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Morphological and physiological characteristics in pomegranate cultivars with different yields

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Abstract. Variations in yield, percentage of hermaphrodite flowers, leaf mass to area ratio, leaf area, stomatal density, leaf gas-exchange and stem water potential (Ψ_{stem}) characteristics were studied in four pomegranate accessions. Yield was greatest in 11005 and Kallisti, compared with 11019 and 11021 accessions. Similarly, percentage of hermaphrodite flowers was greatest in 11005, followed by Kallisti which may be related with greater yields. In addition 11005 had highest photosynthetic rate and leaf area, but leaf mass:area was lowest. Intrinsic water use efficiency was highest, and stomatal conductance and transpiration were usually lowest in 11005 and Kallisti, a characteristic that may enable water conservation. Significant greater stomatal density was also found in Kallisti, which may suggest for a flexible stomata regulation to water deficit conditions. There were no significant differences among the studied genotypes in Ψ_{stem} .

Keywords. Hermaphrodite flowers – Leaf gas exchange – Leaf mass:area ratio – Stem water potential – Stomatal density.

I – Introduction

Understanding the relationship between yield and morphological and physiological characteristics is an important objective in crop breeding. Yield is the most important economic characteristic in pomegranate and is often recorded to vary greatly in different pomegranate genotypes. The abundance of hermaphrodite flowers would have a profound impact on yield since this will affect the number of fruit produced. Leaf gas exchange and leaf morphological characteristics have also been documented to affect yield in many species, however little is known for pomegranate. The present study aims to investigate various morphological and physiological parameters that may be related with increased yield in pomegranate.

II – Materials and methods

The experiment was conducted on the pomegranate accessions 11029 (named as Kallisti), 11005, 11019 and 11021, grown in a collection orchard at the Pomology Institute. The trees were 9 years old and planted in a 5 x 1.5 m distance in a randomised block design of six trees per genotype in two replicate blocks per tree. Mature fruits were harvested when most of their colour was red and total yield was measured. The numbers of hermaphrodite and male flowers were measured in 40 open flowers, three times during the flowering period, and the mean percentage of hermaphrodite flowers was calculated. Measurements of leaf area, leaf mass to area ratio (LMA), and chlorophyll content were made in leaves gathered from the middle part of young shoots in morning of August 19, 2010, from three trees of each accession. Leaf area was measured using a tracing technique in 30 leaves, and LMA was measured from the dry weight and leaf area of 10 leaves. Chlorophyll was measured in 9 leaves after extraction in 96% (v/v) ethanol for 48 h in the dark. Stomatal density was measured in artificial replicas of nail varnish

in the central region around the midrib of lower epidermis. For each leaf impression, five fields of view were selected for analysis.

Leaf gas exchange analysis was carried out with a LI-6400 portable gas exchange system (LI-COR Biosciences, Lincoln, USA). Measurements were carried out employing a PPFD of 1,700 $\mu\text{mol m}^{-2} \text{s}^{-1}$, c_a of 350 $\mu\text{mol m}^{-2} \text{s}^{-1}$ and leaf temperature of 28°C. The parameters determined were CO_2 carbon assimilation (Pn) and stomatal conductance (g_s) following Von Cammerer and Farquhar (1981). The ratio of Pn/ g_s was used as an estimate of short-term (instantaneous) leaf water use efficiency (WUE). Measurements were taken on the 5th of July, and 5th of August, from 9:00 to 11:00 hours, on six replicate leaves.

Stem water potential (Ψ_{stem}) was measured with a pressure chamber (Plant Moisture System, Skye instruments Ltd, Powys, UK). Leaves were previously placed in a black polyethelene bag wrapped in aluminium foil for at least 90 min before measurements to allow leaf water potential to equilibrate with stem water potential. Leaves were placed in the chamber within a few seconds after excision. Measurements were made in the next day after the gas exchange measurements, from 8:30 to 10:00 hours, on six replicate leaves.

Statistical analyses were performed using SPSS (SPSS Inc., Chicago, USA). Data were subject to ANOVA or MANOVA, and then significant differences between individual means were determined using the Duncan's multiple range test at the 5% level.

III – Results and discussion

Yield was greatest in Kallisti and 11005, compared with 11019 and 11021 (mean values 19.2 vs 8.3 kg, respectively), and may have resulted from a greater percentage of hermaphrodite flowers (63.8% in 11005, 37.6% in Kallisti, compared with 15.1% in 11019 and 11021), which suggests for a greater yield potential (Table 1).

Table 1. Mean values (\pm SE) of total yield and various morphological, and physiological characteristics in four pomegranate genotypes

	Kallisti	11005	11019	11021
Total yield (kg)	20.0 \pm 1.5 a	18.3 \pm 3.7 a	7.9 \pm 1.2 b	8.6 \pm 1.1 b
% Hermaphrodite flowers	37.6 \pm 9.7 b	63.8 \pm 6.1 a	13.4 \pm 2.1 c	16.7 \pm 3.8 c
Leaf mass per unit area (LMA, g m ⁻²)	92.8 \pm 4.0 a	79.9 \pm 4.2 b	84.4 \pm 3.9 ab	94.0 \pm 4.1 a
Leaf area (cm ²)	5.5 \pm 0.3 c c	7.8 \pm 0.3 a a	6.8 \pm 0.2 b b	6.8 \pm 0.2 b b
Photosynthetic rate (Pn, $\mu\text{mol m}^{-2} \text{s}^{-1}$)	9.5 \pm 0.7 b	13.0 \pm 0.6 a	9.0 \pm 0.7 b	10.0 \pm 0.6 b
Stomatal conductance (g_s , mmol m ⁻² s ⁻¹)	138.2 \pm 8.1 b	138.9 \pm 8.5 b	207.1 \pm 10.4 a	179.7 \pm 8.3 a
Transpiration rate (E, mmol m ⁻² s ⁻¹)	4.6 \pm 0.1 c	4.7 \pm 0.2 bc	6.3 \pm 0.3 a	5.7 \pm 0.2 ab
Water use efficiency (Pn/ g_s , $\mu\text{mol mol}^{-1}$)	84.2 \pm 10.3 b	96.0 \pm 16.4 a	49.9 \pm 6.5 c	58.2 \pm 4.3 bc
Stomatal density (stomata mm ⁻²)	149.9 \pm 4.1 a	101.0 \pm 7.1 c	122.6 \pm 2.0 b	68.8 \pm 3.1 d
Total chlorophyll _{mass} (mg g ⁻¹ DM)	6.4 \pm 0.6	7.0 \pm 0.5	7.7 \pm 0.2	6.9 \pm 0.6
Total chlorophyll _{area} (mg dm ⁻²)	5.9 \pm 0.5	5.6 \pm 0.4	6.5 \pm 0.2	6.5 \pm 0.5
Stem water potential (Ψ_{stem} , MPa)	-1.15 \pm 0.08	-1.24 \pm 0.07	-1.15 \pm 0.05	-1.08 \pm 0.04

Different letters denote significant differences at P \leq 0.05.

Increased levels of Pn was also found in 11005, compared with the rest studied accessions and may be related with the greatest assimilate demand from fruit (Drogoudi and Ashmore, 2000) and/or better developed photosynthetic mechanism. Leaf area was also greatest in 11005,

which may have provided greater availability of photosynthetic assimilates to accommodate yield. Total chlorophyll content expressed per dry mass or leaf area did not differ among the studied genotypes.

Leaf mass per unit area is an indicator of leaf thickness and the degree of mesophyll development within a leaf blade and is often related with greater photosynthetic capacity (Le Roux *et al.*, 2001). In the present study, although 11005 had highest Pn values and leaf area, LMA was lowest (Table 1). The present results coincide with the study of Mediavilla *et al.* (2001) where in intraspecific comparisons, found that Pn were higher in low LMA leaves, and may be due to a higher proportion of leaf nitrogen in the photosynthetic machinery. Reduced thickness (in low LMA leaves) should also tend to decrease the path length from the stomata to cell wall surfaces reducing gaseous diffusion resistance (Mediavilla *et al.*, 2001).

Intrigliolo *et al.* (2011) found that different irrigation treatments in pomegranate induced considerable differences in Pn and g_s , whereas there were no significant differences in Ψ_{stem} in spring and autumn, suggesting an near-isohydric behaviour of pomegranate trees. Similarly, in the present study there were no significant differences among the studied genotypes in Ψ_{stem} although they varied in Pn, g_s and WUE.

Intrinsic water use efficiency (Pn/g_s) was highest, and g_s and E were usually lowest in 11005 and Kallisti, a characteristic that may enable water conservation. Significant greater stomatal density was also found in Kallisti, which may suggest for a flexible stomata regulation to water deficit conditions.

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