in the irrigation was 1 mm day⁻¹

When deviations were higher than 30%, the variation in the irrigation was 2 mm day⁻¹

240 If according to this approach the water applied was negative the irrigation was stopped.

241

242 **2.3 Measurements**

243 The water status of trees of each treatment was characterised by the midday stem 244 water potential (SWP) and leaf conductance. Leaves near to the main trunk were 245 covered with aluminium foil at least one hour before measurements were taken. The 246 water potential was measured at midday, using the pressure chamber technique every 247 two weeks in 8 trees (Ciudad Real) or 12 trees (Badajoz) per treatment. The comparison 248 of the SWP measurements performed twice a week (for irrigation scheduling, described 249 above) and every two weeks (for water status monitoring) showed that the pattern was 250 similar (data not shown).

In order to describe the effect of the different irrigation strategies, the water stress integral (S_{Ψ}) (as defined by Myers (1988)) was calculated from the SWP data in both locations and three seasons:

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255
$$S_{\Psi} = \sum (\Psi_{m}-c) n$$

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256 Where: $\Psi_{\rm m}$ is the average of stem water potential for any interval

c is the value of the maximum stem water potential in both locations and allthe seasons (-0.5 MPa)

n is the number of the days in the interval

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Abaxial leaf conductance was measured in both locations. In Ciudad Real, leaf conductance was measured around midday in 24 fully expanded sunny leaves per treatment (3 per tree) with a steady state porometer (LICOR 1600, Lincoln, Nebraska, U.S.A). This measurement provided the minimum daily value (Xiloyannis et al, 1988). In Badajoz, leaf conductance was measurement around 10:00 in 18 fully expanded sunny leaves per treatment (3 per tree) with a transient state porometer (AP4, Delta-T
Devices Ltd., Cambridge, U.K.). This measurement provided the maximum daily value
(Xiloyannis et al., 1988). We are aware that both measurements are not comparable.
However, according to literature, leaf conductance (maximum or minimum) is less
sensitive to water stress in olive trees than growth or water potential (Moriana and
Fereres, 2002). We therefore only consider it as indicator of the water stress severity.
Thus, significant reductions of leaf conductance show severe conditions of water stress.

Soil water content was measured in 1m profile along the season with FDR sensors (Diviner2000, Sentenk, Australia) in both locations. Several access tube (from 6 to 8) were installed in each plot between two trees, beside and in the middle of two drips and 0.40m and 1 m from the drip line. The data obtained in both locations did not presented any differences between treatment (data not shown).

277 At the beginning of each season eight shoots per tree were randomly selected, in 278 8 trees per treatment in Ciudad Real and 10 trees per treatment in Badajoz. In each 279 shoot the length, number of inflorescences and fruits were measured periodically. The 280 fruit volume was estimated from a survey of twenty fruits randomly selected in 8 trees 281 (Ciudad Real) or 6 trees (Badajoz) per treatment. Two measurements were made in each 282 fruit of this survey: the longitudinal dimension and the transversal (at the equatorial 283 point) dimension. The pattern of the longitudinal dimension indicated the beginning of 284 the massive pit hardening when the rate of growth of this measurement changed (Gijón 285 et al., 2010). The fruit volume was estimated with the water displacement of the fruit 286 sample. In addition, fresh and dry weight of fruit was also measured.

All of the experimental trees were harvested during Autumn when the maturation index was around 3.5 (Hermoso et al., 1997). The individual fruit yield of

289 each control tree was measured (8 trees for Ciudad Real and 12 trees per treatment for 290 Badajoz) and a sub-sample of 2 kg of fruits taken from each for oil determinations. Oil 291 was extracted using two methods. The Abencor system (Mc2 Ingenieria y Sistemas, 292 Seville, Spain), which emulates commercial oil extraction systems (so called industrial 293 oil content) and was expressed in percentage of the fresh weight. This system extracted 294 the oil only by mechanical methods like the commercial oil industries. The Soxhlet 295 extraction determined the total oil content in the fruit which was expressed in 296 percentage of fresh and dry weight. This system extracted the oil by chemical methods 297 and obtained all the fat in the fruit.

298

299 **2.4 Statistical analysis**

The experimental design was completely randomized blocks, with four blocks in Ciudad Real and three in Badajoz. The data were subjected to one-way ANOVA; means were compared using the Tukey test. Significance was set at P<0.05. The number of samples measured is specified in the text and figures. Regression analysis was performed to determine the relationship between yield, total oil content and shoot length vs the water stress integral.

306

307 **3. Results**

The data of midday stem water potential (SWP) are shown in Figs. 2 (Ciudad Real) and 309 3 (Badajoz). In the three seasons of the Ciudad Real experiment (Fig. 2) there was an 310 increase in the SWP values from the first measurement in February until data in Spring, 311 especially clear during 2006 season (Fig. 2b). Such differences were the same for all the 312 treatments and not related to soil moisture (data not shown). Since these lower values are not present in Badajoz experiment (Fig. 3), they were likely related to low temperature. The mean monthly temperatures during February in Ciudad Real were 3.6° C (2005), 4.9° C (2006) and 8.2 °C (2007), while the mean monthly temperatures during March in Badajoz, were higher mainly in 2005 and 2006 (12.7°C (2005), 12.2° C (2006), 11.5 °C (2007)).

318 Control midday stem water potential (SWP) in Ciudad Real (Fig. 2) and Badajoz 319 (Fig. 3) were similar in the seasonal pattern and even in the absolute values though the 320 canopy shaded areas of the orchards were significantly different (around 15% in Ciudad 321 Real and 40% in Badajoz, data not presented). Maximum values were recorded at the 322 beginning of the spring with values around -1.0 MPa and even higher (Figs. 2 and 3). The Control values slightly decrease until minimum SWP at mid-summer. Such 323 324 decreases usually occurred first in Badajoz, likely related to the evaporative demand in 325 both locations - slightly higher in Badajoz (370 mm ET_o from March to May) than in 326 Ciudad Real (347 mm ET_o from March to May) - and the canopy shaded area (higher in 327 Badajoz than Ciudad Real). However, in both locations minimum SWP was around -1.5 328 MPa (except precise data in 2007 due to a problem with irrigation in Badajoz). From the 329 beginning of September midday SWP tended to give higher values, around -1.0 MPa. In 330 the last data recorded in Control treatments there was a decrease in the SWP data, 331 especially in Ciudad Real (Fig. 2), but also in the 2007 season in Badajoz (Fig. 3) that 332 was likely related to temperatures lower than 10° C.

The SWP values obtained in the different irrigation treatments, however, were clearly different in both locations. In the Ciudad Real experiment, the treatment Deficit Irrigation (DI) was significantly different to Control and Water Potential Irrigation (WI) from the beginning of June (around day of the year, DOY, 150) in the 2005 and 2006 seasons (Figs. 2a and b), and from the beginning of July (DOY 180) in the 2007 season (Fig. 2c). The minimum value of this treatment, DI, was around -2.0 MPa, slightly higher in most of the dates in the 2005 and 2007 seasons. Although the autumn rains rehydrated DI treatment, the SWP values were still significantly lower at the end of the season. The treatment WI was not significantly different to Control in any of the seasons. The SWP values of WI were almost equal to Control on most of the dates, and only a low difference of ± 0.2 MPa was measured, but without a clear trend (Fig. 2).

344 The differences in SWP were faster and clearer in the Badajoz experiment (Fig. 345 3). The SWP values in DI treatment were significantly lower than Control from the 346 beginning of May in the 2005 and 2006 season (around DOY 125) but from beginning 347 of June in the 2007 season (around DOY 150). Therefore, one month before in Badajoz 348 than in Ciudad Real. The minimum values of DI trees were between -2.5 to -3 MPa, 349 lower in the 2007 season (Fig. 3c) than in 2005 and 2006 (Figs. 3a and b). The recovery 350 of DI trees during the Autumn was completed only in the 2005 season, during 2006 and 351 2007 significantly lower values than in Control were measured. There were significant 352 differences between Control and WI treatment in all the seasons. Some of these, only a 353 few, were related to irrigation problems that produced a sharp decrease in SWP as in 354 DOY 150 during 2007 season (Fig. 3c). However, the differences were no higher than 355 0.5 MPa and without a clear trend.

The effect of water stress is the sum of the strength (the values of water potential) and the time that the trees are in such conditions, which is traditionally called the length (Hsiao, 1990). The values of stress integral (Fig. 4) in Control and WI treatmenst were similar in all the locations and seasons. Moreover, in the 2005 and 2006 seasons the values of stress integral in Control and WI were similar even between locations, though the trees were very different in crown volume. The stress integral in
DI treatment was always significantly greater than Control and WI. Such differences
were especially great in the Badajoz experiment (Fig. 4b) where DI values were around
50% greater (they varied from 38% in 2007 to 80% in 2005) than Control and WI, while
in Ciudad Real differences were lower (from 32 to 47%).

366 The data of midday leaf conductance are presented in Figs. 5 (Ciudad Real) and 367 6 (Badajoz). The seasonal pattern of Control treatment in Ciudad Real was similar to the 368 other two treatments (Fig. 5). The values of midday leaf conductance slowly increased 369 along the 2005 season (Fig. 5a) from values around 100 to 500 mmol m^{-2} s⁻¹. However, during the 2006 and 2007 seasons, midday leaf conductance was more stable than in the 370 preceding year with values around 300 mmol $m^{-2} s^{-1}$ (Figs. 5b and c). Only with sharp 371 372 increases likely related to a period of heavy rains (109 mm in October of 2006 and 134 373 mm in April of 2007). In the Badajoz experiment, the seasonal pattern of Control and 374 WI treatment was always similar (Fig. 6). During the 2005 and 2007 seasons, the years of low fruit load for these treatments, midday leaf conductance oscillated between 200-375 300 mmol m^{-2} s⁻¹ (Figs. 6a and c). In 2006, the season of high yield for Control and WI 376 377 treatments, the values of midday leaf water potential increased from values around 200 until higher than 400 mmol $m^{-2} s^{-1}$ from the end of May until the end of September 378 379 (Fig. 6b).

Midday leaf conductance values in Ciudad Real were significantly reduced in DI treatment in comparison to Control and WI treatments in all the seasons (Fig. 5). In the 2005 season the period of lower values occurred from mid-July until the end of September, while no significant differences were found between Control a WI treatments (Fig. 5a). The same period of significant differences between DI and the other two treatments was measured during the 2006 season (Fig. 5b). However, in this
season significantly lower values of WI than Control were also measured in DOY 184
and 194 (Fig. 5b). No significant differences were found during the 2007 season (Fig.
5c).

389 The leaf conductance values of DI treatment in Badajoz (Fig. 6) were similar to 390 the ones described in Ciudad Real but with clearer differences. During the 2005 season, 391 the DI treatment was significantly the lowest value from the beginning of the 392 experiment until early November (Fig. 6a). There were also significant differences 393 between WI and Control, usually with higher values of the former, until mid-August, 394 when no significant differences were measured (Fig. 6a). The differences between DI 395 and the other two treatments were great and significant during the 2006 season, from 396 DOY 129 until 275. After that date, differences were still significant though lower than 397 in the rest of the year (Fig. 6b). The differences between WI and Control were lower, 398 but significant in most of the season. During 2007, DI treatment was more similar to 399 Control than in previous seasons (Fig. 6c). However, the midday leaf conductance 400 values of DI were significantly lower than Control and WI from the beginning until 401 DOY 291. From this date the values of midday leaf conductance in all the treatments 402 decreased but they were similar between them. The differences were lower but 403 significant between Control and WI treatments in most of the dates, but without a clear 404 trend.

The treatments based on water potential (WI and DI) presented clear differences in the irrigation amount. The applied water (AW) in Control and WI treatments were similar in both locations during 2006 and 2007 season, while during 2005 WI treatment used more water than Control (Table 1). Such variations in AW during the first season

409 were related to the threshold value of SWP. During 2005, the SWP threshold was -1.2 410 MPa throughout the complete season and in the summer period though the irrigation was increased greatly SWP values were significantly lower (Fig 2a and 3a). We assume 411 412 that this decrease was a small influence (only around 0.2 MPa) of evaporative demand, 413 mainly, and even fruit load, since the variations were greater in Ciudad Real (with yield) 414 than in Badajoz (without yield). Then, in the next seasons the threshold value was 415 decreased to -1.4 MPa from the beginning of the pit hardening period. This change 416 produced similar water status conditions between WI and Control and almost equal AW 417 in both locations (Table 1, Figs. 2b and c and 3b and c). The threshold SWP in DI 418 produced different needs of water between Ciudad Real and Badajoz (Table 1). While in 419 Ciudad Real the water applied was reduced greatly (AW was 23, 27 and 4% of Control), 420 in Badajoz the amount of water was higher - around 50% of Control (42, 44 and 65% of 421 Control) (Table 1). Such differences in both locations in the AW of DI treatment are 422 related to the difference in crown volume.

423 The vegetative development measured through shoot growth was clearly 424 affected by the irrigation treatment in the Badajoz experiment but only in one season in 425 Ciudad Real (Table 2). The DI values of the shoot length, the number of nodes and the 426 leaf area in the shoots were significantly lower than in Control and WI in all the seasons 427 of the Badajoz experiment. The reduction was higher than 50% in all the parameters 428 measured and especially great in shoot length and leaf area (Table 2). The values 429 obtained in WI and Control in the Badajoz experiment were more similar, though in 430 2005 and 2006 slight, but significant, higher values were found in WI than in Control. 431 However, during 2007, Control values tended to higher values than WI, only 432 significantly higher in shoot length. The differences between WI and Control were

usually lower than 10%, only during 2006 were differences between both treatments
higher than 15%. In the Ciudad Real experiment the values of all vegetative
measurements were similar between treatments and only in the 2007 season were shoot
length and number of nodes significantly lower. However, the trend in all the seasons
was to lower values of DI than in the other two treatments.

438 In order to analyse the influence of irrigation on the vegetative growth 439 throughout the season and not only in the final values, the shoot growth rates (SGR) are 440 presented in Figs. 7 and 8. The seasonal patterns of SGR are similar between the two 441 locations, though the length and the rate of the growth in Control treatment are much 442 greater in Badajoz than in Ciudad Real. The maximum SGR values in Ciudad Real (Fig. 443 7) are usually around DOY 150, while in Badajoz (Fig 8) it is slightly before, with 444 values around double. In the 2005 season the SGR was the lowest of the experiment in 445 Ciudad Real and only significant differences were found around DOY 197 with slightly 446 higher values of WI treatment (Fig. 7a). The period of growth in 2005 was the smallest 447 of the three seasons and no clear influence of irrigation was found. The 2005 winter was 448 extremely severe in Ciudad Real (Fig. 1a) with monthly minimum temperatures below 449 -8.0°C from January to March (-11.9°C, -8.3°C and -8.1°C). In the 2006 and 2007 450 seasons in Ciudad Real, the lowest value of the monthly minimum temperatures was 451 around -5.0°C and only in January of 2006 did it reach -9.2°C (Fig. 1a). On the other 452 hand, the values obtained in Badajoz never dropped below -8.0°C (Fig 1b). In the winter 453 of 2005, also the most severe, monthly minimum temperatures were -7.9°C, -6.0°C and -454 5.2°C from January to March and higher than -5°C in the other two seasons (Fig. 1b). 455 The differences in the maximum values and the growth period between years and 456 locations are likely related to these minimum temperatures. During the 2006 season in

457 Ciudad Real, the values of DI treatment were significantly lower in the periods from 458 DOY 107 to 123 and from DOY 201 to 262 (Fig. 7b). The maximum values were not 459 significantly different, but DI treatment stopped shoot growth around 50 days before 460 that WI and Control treatments. No significant differences were found between WI and Control treatment. During the 2007 season in Ciudad Real, SGR in DI treatment was 461 462 significantly lower than Control in the period from DOY 164 to 278, but no different 463 from WI treatment (Fig.7c). The period of growth was similar in the three treatments. 464 No significant differences were found between WI and Control, but SGR in WI trees 465 tended to produce lower values than Control.

466 The greatest differences in SGR were found in the Badajoz experiment (Fig. 8). 467 In the 2005 season, significant differences in SGR were found between DI and the other 468 two treatments from the beginning of the experiment until DOY 286 (Fig. 8a). The 469 reduction in SGR in DI treatment was very severe though the period of growth was 470 similar. The values of WI and Control SGR were similar, though at the beginning of the 471 experiment (from DOY 126 to 166) significantly higher values in Control than in WI 472 were measured and at the end of the experiment (from DOY 209 to 236), they were 473 significantly higher in WI than Control (Fig. 8a). The differences during the 2006 474 season were even greater between DI and the other two treatments (Fig. 8b) and 475 significant from DOY 131 until the end. The SGR values of WI were significantly 476 higher than Control only from DOY 213 until the end. Although the differences 477 between DI treatment and WI and Control were lower during 2007, they were 478 significant from DOY 92 until DOY 262 (Fig. 8c). The SGR values in WI and Control 479 were again similar. Although significant differences were found between these two

480 treatments, there was no consistent trend and alternate higher and lower values were481 found.

482 The number of inflorescences, number of fruits at 30 and 60 days after full 483 bloom and harvest in the marked shoots are presented in Table 3. In the 2005 season 484 only data in Ciudad Real were measured (Table 3). In Ciudad Real during the 2005 485 season, the number of inflorescences and fruit were significantly lower in DI than in WI 486 treatment. The differences between DI and Control only were significant in the number 487 of inflorescences. The number of fruits was almost constant from 30 days after full 488 bloom. The number of fruits per shoot at harvest was not significantly different between 489 Control and DI though the number of inflorescences in spring was higher in Control. In 490 the 2006 season in Ciudad Real, no significant differences were found in the number of 491 inflorescences but the number of fruits at harvest was significantly higher in DI than in 492 WI treatment. No significant differences were found during the 2007 season in Ciudad 493 Real though the number of inflorescences and fruit at harvest tended to produce lower 494 values in Control trees.

495 In the Badajoz experiment two very different seasons were measured (only 2006 496 and 2007) (Table 3). During the 2006 season WI treatment was significantly the greatest 497 in the number of inflorescences, while Control was slightly lower and DI trees were 498 around half. However, there were a sharp decrease in the number of fruits 30 days after 499 full bloom in WI treatment. At this date Control was significantly the greatest (twice as 500 much as WI) and DI significantly the lowest. Such differences were reduced through the 501 season and at harvest - Control treatment was significantly higher than WI and DI 502 treatment which were almost equal. In this period, from 30 days after full bloom to 503 harvest, the reduction in the number of fruits was higher than 50% in all the treatments.

In the 2007 season, the number of inflorescences was significantly different between all the treatments but in the opposite direction. The number of inflorescences in DI treatment was significantly the highest and Control presented the lowest. The number of fruits was constant from 30 days after full bloom with significantly higher values in WI and DI than in Control. However, the differences in the number of fruits per shoot between Control and DI treatment were lower (30% less in Control) than the differences in inflorescences (50% less in Control).

511 The seasonal pattern of fruit volume and fruit dry weight in the two locations 512 during the 2006 and 2007 season is presented in Figs. 9 and 10. The results of the 2005 513 season in Ciudad Real were similar and are not presented in order to reduce the number 514 figures. In both locations, the seasonal pattern of fruit volume and fruit dry weight was a 515 continuous increase until the end of the summer (Figs. 9 and 10). The fruit volume in 516 Ciudad Real experiment (Fig. 9a and b) was almost equal in all the treatments, without 517 significant differences through the two seasons. However, in 2006 season (Fig. 9c) the 518 fruit dry weight in DI treatment was significantly greater than the other treatments at the 519 end of the experiment (DOY 320 and 339). Such results were not repeated in the next 520 season, when no significant differences were found (Fig. 9d). In Badajoz, fruit volume 521 was significantly affected by water stress in DI treatment during the 2006 season from 522 the beginning of the experiment (Fig. 10a). In the next season the differences were 523 lower and only significant with WI treatment, from the beginning of the experiment 524 (only in the period from DOY 276-311 no significant differences were found), but there 525 were not with Control treatment (Fig 10b). There were no significant differences in fruit 526 volume between WI and Control treatment in 2006, but in the 2007 season Control 527 tended clearly to produce lower values than WI. Similar behaviour was observed in fruit

dry weight between treatments. In both seasons, the fruit dry weight in DI was significantly lower than WI (Figs. 10c and d), only on the period from DOY 260-290 in 2007 season there were no significant differences. The differences in fruit dry weight between DI and Control were significant during the 2006 season and at the end of 2007 (from DOY 325 to DOY 346). No significant differences were found between WI and Control treatment, though WI tended to produce greater values than Control treatment in the 2007 season, especially at the end.

535 The results of yield, oil content and pulp-stone ratio (P/S) are presented in Table 536 4. The yield clearly separated Ciudad Real from the Badajoz experiment. In the Ciudad 537 Real orchard, with a lower canopy shaded area than Badajoz, the yield was smaller in 538 2006 and 2007 (there were no yield in the 2005 season in Badajoz). In the Ciudad Real 539 experiment, only the yield in DI was significantly smaller than WI and Control in the 540 2006 season, though the values of DI treatment in 2005 and 2007 tended to produce 541 lower values. The reduction in yield of DI treatment was 17% (2005), 33% (2006) and 542 7% (2007). The differences in yield between Control and WI were smaller than 4% in all 543 the seasons and they were not significant. In the Ciudad experiment, the values of oil 544 content (industrial and total content) were higher in DI than in Control and WI 545 treatments. Such differences were significant between WI and DI for the percentage of 546 total oil for dry weight values in both years and in fresh weight only in 2006 and for the 547 percentage of oil with industrial extraction in 2007. The differences in the industrial 548 extraction were around 4% higher in DI than in the other two. The total amount of oil 549 (in dry weight) ranged from 3-8%, depending on the treatment - higher in DI than in 550 Control and WI treatment. In comparison to these differences, WI and Control were almost equal in most of the oil parameters, with values slightly higher in Control. TheP/S ratio was not significantly different.

553 In the Badajoz experiment, the yield was significantly different in both seasons 554 but in the opposite way. In the 2006 season, WI and Control were significantly higher 555 than DI, but in 2007 DI yield was higher. The percentage of yield reduction and the 556 yield values itself were similar between seasons. The percentage of oil with industrial 557 extraction was not significantly different between treatments and only slightly higher in 558 WI and Control than in DI treatment. When the total amount of oil is considered in fresh 559 weight, WI and Control tended to produce higher values than DI, even significant in 560 2006. However, when total oil content is considered in dry weight DI reached the 561 highest values, though no significant differences were found. The P/S ratio was only 562 significantly higher in DI treatment than in Control and WI during the 2006 season. In 563 the next year, though the WI and Control yield were similar to the previous DI result, 564 the P/S ratio was almost equal.

565 Fig. 11 shows the relationship between the stress integral (SI) data (Fig. 4) with 566 the yield, total oil content (Table 4) and shoot length (Table 2). The relationship 567 between SI and yield was clearly different with the locations (Fig. 11a). In Ciudad Real the relationship was stronger than in Badajoz (r= 0.78^* ; R²=0.55 in Ciudad Real and 568 569 non-significant in Badajoz). In the Ciudad Real experiment, the increase in SI in the 570 season decreased the yield. While in the Badajoz experiment the alternate bearing 571 produce that for a similar SI the yields obtained were different. But, also in Badajoz, the 572 trend of decrease in yield with SI is also clear. However, the reductions are very 573 different. While in Ciudad Real, SI values around 250 MPa day reduced the yield by 574 70%, in Badajoz SI values around 350 MPa day only reduced the yield by 30%. The

575 relationship between SI and shoot growth was also different between locations (Fig. 576 11b). In both locations, an increase in SI reduced the shoot length, but such reductions 577 were slower in Ciudad Real than in Badajoz. In addition, no significant correlations 578 were found in Ciudad Real, but they were in the Badajoz experiment (r=0.96**; 579 R^2 =0.89). In the Ciudad Real experiment the increase of SI from around 100 to higher 580 than 200 MPa day, reduced the shoot length by 35%. While in the Badajoz experiment, 581 maximum shoot length was around 200 MPa day and when values higher than 300 MPa 582 day was measured, length was reduced by more than 70%. Finally, the relationship 583 between the total oil content expressed in dry weight (TD) and integral stress was 584 similar between locations (Fig. 11c). If the data of Fig. 11c of locations are considered 585 together a significant correlation is calculated (r=0.77**). The increase in SI produced 586 and increase in TD, from 150 MPa day with 42% until 55% with a SI of 350 MPa day.

587

588 **4. Discussion**

589 The influence of evaporative demands and cultivar was low in the midday stem water 590 potential values and the same threshold of SWP (-1.2 MPa before and -1.4 MPa after 591 massive pit hardening) for no water stress conditions was reliable in the two locations studied. In addition, the irrigation scheduling of WI treatment provided a similar 592 593 amount of water applied as Control trees in both locations. Therefore, the same 594 threshold values are useful for different conditions. There are a few publications, from 595 our knowledge, that reported a significant relationship between SWP and vapour 596 pressure deficit (VPD). Moriana and Fereres (2004) reported that the influence of 597 vapour pressure deficit VPD in the SWP values in olive trees is small in no water stress 598 trees. In addition, since the period of irrigation scheduling in olive orchards is

599 commonly characterised by a very high and stable VPD, then no great variations would 600 be expected especially at midday. Other studies in the literature suggest irrigation scheduling in fruit trees with SPW and do not consider the effect of evaporative demand 601 602 (i.e. in prunes, Lampinen et al. 2001). Another factor in the SWP values is the fruit load. 603 Martín-Vertedor et al (2011) reported a significant influence of fruit load in the water 604 relations of olive trees. However, since the data of Badajoz during the 2005 season with 605 no yield were also very different in AW between WI and Control treatment (12% 606 higher) the influence on the selected threshold values was also not very great. We 607 assumed, from the data of the first year, a slight influence of evaporative demand and 608 irrigated with values of -1.2 MPa as reference before the massive pit hardening period 609 started and -1.4 MPa from the beginning of the massive pit hardening period. Although 610 in the present study, and in others in the literature, values of SWP are higher sometimes 611 in the irrigation season than these proposed (Moriana et al., 2003; Grattan et al., 2006; 612 Tognetti et al., 2006; Fernández et al., 2008; Iniesta et al., 2009; Correa-Tedesco et al., 613 2010; Gómez-del-Campo, 2010), we considered that such values are exceptional and the 614 influence is low. Only at the beginning and at the end of the irrigation period, when 615 VPD is very low, are higher values of SWP common than the ones suggested (-1.2 MPa 616 and -1.4 MPa), especially in autumn when -1.4 would be the threshold value. We 617 assume that during these short periods, especially autumn, we will apply mild water 618 stress conditions but, according to the results in the literature even around full bloom, 619 they will not reduce yield (Moriana et al., 2003; Iniesta et al., 2009; Fernandes-Silva, 620 2010). In addition, WI treatment was, in both locations, almost equal to Control 621 treatment in water relations, vegetative and reproductive growth and in yield. Only 622 outside of the irrigation period, in winter time, are the values suggested in WI treatment

clearly different to the ones suggested in full irrigated conditions. During the low
temperature period the SWP measurements are lower than the ones suggested (-1.2 and
-1.4 MPa) due to the chilling-induced-dehydration (Pavel and Fereres, 1998; PérezLópez et al. 2010), but in such conditions there is no irrigation needed.

627 The other main limitations to SWP measured as an irrigation tool is the 628 estimation of water applied. Traditionally, water potential has been used as a correction 629 of the traditional water balance method (Shackel et al., 1997; Gucci et al., 2007). 630 However, the estimation of crop coefficient (K_c) and coefficient of ground cover (K_r) is 631 difficult. As an example, in olive trees the model of Orgaz et al. (2006) that estimated 632 K_c and K_r in a unique crop coefficient, demonstrated that the traditional water balance 633 sub-irrigated most of the olive orchard, especially the youngest (Pérez-López et al., 634 2007). The approach suggested in the present study used SWP as the main tool in the 635 decision of water applied. Only if the SWP obtained is lower than the threshold, trees 636 are irrigated. Therefore, SWP values are an objective of irrigation and not a simple 637 control of the amount of water applied. In the present study, the applied water was 638 similar in WI and in Control treatment, a traditional water balance, with differences 639 lower than 7% in 2006 and 2007. Only during 2005, when the SWP threshold during 640 summer was the highest, were such differences were clearly marked. Pérez-López et al 641 (2007) reported clear differences between Orgaz's model and water balance method 642 with a reduction of around 20% in crown volume. However, such differences may only 643 be important in young orchards and probably less important that the ones reported by 644 Pérez-López et al. (2007) when pruning was considered. The main limitation of our 645 approach is the interval between SWP measurements. In the present study irrigation 646 scheduling was done every two-three days, but the pattern of the SWP was similar in

both locations to the ones obtained every two weeks (data not shown). However, we areaware that this interval, two weeks, may be too long in soil with low water retention.

649 The reduction in the vegetative growth in DI treatment was more severe in 650 Badajoz than in the Ciudad Real experiment. Such differences were related to the length 651 and the severity of the water stress (Hsiao, 1990) and the canopy development. 652 Although the minimum values were similar (around -2 MPa), the higher stress integral 653 reduced all the growth measurements more in Badajoz than in Ciudad Real. Growth is a 654 very sensitive process to drought (Hsiao, 1990) and in young olive trees it is reduced 655 even when no clear differences in water potential have been reported (Pérez-López et 656 al., 2007; Correa-Tedesco, 2010; Fernandes-Silva, 2010). Gómez-del-Campo (2008) 657 suggested that values around -1.5 MPa of SWP in young olive trees would reduce shoot 658 growth to 66% of the maximum, while at around -1.8 MPa the reduction would be 659 around 50%. However, similar reductions were not found in the same orchard the year 660 before when SPW reached these values (Gómez-del-Campo, 2010). Such results are 661 probably related to the length of the water stress, higher in one year than in other 662 (Gómez-del-Campo et al., 2010). Therefore, in young olive orchards (orchards with no 663 or very low yield) deficit irrigation scheduling with precise threshold values of SWP 664 would not be the most accurate recommendation in order to optimise vegetative growth. 665 However, if irrigation scheduling maximizes the period of low water stress conditions 666 (with SWP around -1.2 MPa), the reduction in the seasonal stress integral would likely 667 provide the best results.

668 The yield response to the irrigation treatment proposed was different between 669 locations. In the Ciudad Real experiment, DI trees tended to produce lower yield in all 670 the years and the differences were significant in 2006. The biannual values in Control

(9.7 and 11.5 Kg tree⁻¹) and WI (9.5 and 11.1 Kg tree⁻¹) were greater than DI treatment 671 (7.2 and 9.2 Kg tree⁻¹). However, in Badajoz the biannual yield was almost the same 672 between treatments (22, 19.4 and 19.2 Kg tree⁻¹). The canopy shaded area in the orchard 673 674 of the Ciudad Real experiment was low, with the first yield in 2003. Therefore, crown 675 volume limited the yield in comparison with Badajoz. In the Ciudad Real experiment, 676 no differences were found in the number of fruits per shoot or in the fruit volume. In 677 such conditions, the yield was very linked to growth and since growth was reduced, the 678 yield was also affected. Such a response would be likely related to the number of shoots 679 which were lower when we consider a young tree with a small canopy. Therefore, if a 680 limited number of shoots, for the size of the crown, is reduced by water stress 681 conditions, yield would be more affected than in a mature tree, where the great crown 682 volume will reduce the effect of lower shoot growth (as we discuss below).

683 On the other hand, the response of yield in the Badajoz experiment, a mature 684 orchard with a high canopy shaded area, is the sum of several factors. The water stress 685 conditions in DI treatment controlled the growth of the trees and reduced the alternate 686 bearing pattern produced for the 2005 season (no yield). Shoot growth is very important 687 in the yield of the next season in olive trees. The significant reduction in yield during 688 the 2006 season in DI treatment was less affected by the number of fruit per shoots or 689 fruit volume of the 2006 season in comparison with Control or WI than for the great 690 reduction in vegetative growth of the 2005 season. The growth of DI during the 2006 691 season in DI treatment was similar to 2005 and produced a similar number of 692 inflorescences per shoot in 2007 than in 2006. Therefore, the increment of around 44% 693 in yield in 2007 compared to 2006 in DI treatment was likely related to a better fruit set, 694 the same number of inflorescences per shoot produced greater fruit per shoots in 2007

695 than in 2006. This better yield result was likely related to a clear reduction in the stress 696 integral of DI treatment in 2007 in comparison to 2006, especially with the higher 697 values of SWP of this treatment at the beginning of the season. Until the beginning of 698 the pit hardening the stress integral during 2007 was 62.6 MPa day, while in 2006, 699 though the pit hardening occurred 13 days before, it was 96.6 MPa day. Fruit set is the 700 most sensitive phenological period in olive trees to water stress (Moriana et al., 2003) 701 but is less common in the climatic conditions where olive trees are grown. The 702 reduction in number of fruits per shoot in comparison with the number of inflorescences 703 was greater in WI and Control than DI during 2006, but it was likely related to fruit 704 load. Lavee et al (1999) reported an improvemnt in the fruit set when the number of 705 inflorescence is reduced. However, in the 2007 season, fruit set was even better in 706 Control and WI than DI treatment. Therefore, the threshold value of -1.2 MPa is likely 707 to be a reliable indicator for minimizing the water stress integral and obtaining an 708 adequate fruit set.

709 The pattern of the yield in Control and WI treatment in Badajoz showed that no 710 water stress conditions provided excessive vegetative growth, which induces more 711 severe alternate bearing. The level of water stress during the massive pit hardening 712 period, which was the most severe, affected the fruit growth but not the fruit number per 713 shoot in Badajoz. In summary, the level of water stress was apparently low during pit 714 hardening, even in DI treatment, and though the above effects were produced, they 715 permitted less alternate bearing in DI trees and a biannual yield similar to Control and 716 WI treatments. Water withdrawal during the massive pit hardening period is the 717 common recommendation in regulated deficit treatment in olive trees (Goldhamer, 718 1999; Alegre et al., 2002; Moriana et al., 2003). The level of water stress in our work

719 was lower than others reported (Alegre et al., 2002; Moriana et al., 2003) and, as such 720 then, even lower SWP threshold would be suitable. This lack of effect in water stress 721 during this period may have led, in recent years, to several authors suggesting water 722 stress conditions until the beginning of pit hardening (Patumi et al, 1999; Tognetti et al., 723 2006; Lavee et al., 2007). However, such a recommendation is sustainable only when 724 the water stress level is not severe (Goldhamer, 1999) and is not produced during fruit 725 setting period (Moriana et al., 2003). Some recent studies suggest continuous deficit 726 irrigation in olive trees (Moriana et al, 2003; Iniesta et al, 2009). These irrigation 727 schedules are only sustainable when the water stress levels are controlled, otherwise the 728 results will be changeable every season.

729 The accumulation of oil in the fruit was improved with water stress. The 730 industrial extraction, especially the total amount of oil in the fruit, was greater in DI 731 than in Control and WI treatment. Lavee and Wonder (1991) reported a decrease in the 732 oil accumulation in conditions of water stress. However, other authors reported an 733 increase in the percentage of oil in conditions of moderate water stress (Girona et al., 734 2002; Moriana et al., 2003; Lavee et al 2007; Iniesta et al., 2009). The relationship in 735 the present study between the stress integral and the total amount of oil in the fruit was 736 similar between both locations, though the cultivar and the soil cover were different. 737 The level of water stress and the phenological stage when drought promotes the increase 738 of oil content is not clear. There is no phenological indicator that provides information 739 about the oil accumulation. Moriana et al (2003) reported a significant increase in the 740 amount of oil from the end of July and several studies have reported significant changes 741 in the oil composition with water stress conditions during summer (Patumi et al., 1999; 742 Mangliulo et al., 2003; Moriana et al., 2007). However, Inglese et al (1996) increased the amount of oil with the irrigation of rain fed trees 80 days before harvest. More
information is needed to suggest a RDI schedule based on SWP in relation with fat
accumulation.

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747 **5.** Conclusions

The irrigation scheduling of olive trees with midday stem water potential (SWP) was performed successfully. The threshold values of -1.2 MPa before the beginning of the massive pit hardening and -1.4 MPa during this period and until harvest provided an irrigation scheduling almost equal to a traditional water balance method. Only during the chilling period would these threshold values not be adequate. The same SWP values in the two different orchards produced the same result of water applied and water status in comparison with Control treatment.

755 The treatments with a threshold value of -2.0 MPa clearly reduced vegetative 756 growth. Such reductions were clearly related not only to the minimum water potential 757 measured, but also to the length of the period of water stress. This irrigation scheduling 758 produced a different yield response. In an orchard with a low canopy shaded area, which 759 we may assimilate to a young orchard, the reduction in yield was strongly related to 760 vegetative growth. In a mature orchard, although the vegetative reduction was even 761 greater, the yield response was not always linked to it. Moreover, though the water 762 stress of this treatment slightly affected the fruit set and fruit growth, such effects 763 clearly only limit the yield in a single year, but not when biannual yield are considered. 764 In addition, in both locations, water stress increased the amount of total oil in the fruit. 765 Therefore, DI treatment was better irrigation scheduling in the mature olive orchard but

766	not in young	orchard.	However,	more	accurate	SWP	management	in	the	different
767	phenological s	stages in r	nature trees	shoul	d be inves	tigated	1.			

768

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930 Figure captions

Figure 1. Monthly rainfall (vertical bars) and maximum (solid lines) and minimum
(dash lines) temperature in Ciudad Real (a) and Badajoz (b) experiments from January
of 2005 until December of 2007.

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Figure 2. Seasonal patterns of midday stem water potential (SWP) during 2005 (a),
2006 (b) and 2007 (c) seasons in Ciudad Real experiment. Each point is the average of 8
measurements. Stars in the bottom represent the date when significant differences
between treatment were found. Symbols represent: ▲Control treatment; □ WI
treatment; ■ DI treatment.

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Figure 3. Seasonal patterns of midday stem water potential during 2005 (a), 2006 (b)
and 2007 (c) in Badajoz experiment. Each point is the average of 12 measurements.
Stars in the bottom represent the date when significant differences between treatments
were found. Symbols represent: ▲Control treatment; □ WI treatment; ■ DI treatment.

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Figure 4. Stress integral values during the three seasons of the experiment in Ciudad Real (a) and Badajoz (b). Stress integral were calculated with data of the midday stem water potential of Figs. 1 and 2 in the period 100 to 315. There were no significant differences between Control (solid box) and WI (oblique line box) treatments in any of the season or places. In all them, DI treatment (vertical line box) is significantly higher (Tukey Test; P<0.05).

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Figure 5. Seasonal patterns of midday leaf conductance during 2005 (a), 2006 (b) and
2007 (c) seasons in the Ciudad Real experiment. Each point is the average of 24
measurements. Stars in the bottom represent the date when significant differences
between treatments were found. Symbols represent: ▲Control treatment; □ WI
treatment; ■ DI treatment.

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Figure 6. Seasonal patterns of maximum leaf conductance during 2005 (a), 2006 (b) and
2007 (c) seasons in Badajoz experiment. Each point is the average of 18 measurements.
Stars in the bottom represent the date when significant differences between treatments
were found. Symbols represent: ▲ Control treatment; □ WI treatment; ■ DI treatment.

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Figure 7. Seasonal patterns of shoot growth rate (SGR) during 2005 (a), 2006 (b) and
2007 (c) seasons in Ciudad Real experiment. Each point is the average of 64
measurements. Stars in the top represent the date when significant differences between
treatments were found. Symbols represent: ▲ Control treatment; □ WI treatment; ■ DI
treatment.

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Figure 8. Seasonal patterns of shoot growth rate (SGR) during 2005 (a), 2006 (b) and
2007 (c) seasons in Badajoz experiment. Each point is the average of 80 measurements.
Stars in the top represent the date when significant differences between treatments were
found. Symbols represent: ▲ Control treatment; □ WI treatment; ■ DI treatment.

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Figure 9. Seasonal pattern of fruit volume (a and b) and fruit dry weight (c and d) in the
experiment of Ciudad Real during 2006 (a and c) and 2007 (b and d) seasons. Stars in

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977 the top represent the date when significant differences between treatments were found.
978 Each symbol is the average of 160 data. Symbols represent: ▲ Control treatment; □ WI
979 treatment; ■ DI treatment.

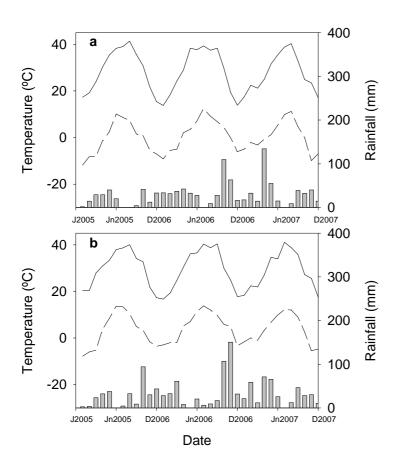
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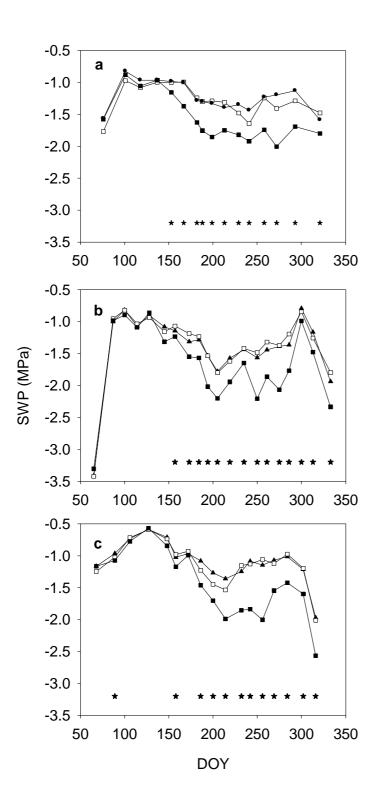
Figure 10. Seasonal pattern of fruit volume (a and b) and fruit dry weight (c and d) in
the experiment of Badajoz during 2006 (a and c) and 2007 (b and d) seasons. Each
symbol is the average of 120 data. Symbols represent: ▲Control treatment; □ WI
treatment; ■ DI treatment.

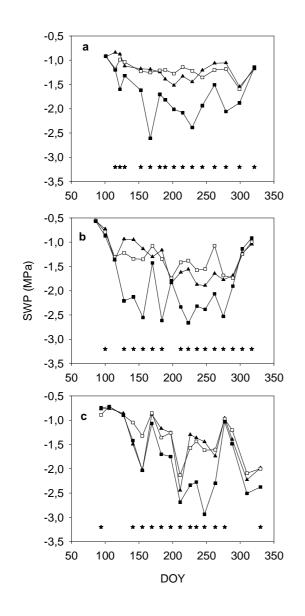
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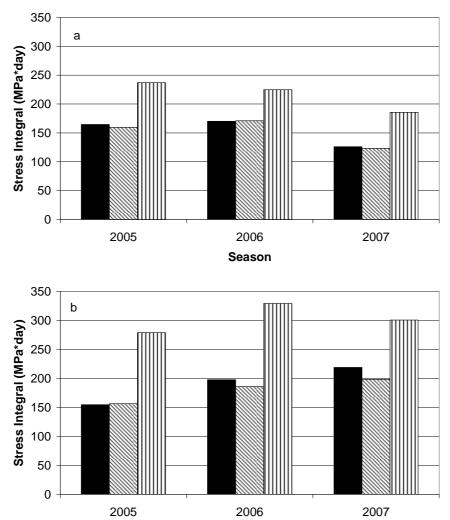
986 Figure 11. Relationship between the data of Stress integral (Fig. 3) and the yield (a), 987 shoot length (b) and total oil content (c) in Ciudad Real (\blacksquare) and Badajoz (\Box) 988 experiments. The data of yield and total oil content are from Table 4, the ones of shoot 989 length are from Table 2. Data of shoot length in 2005 season are not included. Lines 990 represent the equation regression of yield vs SI in Ciudad Real (a; Y=19.5-0.05X; 991 r=0.78; R^2 =0.55***; RMSE=1.7; n=9), shoot length vs SI in Badajoz (b; Y=37.9-0.01X; r=0.96; R^2 =0.89***; RMSE=2; n=6), total oil content vs SI with all the data (c; 992 Y=34.9+0.06X; r=0.77; R²=0.55***; RMSE=3; n=12). The regression in a and b which 993 994 are not presented are not significant.

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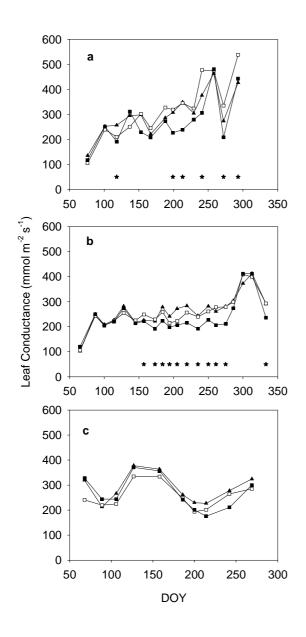


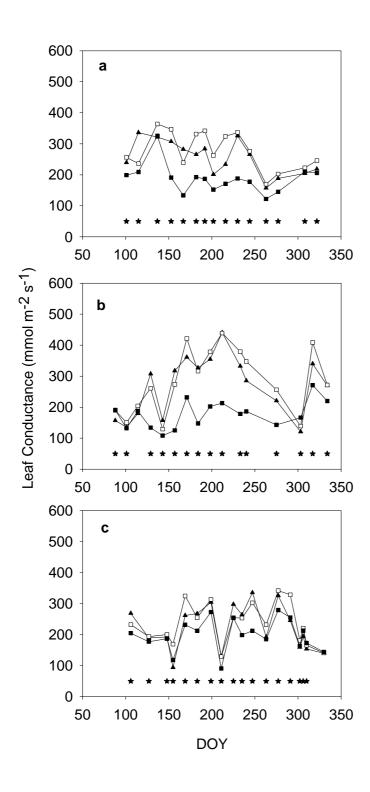


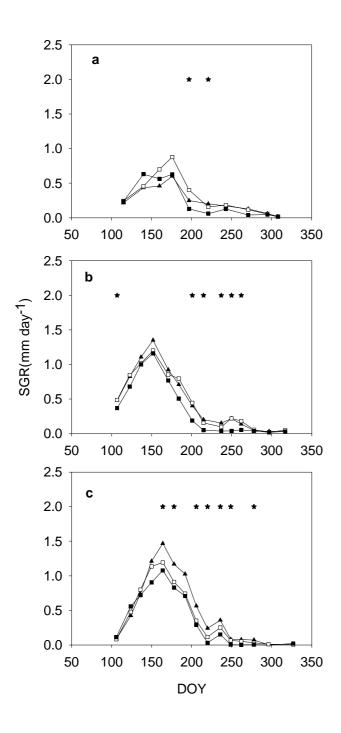


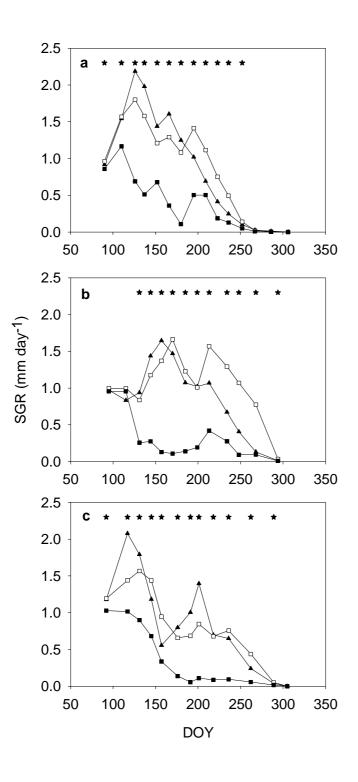
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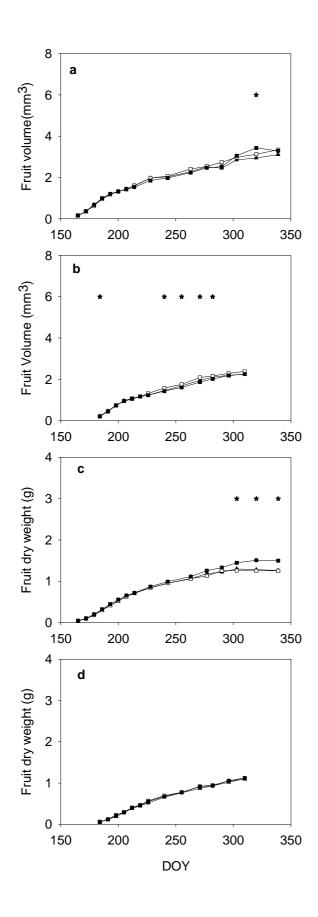
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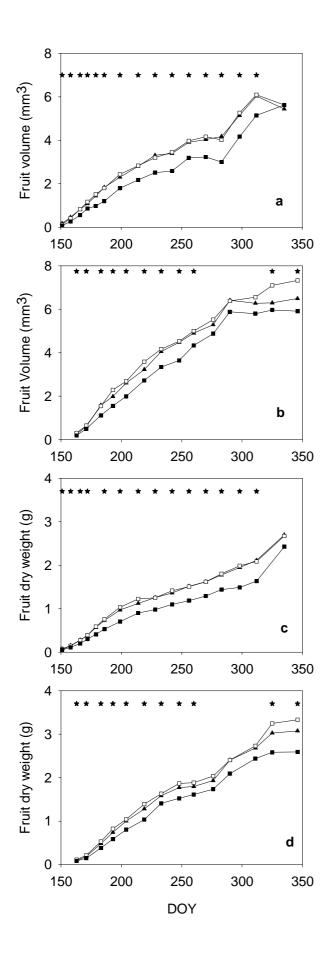


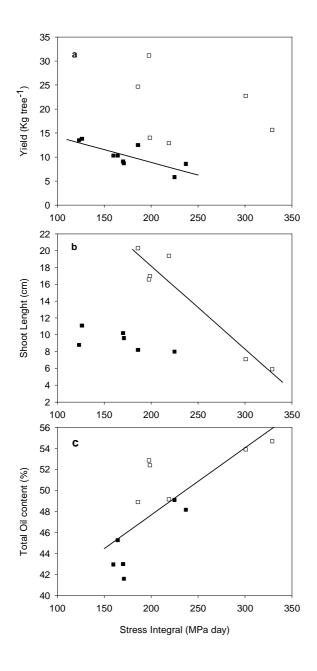












		Ciudad Real	1	Badajoz			
	2005	2006	2007	2005	2006	2007	
	AW (mm)	AW (mm)	AW (mm)	AW (mm)	AW (mm)	AW (mm)	
Control	125	93	113	380	410	296	
WI	180	101	108	427	388	305	
DI	29	25	5	161	180	193	
ET _o (mm)	1160	1299	1207	1420	1315	1263	
ET _c (mm)	190	222	248	546	597	529	
Rain (mm)	225	431	404	250	463	356	

Table 1. Applied water (AW, mm) in each treatment and location along the experiment. The seasonal reference evapotranspiration (ET_o) , seasonal crop evapotranspiration (ET_c) and total rainfall is also included.

Table 2. Shoot length (SL, cm), number of nodes (NN) and leaf area (LA, mm^2) of selected shoots at the end of the three seasons of the experiment and in the two locations (Badajoz and Ciudad Real). Different letter in the columns indicates significant differences within the location and the year (P<0.05. Tukey).

			Badajoz		(Ciudad Re	eal
		SL	NŇ	LA	SL	NN	LA
	Control	19,3 a	12,2 b	98,9 a	7.6	8.1	
2005	WI	20,2 a	13,3 a	103,5 a	7.6	7.7	
	DI	8,8 b	7,2 c	42,7 b	6.4	8.2	
	Control	16,6 b	11,6 b	73,9 b	10,2	9,6	
2006	WI	20,3 a	14,3 a	84,8 a	9,6	9,6	
	DI	5,9 c	6,5 c	34,0 c	8,0	8,5	
	Control	19.4 a	13.3 a	110.2 a	11.1 a	8.1 a	
2007	WI	17.0 b	13.0 a	106.6 a	8.8ab	6.7 ab	
	DI	7.1 c	7.1 b	56.0 b	8.2 b	6 b	

Table 3. Number of inflorescences (NI), number of fruit at 30 days after full bloom (NF1), number of fruit at 60 days after full bloom (NF2) and number of fruit at harvest (NFH) in the same selected shoots that Table 1. Different letters within the season and the location indicates significant differences (P<0.05. Tukey). There was no fruit yield in Badajoz in the 2005 season.

			Badajo	Z	Ciudad Real				
		NI	NF1	NF2	NFH	NI	NF1	NF2	NFH
	Control					14 a	4 ab	4ab	4 ab
2005	WI					16 a	5 a	5 a	5 a
	DI					11 b	3 b	4 b	4 b
	Control	22 b	8 a	4 a	3 a	7			2ab
2006	WI	23 a	4 b	2 b	1 b	8			2 b
	DI	12 c	2 c	1 b	1 b	6			3 a
2007	Control	5 c	2 b	2 b	2 b	9			2
	WI	9 b	3 a	3 a	3 a	11			3
	DI	11 a	3 a	3 a	3 a	12			3

Table 4. The table shows the data of yield, total percentage of oil (fresh weight) (TF) and with industrial extraction (IF), total percentage of oil (dry weight) (TD) and the pulp-stone ratio (P/S) in Ciudad Real and Badajoz during the experimental seasons. Different letters in the same row indicate significant differences within the location and the year (Tukey; p<0.05).

			Ciudad Real			Badajoz		
		Control	WI	DI	Control	WI	DI	
	Yield(kg tree ⁻¹)	10.3±1.5	10.3±0.5	8.6 ±0.9				
	% Oil (IF)	18.5 ± 1.1	18.7 ± 0.8	22.4 ± 1.0				
2005	% Oil (TF)	23.2±1.5ab	$22.3 \pm 0.8 b$	26.9±0.8a				
	% Oil (TD)	45.2±1.5ab	43.0±1.1b	48.1±0.8a				
	P/S	5.2±0.3	4.8 ± 0.1	4.6±0.1				
	Yield(kg tree ⁻¹)	9.1±0.5a	8.7±0.5a	5.8±0.3b	31.1±1.4a	24.7±2.1a	15.7± 2.9b	
	% Oil (IF)	12.2 ± 0.6 ab	10.7±0.6b	15.2±1.2a	17.9±0.3	16.8 ± 1.0	15.1 ± 0.3	
2006	% Oil (TF)	17.9±04	17.1±0.9	20.3±1.5	25.9±0.8a	$22.4 \pm 0.7 ab$	$21.1 \pm 0.5b$	
	% Oil (TD)	43.0±0.5ab	41.6±1.7b	49.1 ±3.3a	52.9±1.6	48.9±1.3	54.7 ± 1.5	
	P/S	4.4 ± 0.1	4.7±0.1	5.4 ± 0.2	6.9±0.3b	6.8±0.1b	9.1±0.2a	
	Yield(kg tree ⁻¹)	13.8±0.6	13.5±0.6	12.5±0.4	12.9±2b	$14.0\pm1.8b$	$22.7 \pm 0.8a$	
	% Oil (IF)				17.9±0.6	18.1±0.3	17.8 ± 1.3	
2007	% Oil (TF)				23.0±0.2	23.7±0.4	24.4 ± 0.5	
	% Oil (TD)				49.2±0.3	52.4±0.7	53.9 ± 1.4	
	P/S				6.9 ± 9.4	7.9 ± 0.04	7,7±1.3	