



Research article

Yield and seed quality parameters of common bean cultivars grown under water and heat stress field conditions

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Abstract: Drought and heat stress strongly influence common bean development. The aim of this work was to study the effect of water stress on yield and quality traits of four common bean cultivars under *xerothermic* Mediterranean conditions. The field experiments were conducted under normal and water stress (50% of the normal) conditions applied 25 days after seed emergence (first flower buds visible). Agronomic, physiomorphological, quality traits and drought indices were assessed. Water stress reduced the number of pods plant⁻¹ (53%), seeds pod⁻¹ (9.7%), harvest index (49%) and seed yield (58%). Cultivar ‘Cannellino’ provided the higher performance both under normal and water stressed conditions and exhibited the lower drought susceptibility index and the highest mean productivity and geometric mean productivity across water treatments. These results were attributed primarily to the earliness of the cultivar ‘Cannellino’ which enabled avoidance of very high temperatures and severe drought and to the robustness and quick pod set. Valuable genetic variability was also observed for important quality seed traits (cooking time, protein). In conclusion, water stress is a significant limiting factor for seed yield and quality traits eventhough, suitable cultivars (i.e. ‘Cannellino’, ‘G. Northern’) were indentified in the course of this study. It is suggested that breeders select for early-flowering and quick pod-setting varieties to reduce the negative effects of water and heat stress in these environments.

Keywords: water stress; heat stress; *Phaseolus vulgaris* L.; seed yield; yield components; seed quality traits

Abbreviations: WT: water treatment levels; SY: seed yield; PPL: pods plant⁻¹; SP: seeds pod⁻¹; HSW: 100 seed weight; HI%: harvest index; PH: plant height; LEN: seed length; WID: seed width; HEI: seed height; HYI: hydration increase; HC: hydration capacity; SCP: seed coat proportion; CT: cooking time; SPC: seed protein content; DSI: Drought susceptibility index; DTI: Drought tolerance index; MP: Mean productivity; GMP: Geometric mean productivity; YRR: Yield reduction rate from non-stressed (%); YSI: Yield stability index; DII: Drought intensity index

1. Introduction

Common bean (*Phaseolus vulgaris* L.) is grown throughout the world as an important Legume crop and main dietary source of proteins, carbohydrate, dietary fiber, and minerals [1–5]. Moreover, amino-acids contained in common beans are of high dietary value [6] and help to reduce the risks of serious human diseases [7].

Drought and uneven rainfall are linked to climate change and such conditions pose a significant growth constraint for many field crops, including dry beans, especially, in the *semiarid* Mediterranean regions [8,9]. These conditions greatly influence production characteristics (yield, seed quality) which in turn impact the commercial value of the end product and the farmers' income. Among grain legumes, common beans are relatively sensitive to drought stress and these findings have been confirmed by experiments in the field, in greenhouses, and in controlled conditions [10]. More than 60% of the world's common bean is cultivated under non-irrigated conditions, and drought is estimated to cause up to 80% yield losses in many regions of the world [11]. The detrimental effects can affect common bean on several stages of development either during early growth, during the vegetative development or at flowering or pod/seed filling (flower and pod abortion and yield reduction) [12,13].

In Greece, common bean cultivation dates back to the 16th century AD [14]. In recent years bean is traditionally cultivated from April to September under different irrigation schemes to ensure acceptable production levels. The mean annual output of beans for the period 2012–2014 was 2.3 t ha⁻¹, while the mean acreage was 7870 ha [15]. Most bean fields are localized in the continental region and specifically in relatively cool and wet areas or around lakes. In these areas one of the main considerations is water availability for beans and other water-demanding spring crops like maize or cotton. Different bean genotypes have diverse responses to drought. Therefore, identifying drought tolerant genotypes would increase dry bean production in areas where irrigation water is a limiting factor [16].

Development of drought adapted common bean cultivars is an important strategy to minimize crop failure and increase food security in the face of climate change. Identification of key plant traits and mechanisms that contribute to improved drought adaptation can increase the efficiency of breeding programs through the selection of superior genotypes.

However, drought tolerance is a complex trait that cannot be investigated solely, as it is influenced by many factors including morphological and phenological characteristics. Moreover, selection under stress for yield can underestimate drought resistance, since resistance to extreme stress may be associated with yield penalty [17]. Most studies have focused on the effects of drought stress on agronomic or physiological variations with less effort on the combination of these factors [18,19]. The identification of key factors and patterns of combined variability in agronomic, morphological, phenological and physiological traits under drought can draw a picture on how these factors contribute to final grain yield in a consorted way [20]. Moreover, drought indices and yield means have been jointly used to identify genotypes exhibiting consistent performance across stress treatments. Such variables

include the percentage reduction in yield (YRR), the drought susceptibility index (DSI), harvest index (HI), and the geometric mean productivity (GMP) from contrasting environments [21].

The objectives of this study were to compare four dry bean cultivars on the agronomic, physicochemical and seed quality traits performance under water stressed conditions and irrigated conditions. The contribution of all these traits to seed yield and crop survival under water stress conditions would provide valuable information for determining trait evaluation in the breeding program.

2. Materials and methods

2.1. Experimental conditions

The experiment was carried out in 2014 at the main experimental field of the Industrial and Fodder Crops Institute (IFCI–‘ELGO ‘Demeter’; longitude 22°25’ E and latitude 39°36’ N) in Larissa prefecture in Central Greece and at an elevation of 73 m above sea level.

The soil of the experimental site was heavy-clayey (31% sand, 24% silt, 45% clay; Bouyoucos hydrometer method) [22], poor in organic matter content (1.3%, Walkley and Black method) [23] and classified as *Vertisol* [24]. The soil was sufficient in CaCO₃ (1.8%), low to medium in phosphorous (14 mg P kg⁻¹) [25] and high in exchangeable potassium content (Ammonium acetate method, 1.3 cmol K⁺ kg⁻¹) [26] to a depth of 30 cm, and a slightly alkaline (pH = 7.4; 1:1 soil–H₂O suspension) [27].

Genetic material included four dry bean cultivars (*Phaseolus vulgaris* L.) that corresponded to the dwarf, non-trailing type according to UPOV guidelines [28]. Two of them were cultivars ‘Iro’ and ‘Pyrgetos’ registered in the Greek National Variety Catalogue (selected and released by the IFCI) whereas, the other two commercial varieties were ‘Cannellino’ which is registered in the EU horticultural catalogue and the ‘Great Northern’ type.

‘Iro’ is a relatively early maturing cultivar (110 days to maturity) with medium-sized spheroid seeds (1000-seed weight 340–380 g, 1.2 cm in length and 0.8 cm in width, 5–6 seeds/pod) and average seed yield 2.5–3.0 t ha⁻¹ under irrigation. ‘Pyrgetos’ is earlier than ‘Iro’ (100 days to maturity) with a higher 1000-seed weight (380–400 g) and an average seed yield of 2.4–3.0 t ha⁻¹ under irrigation. Seed dimensions at maturity were 1.3 cm in length and 0.7 cm in width, while each pod contain 5–7 seeds. Both cultivars are well-adaptive under a wide range of soils. They have short cooking time (CT), extraordinary tasting characteristics and under favorable growing conditions can reach (‘Iro’) or even exceed (‘Pyrgetos’) 4.0 t ha⁻¹ seed yield.

The Italian cultivar ‘Cannellino’ known also as ‘Lingot’ is an early maturing cultivar (90–100 days to maturity) with medium-sized kidney-shaped seeds (1000-seed weight 280–340 g) and average seed yield 2.7–3.2 t ha⁻¹ under irrigation. The seeds at maturity reach 1.4 cm in length and around 0.63 cm in width, and contain 6–7 white seeds/pod. It is preferably harvested as dry bean even though can be also fresh consumed. Official data about the entry named ‘Great Northern’ are not available. ‘Great Northern’ corresponds to a commercial type of significant interest due to its popularity among Greek farmers.

Sowing was performed on the 9th of May 2014 (*late sowing*) so as the growth stages of flowering and pod filling coincide with the heat stress period (temperature > 35 °C and dry air condition). Emergence of seeds was observed 8–10 days after sowing.

Plot size was 4.5 m². Each plot consisted of four rows with 0.5 m distance apart. A dense seeding system was selected (90–100 kg ha⁻¹) on furrows and after seed germination (on 15 May

2014) plants were thinned to a distance of 5–7 cm on the row. Seeding depth was 2 to 3 cm.

Reduced tillage techniques (ploughing depth about 10 cm, motor rotary cultivation) were applied in order to ensure favorable seedbed conditions (minimize loss of soil moisture and eliminate weeds). At the time of sowing, a basic fertilization with a balanced complex mineral fertilizer (15 N–15 P₂O₅–15 K₂O; 200 kg ha⁻¹), was pre-plant incorporated, whereas no extra fertilization was added during the growing period. The trial was left unseeded (fallow) for the previous season. Hand-weeding was performed at regular intervals. Diseases and pests were controlled by conventional pesticides. Small scale aphid detection resulted to application of a non-systematic insecticide (deltamethrin 2.5 EC) on 13 June 2014 to prevent further aphid infection.

2.2. Irrigations and weather conditions

The trial received pre-plant irrigation (10 mm) to stimulate seed germination and thereafter 25 mm water through a sprinkler irrigation system to sustain further plant growth. Two levels of water treatment (WT) as a percentage of field capacity (by weight) were applied 25 days after seed emergence (first flower buds visible). The first water treatment was considered as normal-non stressed (NWT = 300 mm) in which the cultivars were irrigated until soil moisture level was depleted to 30% field capacity. The second water treatment was drought-stressed (SWT) in which the cultivars were irrigated with 150 mm of water (50% of the normal). Both water levels were applied through an automated drip irrigation system (spaced every 0.5 m; flow 5.1 m³ h⁻¹) with separate timers on a weekly basis during night windless hours until completion of pod filling. Data on mean, minimum and maximum air temperature and precipitation were acquired by a nearby (1 km) automatic weather station (<http://penteli.meteo.gr/stations/larissa/>) in Larissa (Figure 1).

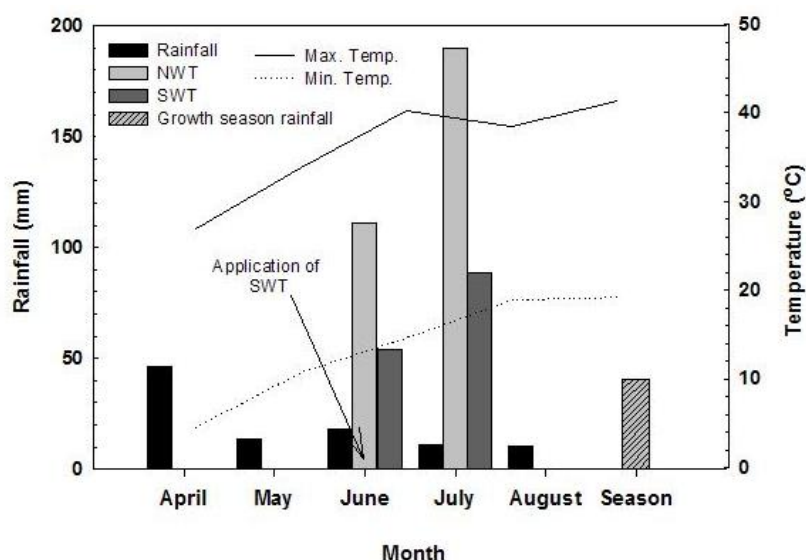


Figure 1. Minimum and maximum temperatures and rainfall distribution during the crop growing period at the Industrial and Forage Crops Institute in Larissa prefecture in Central Greece. On mid-June (36 days after sowing) two levels of water treatment (WT) were applied: normal = NWT and water stressed = SWT (indicated by the arrow on the graph). Growth season rainfall corresponds to amount from sowing till harvest.

2.3. Data sampling and calculations

2.3.1. Physio-morphological and agronomic traits

Physiological plant traits of the cultivars included: (i) Plant earliness (assessed non-distractively) as the days after sowing (DAS) to flower opening of at least 50% of plants per plot (days to flowering, DF); (ii) Leaf chlorophyll content (SPAD) using a hand held chlorophyll meter (SPAD-502, Minolta Co, Osaka, Japan). The leaf greenness was determined on the blades, midway between the leaf edge and midrib [29] of fully expanded terminal leaves. Measurements were recorded as the mean of 10 randomly selected plants per plot (5 plants per row of the two central rows per plot) at different stages of development full anthesis (35–40DAS), end of anthesis (55–60DAS), pod filling (70–75DAS); (iii) Plant height (PH) of 10 randomly chosen plants per plot (5 plants per row) at mid pod filling (70–75DAS).

Agronomic traits included yield components namely: (i) Number of pods per plant (PP) measured 70–80DAS from ten single plants of the two central rows in each plot; (ii) Number of seeds per pod (SP) from ten single plants of the two central rows in each plot; (iii) 100-seed weight (g) using a sensitive digital balance; (iv) Seed yield (SY) was calculated from the two central plant rows of each plot using sensitive digital balance and corrected based on seed moisture content. The plot yield was converted to kg per hectare (kg ha^{-1}) after adjusting to 13% moisture content. Seed yield was assessed at physiological maturity for each cultivar in order to avoid seed loss; (v) Harvest index (HI, dimensionless) as the ratio of SY per total above ground dry matter [30]. Plants first from the SWT and afterwards from the NWT were cut at ground level using hand sickles and threshed using a Wintersteiger plot combine.

Seed morphological characteristics included seed length (LEN), seed width (WID) and seed height (HEI) in mm measured on 20 seeds per plot using an electronic pachymeter.

2.3.2. Physicochemical and quality seed traits

The following physicochemical bean seed properties were assessed: (i) Hydration increase (HYI) calculated as the percentage of increase in mass of beans soaked in distilled water for 12 h; (ii) Hydration capacity (HC) expressed as hydration capacity per seed, determined by dividing the mass gained by the seeds in 12 h by the number of seeds present in the sample [31]; (iii) Seed coat proportion (SCP%) was determined on 10 seeds per plot, as the ratio in weight between coat and cotyledon expressed in percentage, after removing the seed coat from the cotyledons, both after soaking and keeping them for 24 h at 105 °C. Seed mass was assessed using a KERN precision and analytical balance.

Qualitative and nutritional seed traits included: (i) Cooking time (CT) which was recorded according to the method described by Iliadis [32]; (ii) Seed protein content (SPC %) determined in the finely grounded samples obtained from all plots for each cultivar using the Kjeldahl method (% protein = total N multiplied by 6.25); (iii) Seed pH in finely grounded samples of 5 g seeds per plot diluted in 20 mL distilled water and measured using a laboratory pH-meter (GLP 21-CRISON).

2.3.3. Drought tolerance indices

Additionally to direct parameter calculations drought tolerance indices for individual cultivars were also calculated using primary data according to Darkwa et al. [21]:

$$\text{Drought intensity index} = \text{DII} = 1 - X_{\text{DS}}/X_{\text{NS}} \quad [33] \quad (1)$$

$$\text{Drought susceptibility index} = \text{DSI} = \frac{1 - Y_{\text{DS}}/Y_{\text{NS}}}{1 - X_{\text{DS}}/X_{\text{NS}}} \quad (2)$$

$$\text{Drought tolerance index} = \text{DTI} = Y_{\text{DS}}Y_{\text{NS}}/X_{\text{NS}}^2 \quad (3)$$

$$\text{Mean productivity} = \text{MP} = (Y_{\text{DS}} + Y_{\text{NS}})/2 \quad (4)$$

$$\text{Geometric mean productivity} = \text{GMP} = \sqrt{Y_{\text{DS}}Y_{\text{NS}}} \quad (5)$$

$$\text{Yield reduction rate from non-stressed (\%)} = \text{YRR} = \frac{Y_{\text{NS}} - Y_{\text{DS}}}{Y_{\text{NS}}} \times 100 \quad (6)$$

$$\text{Yield stability index} = \text{YSI} = Y_{\text{DS}}/Y_{\text{NS}} \quad (7)$$

where, Y_{DS} and Y_{NS} are the mean yields for a particular bean cultivar under drought stress (SWT) and non- stress conditions (NWT) respectively, and X_{DS} and X_{NS} are the mean seed yields over all cultivars under drought stress and non- stress conditions respectively.

2.4. Statistical analyses and calculations

The treatments (level of irrigation and cultivars) were arranged in a split-plot design with three replications. Levels of water treatment-irrigation (WT) (NWT, SWT) were selected to be in the main plots and bean cultivars were selected to be in sub-plots. The results were analyzed by means of analysis of variance (ANOVA) using SPSS [34] and means were separated with Duncan's Multiple Range Test at $p < 0.05$. Pearson's correlation coefficients between all parameters were also determined whereas due to very low values data transformation was performed for harvest index (HI) [35].

3. Results

3.1. Weather conditions and plant growth

Data on monthly rainfall and minimum and maximum temperatures received during the growing season are compiled in Figure 1. Late June was extremely hot reaching peak temperatures close to 40 °C. A total of 40.4 mm precipitation was recorded during the growing season whereas around one fourth of it (~10.8 mm) was registered during the application of irrigation treatments (NWT and SWT) from 14th of June onwards.

On this date plant earliness, assessed non-destructively, showed that 'Cannellino' was the earliest cultivar (reached full flowering) followed by 'G. Northern', 'Pyrgetos' and 'Iro' respectively. Specifically, 'G. Northern' reached full flowering three days later (on 17th of June) than 'Cannellino' whereas 'Pyrgetos' and 'Iro' reached full flowering more than one week later (on 21st and 25th of June respectively).

Cultivars differed significantly in PH ($p < 0.001$), but the WT level did not influence PH. It was on average reduced by only 6.6 % under SWT conditions. ‘Cannellino’ was the tallest and ‘Iro’, the shortest among the cultivars under both WT levels. The former cultivar reduced its PH only by 2.6% whereas the later by 13.3% under SWT conditions (Table 1).

The chlorophyll content, assessed by SPAD readings, were not significantly different either between WT levels or among cultivars. Differences were more pronounced for ‘Cannellino’ especially at the late stages of crop growth (from 64 to 75 days after sowing) which reduced its leaf greenness by 16.4% under SWT conditions (data not shown).

3.2. Yield related traits

Cultivar (C), WT and their interaction $C \times WT$ effects on seed yield (SY) were found significant, whereas for the yield components (PPL, SP, HSW, and HI%) results were inconsistent.

SY was significantly reduced under SWT by on average 58.4% (Table 1) and cultivars responded significantly different to WT levels (Figure 2). ‘Cannellino’ showed the highest SY both under NWT (1065 Kg ha⁻¹) and SWT (654 Kg ha⁻¹), whereas the late maturing cultivar ‘Iro’ even though performed very well under NWT (808 Kg ha⁻¹), showed practically zero seed yield under SWT reaching not more than 10 Kg ha⁻¹. Responses to NWT of ‘G Northern’ were the lowest among the cultivars whereas, ‘Pyrgetos’ the least yielding cultivar under NWT (636 Kg ha⁻¹), showed 73.5% yield reduction under SWT (Table 1, Figure 2).

Among the yield components PPL and HI% were significantly affected by the cultivars, HSW was significantly affected by WT level and the $C \times WT$ was only significant for PPL. On average PPL reduced by 53.1% under SWT with ‘Iro’ showing the highest PPL reduction (95.8%) and ‘Cannellino’ the smallest one (7.9%) (Table 1). SP were not affected either from cultivars or WT level and reduced by 9.7% on average under SWT. ‘Cannellino’ again showed the smallest reduction (5.9%) and ‘Iro’ the largest one (14.1%). On average, HSW reduced by 18.3% under SWT with ‘Iro’ having the highest HSW (30 g) among cultivars under NWT whereas, under SWT ‘G. Northern’ exhibited significantly higher HSW. The cultivar ‘Cannellino’ had the lowest HSW under both WT levels. The ratio of seed weight over total plant dry weight (HI%) was affected only by the cultivars. However, values of HI% were rather low ranged on average from 0.21 to 0.11 under NWT and SWT respectively. ‘G Northern’ under NWT and ‘Cannellino’ under SWT had the highest HI%, whereas ‘Iro’ showed the largest HI% reduction (98.3%) under SWT (Table 1).

Table 1. Agronomic data (SY = Seed yield in kg ha⁻¹; yield components: PPL = pods plant⁻¹, SP = seeds pod⁻¹, HSW = 100 seed weight; HI = harvest index; PH = plant height) and morphological (LEN = seed length, WID = seed width, HEI = seed height), physico-chemical (HYI = hydration increase, HC = hydration capacity, SCP = seed coat proportion) and quality (CT = cooking time, SPC = seed protein content and seed pH) seed traits of four bean cultivars (*Phaseolus vulgaris* L.) as affected by two levels (NWT = normal, SWT = stressed) of water treatment (WT). Data represent means of three replications.

| Cultivar | SY | PPL | SP | HSW(g) | HI(%) | PH(cm) | LEN | WID | HEI | HYI(%) | HC | SCP(%) | CT(min) | SPC(%) | Seed(pH) |
|----------------------------|-------------------|------------------|------|------------------|--------------------|-------------------|--------------------|------------------|------------------|--------------------|--------------------|-------------------|-----------------|-------------------|--------------------|
| | | | | | | | 10 ⁻³ m | | | | | | | | |
| <i>Non stressed WT</i> | | | | | | | | | | | | | | | |
| Pyrgetos | 636 ^c | 8.8 | 2.1 | 29 ^{ab} | 0.13 ^c | 46.9 ^b | 12.8 ^b | 7.1 ^b | 5.1 ^b | 119.2 ^b | 0.43 ^a | 7.7 ^c | 15 ^c | 29.2 ^a | 6.45 ^a |
| Iro | 808 ^b | 8.6 | 2.4 | 30 ^a | 0.19 ^{bc} | 42.4 ^c | 12.1 ^c | 7.7 ^a | 4.9 ^b | 113.7 ^c | 0.42 ^{ab} | 9.5 ^b | 17 ^b | 24.8 ^b | 6.35 ^b |
| G. Northern | 767 ^b | 8.1 | 2.2 | 26 ^{bc} | 0.30 ^a | 58.1 ^a | 12.4 ^{bc} | 7.0 ^b | 6.0 ^a | 126.0 ^a | 0.40 ^{ab} | 8.0 ^c | 15 ^c | 24.6 ^b | 6.41 ^a |
| Cannellino | 1065 ^a | 9.3 | 2.3 | 24 ^c | 0.24 ^{ab} | 58.5 ^a | 13.4 ^a | 6.3 ^c | 5.0 ^b | 124.3 ^a | 0.37 ^c | 10.3 ^a | 22 ^a | 24.2 ^b | 6.44 ^a |
| <i>Mean</i> | 819 | 8.7 | 2.2 | 27 | 0.21 | 51.5 | 12.7 | 7.0 | 5.2 | 120.8 | 0.41 | 8.9 | 17 | 25.7 | 6.41 |
| <i>Stressed WT</i> | | | | | | | | | | | | | | | |
| Pyrgetos | 168 ^b | 1.8 ^c | 1.9 | 23 ^{ab} | 0.06 ^{bc} | 44.4 ^c | 11.5 ^b | 6.6 ^c | 4.9 ^b | 129.1 ^a | 0.30 ^a | 9.3 ^c | 18 ^c | 31.2 ^a | 6.43 ^a |
| Iro | 8 ^c | 0.4 ^c | 2.0 | 21 ^{bc} | 0.003 ^c | 36.8 ^d | 10.8 ^c | 6.8 ^b | 4.8 ^b | 112.5 ^c | 0.23 ^b | 12.3 ^a | 20 ^b | 30.3 ^a | 6.35 ^{bc} |
| G. Northern | 531 ^a | 5.6 ^b | 2.0 | 25 ^a | 0.14 ^b | 54.1 ^b | 12.1 ^a | 7.0 ^a | 5.6 ^a | 121.2 ^b | 0.35 ^a | 8.5 ^d | 18 ^c | 26.7 ^b | 6.31 ^c |
| Cannellino | 654 ^a | 8.6 ^a | 2.1 | 19 ^c | 0.24 ^a | 57.0 ^a | 12.6 ^a | 6.0 ^d | 4.8 ^b | 123.5 ^b | 0.35 ^a | 11.1 ^b | 30 ^a | 26.9 ^b | 6.37 ^b |
| <i>Mean</i> | 340 | 4.1 | 2.0 | 22 | 0.11 | 48.1 | 11.7 | 6.6 | 5.0 | 121.6 | 0.31 | 10.3 | 22 | 28.8 | 6.37 |
| CV% | 25.4 | 28.9 | 13.8 | 13.1 | 33.9 | 3.59 | 4.60 | 3.29 | 5.98 | 3.69 | 14.05 | 5.66 | 10.1 | 5.66 | 0.80 |
| LSD _{0.05} | 13.1 | 1.64 | 0.26 | 28.7 | 0.89 | 1.59 | 0.50 | 0.20 | 0.27 | 3.98 | 0.05 | 0.48 | 3.48 | 1.37 | 0.05 |
| <i>F-test significance</i> | | | | | | | | | | | | | | | |
| Cultivar (C) | *** | ** | n.s. | n.s. | * | *** | ** | *** | *** | ** | n.s. | *** | *** | *** | * |
| WT | * | n.s. | n.s. | * | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. | ** | * | ** | ** | ** |
| C×WT | * | * | n.s. | n.s. | n.s. | n.s. | n.s. | * | n.s. | n.s. | n.s. | * | n.s. | n.s. | n.s. |

*, **, ***: Significant differences at 0.05, 0.01 and 0.001 respectively and n.s. is not significant. Means followed by different letters within the same column and water treatment (WT) are different at $p < 0.05$ according to Dunncan's Multiple Range Test (DMRT). LSD_{0.05}: Least significant difference for comparing the cultivars within the WT at 5% probability level.

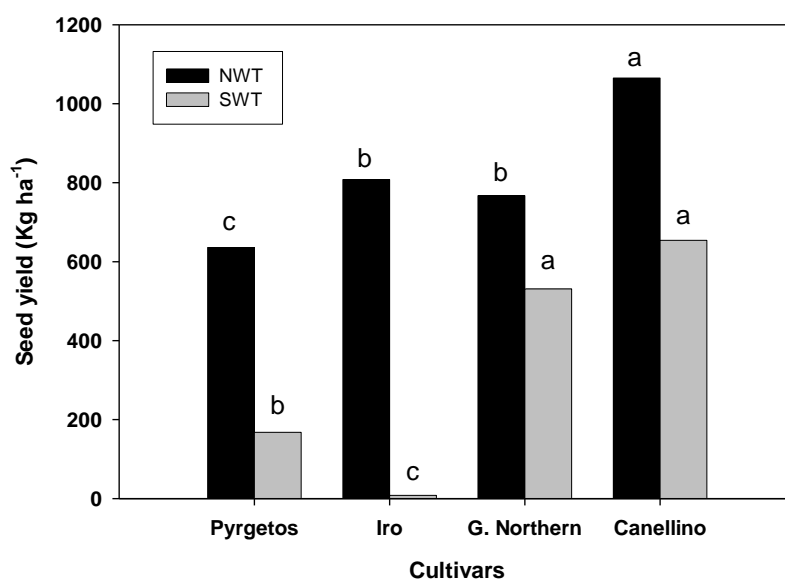


Figure 2. Seed yield of four bean cultivars (*Phaseolus vulgaris* L.) as affected by two levels of water treatment (NWT = normal, SWT = stressed). Mean seed yield followed by different letters within water treatment (WT) are different at $p < 0.05$ according to Dunncan's Multiple Range Test.

3.3. Morphological, physicochemical and quality seed traits

Except hydration capacity (HC), cultivars affected all the rest morphological (LEN, WID, HEI), physicochemical (HYI and SCP%) and quality seed traits (CT, SPC%, pH). The effect of WT was significant on all qualitative seed traits (CT, SPC %, pH) and on HC and SCP% whereas significant $C \times WT$ were seldom only for WID and SCP% (Table 1). Among cultivars 'Cannellino' exhibited the highest LEN under both WT levels, and 'Iro' reduced its LEN by 10.5% under SWT. The later cultivar had the greatest reduction for WID whereas, 'G Northern' showed none WID reduction under SWT. The highest HEI was found for the cultivar 'G Northern' under both WT levels. As far as seed physicochemical traits are concerned, 'G Northern' even though under NWT showed among cultivars the highest HYI, under SWT exhibited the largest HYI reduction (3.8%). HC was mostly reduced (45.6%) for 'Iro' followed by 'Pyrgetos' (30%) under SWT. SCP was greater (10.3%) for 'Cannellino' under NWT and for 'Iro' (12.3%) under SWT. Over all cultivars SCP increased by 16.0% under SWT. 'Iro' increased more its SCP (29.5%) followed by 'Pyrgetos' (21.7%) under SWT (Table 1). CT was on average increased by 29.4% under SWT. Among cultivars 'Cannellino' showed the highest CTs under both WTs and the higher CT increase under SWT (36.5%). Seed protein content (SPC%) increased for all cultivars under SWT. 'Iro' showed the greater SPC increase (22.2%) and 'Pyrgetos' the lowest one since exhibited the highest SPC% under both WT. Seed pH was on average reduced only by 0.7% under SWT; the higher among cultivars reduction was found for 'G Northern' (1.5%).

3.4. Drought indices

Drought intensity index (DII) over all cultivars was found moderate (0.58). Drought tolerance/susceptibility indices were calculated for each cultivar (Table 2). With regard to indices related to mean productivity (MP) and geometric mean productivity (GMP) under the two WT levels ‘Cannellino’ was the higher-yielding cultivar followed by ‘G Northern’. These two cultivars were considered tolerant to drought stress because exhibited the lower DSI and YRR and the higher values of DTI and YSI. However, ‘G Northern’ was not after ‘Cannellino’ the higher yielding cultivar under NWT; this was the case for ‘Iro’ which was considered the most susceptible cultivar to drought stress since showed minimal DTI and YSI even though yielded very well under NWT.

Table 2. Yield related drought tolerance indices of four bean cultivars (*Phaseolus vulgaris* L.) grown under normal and water stress conditions in Larissa, Central Greece.

| Cultivar | Drought tolerance indices ¹ | | | | | | DII |
|-------------|--|------|-------|-------|------|------|------|
| | DSI | DTI | MP | GMP | YRR | YSI | |
| Pyrgetos | 1.26 | 0.16 | 402.0 | 326.9 | 73.6 | 0.26 | |
| Iro | 1.69 | 0.01 | 408.0 | 80.4 | 99.0 | 0.01 | 0.58 |
| G. Northern | 0.53 | 0.61 | 649.0 | 638.2 | 30.8 | 0.69 | |
| Cannellino | 0.66 | 1.04 | 860.5 | 834.6 | 38.6 | 0.61 | |

¹where: DSI=Drought susceptibility index, DTI=Drought tolerance index, MP=Mean productivity, GMP=Geometric mean productivity, YRR=Yield reduction rate from non-stressed (%), YSI=Yield stability index, DII=Drought intensity index (over all cultivars).

3.5. Correlation between traits

As found in Table 3, SY was significantly and positively correlated to most yield components (PPL, SP and HI) except to HSW. Strongest was found the correlation between SY and PPL ($r = 0.888, p < 0.01$) as compared to SY and SP ($r = 0.622, p < 0.01$) (Figure 3). Relations between PH and various yield components were also positive and significant except for SP. SY was also positively correlated to LEN and HC and as expected negatively to SPC ($r = -0.828, p < 0.01$). PPL was correlated to other yield components (SP, HI, HSW), to LEN, HC, and negatively correlated to SCP and SPC. HI was positively correlated to LEN and to HC. PH was positively correlated to seed morphological traits (LEN and HEI) and to HYI and negatively to SPC. Except the negative correlations between SPC and various yield components negative correlations were also observed between this trait and LEN ($r = -0.573, p < 0.01$) and SCP ($r = -0.486, p < 0.05$). The correlations between CT and HSW, WID, HEI, SCP were also negative. No significant correlations were observed between seed pH and any other examined trait.

Table 3. Pearson's correlation coefficients and levels of significance between agronomic traits and several morphological, physicochemical and quality seed traits under normal and water stress conditions. Data were combined over cultivars, water treatments and replications (n = 24).

| | SY | PPL | SP | HI | PH | HSW | LEN | WID | HEI | HYI | HC | SCP | SPC | CT |
|-----|----------|----------|----------|----------|----------|----------|----------|----------|----------|------|----------|---------|------|------|
| PPL | 0.888** | | | | | | | | | | | | | |
| SP | 0.622** | 0.624** | | | | | | | | | | | | |
| HI | 0.784** | 0.752** | 0.715** | | | | | | | | | | | |
| PH | 0.661** | 0.594** | n.s. | 0.644** | | | | | | | | | | |
| HSW | n.s. | 0.439* | n.s. | n.s. | n.s. | | | | | | | | | |
| LEN | 0.756** | 0.739** | n.s. | 0.662** | 0.703** | n.s. | | | | | | | | |
| WID | n.s. | n.s. | n.s. | n.s. | n.s. | 0.684** | n.s. | | | | | | | |
| HEI | n.s. | n.s. | n.s. | n.s. | 0.464* | n.s. | n.s. | n.s. | | | | | | |
| HYI | n.s. | n.s. | -0.414* | n.s. | 0.448* | n.s. | n.s. | -0.508* | n.s. | | | | | |
| HC | 0.612** | 0.663** | n.s. | 0.423* | n.s. | 0.593** | 0.652** | n.s. | n.s. | n.s. | | | | |
| SCP | n.s. | -0.422* | n.s. | n.s. | n.s. | -0.611** | n.s. | -0.469* | -0.620** | n.s. | -0.699** | | | |
| SPC | -0.828** | -0.737** | -0.552** | -0.711** | -0.584** | n.s. | -0.573** | n.s. | n.s. | n.s. | -0.486* | n.s. | | |
| CT | n.s. | n.s. | n.s. | n.s. | n.s. | -0.553** | n.s. | -0.676** | -0.450* | n.s. | n.s. | 0.628** | n.s. | |
| pH | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. |

For explanation of abbreviations please refer to Table 1 and to abbreviation list. *,** significant correlations at $p \leq 0.05$ and $p \leq 0.01$ respectively, n.s. = not significant.

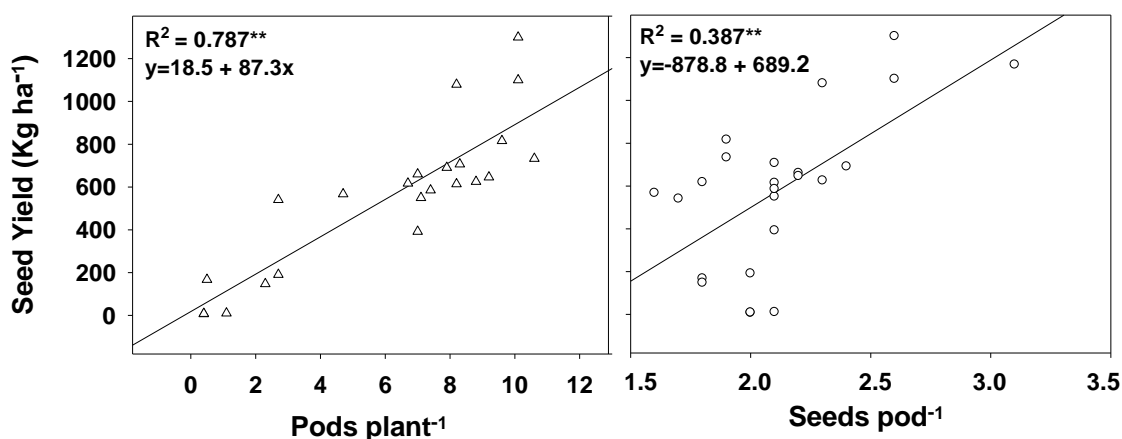


Figure 3. Relationship between bean seed yield with pods plant⁻¹ (left) and seed yield with seeds pod⁻¹ (right). Regression is indicated by the solid line. ** p value of the fitted model is significant at $p < 0.01$.

4. Discussion

The uncharacteristically warm/hot, growing conditions (~ 40 °C, from 25–27 of June, Figure 1) especially during crucial stages of development (flowering) imposed water stress and resulted in pronounced cultivar yield differences. These temperatures are considered unfavorable for common bean production [13,36]. The detrimental combination of high temperatures and water shortage (SWT) was especially evident for the cultivar ‘Iro’ which showed flower drop and absence of pod filling. The yield of ‘Iro’ was negligible (8 kg ha^{-1}), whereas ‘Pyrgetos’ (168 kg ha^{-1}) and the early cultivars ‘G. Northern’ and ‘Cannellino’ reached acceptable yields under SWT (Table 1, Figure 2).

Plant height (PH) was significantly different for the cultivars and was reduced on average by 6.6% under SWT. Cultivar differences in plant height may be also attributed to differences in earliness. Specifically, the early cultivars ‘G. Northern’ and ‘Cannellino’ had already completed their development when SWT took place. However, this was not the case for ‘Iro’, the later among the cultivars, which reduced its PH by 13.3% under water stress. These findings are similar to those observed by other studies [37–39]. Leaf chlorophyll content (SPAD) measured from flowering to maturity was lower on average under SWT compared to NWT (data not shown) confirming the findings by Ghanbari et al. [40]. Chaves et al. [41] also reported that drought stress reduces leaf chlorophyll content. The cultivars ‘G. Northern’ and especially ‘Cannellino’ reduced their leaf greenness more under SWT later in the season. According to Mafakheri et al. [42] chlorophyll decrease under SWT is attributed to chloroplast breakdown.

The magnitude of stress over all cultivars measured by the drought intensity index (DII) was found moderate (0.58, Table 2). These conditions are considered sufficient to express genotypic differences for the measured traits. Darkwa et al. [21] in their study were able to discriminate a set of large number (67) of common bean genotypes under moderate drought stress conditions ($\text{DII} = 0.30$). Normally DII values exceeding 0.70 indicate severe drought conditions. Several other studies indicate that under severe drought stress ($\text{DII} > 0.70$) genotypic differences could be eliminated because of the extremely low yield potential [43,44] whereas, evaluation of common bean under high to moderate stress is considered ideal [45,36].

The significant effect of the C, WT treatments and $C \times \text{WT}$ interaction for seed yield indicated

that common bean cultivars responded differently across the two WT regimes. These results are in compliance to those of other authors [45–47] who reported differential response of common bean varieties to drought-stressed and non-stressed conditions. The results of our study showed that seed yield reduced by more than a half (58.4%) as compared to NWT (Table 1). Similarly, Singh [48] recorded mean yield reductions by 52% for dry bean landraces under water stress conditions, whereas Urrea et al. [49] in their study found yield reductions from 47% to 69%. Cultivars responded differently to SWT reducing their SY potential from 30.8% to 99.0% (Table 1, Figure 2). The superiority of ‘Cannellino’ for SY under both stress and non-stress conditions could be attributed to plant earliness (reached full flowering when WT were applied) and to its high HI. These traits contribute significantly to elevated yield potential especially under water stress conditions [50–52] coupled with high total biomass and number of pods per plant [53,54].

Yield components are good indicators of overall water stress and the present study revealed that significant yield reductions are attributed to reductions to individual yield components under SWT. Similarly Asfaw and Blair [45] reported reductions in PPL, SP, HSW and consequently seed yield of common bean under drought-stressed conditions. These reductions were attributed to unsuccessful flower fertilization which is an important consideration for determining seed yield under drought-stress conditions [43]. In our experiments PPL were unaffected by the WTs however, cultivar differences were pronounced. This may indicate that this trait is more genotype dependent. SP was not affected either by cultivars or WT and HSW was only affected by WT.

SY was significantly correlated PPL and to SP as was also found in the study by Abebe and Brick [55], with the former correlation being stronger (Table 3, Figure 3) indicating that PPL could be a reliable indirect selection criterion for high yielding bean genotypes. Accordingly, Nielsen and Nelson [39] found that PPL was closer correlated to SY than SP under water stress conditions.

Harvest Index, that is used to express the proportional remobilization of photosynthates to grain [56] was found different ($p < 0.05$) between the cultivars and on average very low (0.11) especially under SWT. Intense water stress results to low harvest indices as indicated by Munoz-Perea et al. [51] even though our study was conducted under moderate drought stress (DII = 0.58, Table 2). According to Jain [57] legumes produce high biomass in the expense of seeds leading to low harvest indices. Similar results are presented by Vlachostergios and Roupakias [58] for lentil. Moreover HI was strongly correlated to SY (Table 3) and as indicated by Solanki [59] this could be further studied and proposed as an indirect selection criterion for high yield potential.

All cultivars exhibited lower HSW under SWT (on average reduced by 18.3%) confirming similar results by Castaneda-Saucedo et al. [60] who reported HSW reduction from 10.0% to 14.8% depending on the developmental stage of water stress application.

Seed morphological traits (LEN, WID, HEI) were affected to a lesser extent by the WT levels and cultivars demonstrated significant differences for all traits. Some genotypic effects were modified by the WT levels only for seed width (WID). Even though ‘Cannellino’ exhibited the highest seed length, it had the lowest WID under both WT levels, whereas ‘G Northern’ showed no WID reduction under SWT. From the Greek cultivars, ‘Iro’ showed the lowest LEN under both WT and ‘Pyrgetos’ had intermediate seed size. It seems that seed size is mainly genetically driven even though some morphological traits could be affected by water shortage. On the other hand consumers are attracted by the large or medium-large bean cultivars even though small-seeded genotypes normally exhibit higher yield potential [61].

Except seed hydration capacity (HC) cultivars significantly affected all physicochemical (HYI, SCP) and qualitative seed traits (CT, SPC, pH). Accordingly, these traits, with the exception of HYI, were

affected by water stress (SWT) and either on average increased (SCP, CT, SPC) or decreased (HC, pH). The Greek cultivar 'Iro' exhibited the largest reduction in HC under water stress conditions (45.7%). Moreover, HC was significantly and negatively associated to SCP which in turn is positively related to cooking time (CT) (Table 3). It is evident that bean cultivars with high HC or HYI require less time for boiling. This is a major quality trait for market consumption. Rapid water uptake in seeds is also an important factor to the processing industry. Bean cultivars with exceptional physicochemical characteristics influence the acceptance both by consumers and by farmers.

Cooking time is a seed quality trait that is related to energy consumption. Storage of bean seeds under elevated temperature and high relative humidity conditions increase cooking time. The imbibition of water by bean seeds during soaking leads to the softening of the seed coat and cotyledon because of cellular expansion [62]. Agbo et al. [63] postulated that cooking time increased because of hard bean seed coat. Bean cultivar differences in CT may be attributed to different micropyle size which is related to seed coat water penetration. Seed perisperm leaves passage to water and/or oxygen entrance within the seed resulting in a state of forced dormancy. However, the only entry of these elements is the micropyle. Other studies indicate that, CT as well as seed coat hardness is increased by high content of secondary macronutrients like Ca and Mg whereas, these characteristics decrease in soils poor in P and Mg [64]. Stoyanova et al. [65] found that bean seeds produced on a *calcic chernozem* in Bulgaria needed a longer cooking time compared to those produced in soils with lower Ca levels.

Seed protein content (SPC%) increased under water stress conditions by on average 12.0%. 'Iro' responded better exhibiting 22.2% SPC increase under water stress conditions, but 'Iro' had the lowest yield under water stress. The Greek cultivars showed the higher SPC; 'Pyrgetos' under NWT and 'Pyrgetos' and 'Iro' under SWT had the higher SPC. The higher SPC for the late cultivar 'Pyrgetos' is also supported by the higher chlorophyll meter readings recorded for this cultivar at the end of the growing season (data not shown) indicating that available photosynthates are mobilized to the seed and translated into proteins at maturity. There was a negative relation between SY and SPC (Table 3) confirming results of other studies on the well known inverse yield-protein relation either on cereals [66] or in legumes [67]. In common bean, phenotypic relations between these two traits (GY vs SPC) vary greatly and these relations are neutral or negative [68,69]. It was suggested that improving simultaneously both traits is feasible [70]. One way is to select for high seed yield potential and then selecting within the high yielding families single plants with high seed protein concentration [71]. The higher SPC under SWT could also be attributed to the increase of *de novo* proteins due to water limitation [72]. Our results agree with those of Sadeghipour and Aghaei [73] but conflicting to those of Ghanbari [74].

5. Conclusion

Significant differences were detected between WT levels for SY (58.4%) and PPL (53.1%). A positive correlation between SY and PPL was found, indicating that PPL could be a reliable selection criterion for high yielding bean genotypes evaluated under water and heat stress. Earliness was a crucial factor which determined yield potential. Among cultivars the early 'Cannellino' and 'G. Northern' exploited the available soil moisture efficiently and reached stable yields. Such cultivars may be a good source genetic material for breeding programs and could be suggested for cultivation in drought prone areas where irrigation water is scarce. Valuable genetic variability was observed for CT and SPC both important quality criteria attracting consumers, farmers and the industry. Among cultivars, the Greek cultivar 'Pyrgetos' was superior for these seed quality traits under both WT levels.

Conflict of interest

The authors declare no conflict of interest in this paper.

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