Effects of nutritive solution electrical conductivity and plant density on growth, yield and quality of sweet basil grown in gullies by subirrigation

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Key words: leaves quality, nutrient uptake, Ocimum basilicum L., plants number per pot, production, soilless.

Abstract: The increasing demand for basil in the last decade has arisen from consumer tendency towards high nourishing produce. Soilless growing of this crop is a current farm strategy and the quality targets are affected by nutritive solution as well as by plants density per pot. Research was carried out with the aim of assessing plant growth, yield and leaves quality of basil (*Ocimum basilicum* L., cv. Gecom FT) grown in pots (peat-lapil) and fed by subirrigation inside plastic gulles, under a heated greenhouse. Comparisons were made of four electrical conductivities (EC: 2.2, 2.5, 2.8, 3.1 mS·cm⁻¹) in factorial combination with four plant densities (9, 12, 15, 18 plants per pot) and a split plot design was arranged with three replicates. The 2.8 mS·cm⁻¹ EC resulted in the best yield, growth indexes and biometrical parameters values. Water absorption was highest under the 2.8 mS·cm⁻¹ EC, whereas the highest nutrient consumptions as well as the best quality indicators and chemical composition corresponded to the 2.8 to 3.1 mS·cm⁻¹ EC range. The 12 plants per pot density gave the best results, in terms of yield, growth indexes and biometrical parameters, also showing the highest plant water and nutrient uptakes. The leaves quality attributes and chemical composition always displayed decreasing trends as a function of the plant density increase, the highest values corresponding to 9 and 12 plants per pot; only the nitrates concentration showed an opposite trend compared to the other nutrients. In conclusion, the 2.8 mS·cm⁻¹ nutritive solution and the 12 plants per pot density resulted in the best yield and leaves quality. Further enhancement of both experimental factors level even caused the reduction of water and nutrient efficiency use.

1. Introduction

The increasing demand for basil resulted in cropping area extension by 66% since 2001 in Italy (www.istat.it), with the frequent use of local ecotypes (Zecchinelli, 1999; Tesi and Lenzi, 2002). Basil (*Ocimum basilicum* L.) is a high marketable value vegetable, which is consumed both as a fresh aromatic ingredient and combined with pasta in a cooked dish (pesto). As today's consumer choices are oriented towards high quality produce, not only from a sensorial point of view but also in terms of nutritional properties, a particular emphasis is given to this product. In this direction, soilless growing could represent an effective crop management in order to

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enhance product quality attributes (Sgherri et al., 2010), which mainly depend on variety (Tesi et al., 1991) but they are also affected by crop system (Tesi et al., 1997). Plant growing in pots sown with several seeds per pot, to be sold when the plants set reaches a scheduled size, is one of the current farm strategies and interesting market perspectives mainly arise from the winter crop cycle. In this season, light intensity is sufficient for carrying out efficient crop cycles (Beaman et al., 2009), but it is necessary ensuring the adequate minimum temperature, which is also positively correlated with basil flavour (Chang et al., 2007). Moreover, the nutritive solution supplied to plants plays a crucial role as it significantly affects yield (Bekhradi et al., 2015) and plant features, such as stem height and dry weight (Adler et al., 1989; Bione et al., 2014); in this respect, basil is considered a moderately tolerant species (Herrera, 2005). The plants density is also of primary importance for the

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produce quality (Bohme and Pinker, 2014), as the individual space available has a major impact on the balance among the different plant parts.

Due to literature shortage on the above mentioned topics, research was carried out on soilless pot-grown basil in Naples (southern Italy), with the aim of evaluating the effects of nutritive solution electrical conductivity and pot plants density on crop growth, yield and leaves quality.

2. Materials and Methods

Research was carried out on basil (Ocimum basilicum L. cv. Gecom FT) at the experimental site of Naples University Federico II in Portici (Naples, southern Italy, 40°49' N, 14°20' E, 63 m a.s.l.), in a Mediterranean or Csa climate (Peel et al., 2007), during winter season in 2007 and 2008. The crops were soilless grown in pots, placed in gullies and fed by sub-irrigation, under plastic (IR-PE) tunnels equipped with an air heating system set to 16°C. Comparisons were made of four electrical conductivities (EC: 2.2, 2.5, 2.8, 3.1 mS·cm⁻¹) in factorial combination with four plant densities (9, 12, 15, 18 plants per pot) and a split plot design was arranged with three replications; each treatment included 30 pots. The hydroponic equipment consisted of: a) 48 rigid PVC gullies (each 12 cm wide and deep, 300 cm long) supported by plastic elements at 70 cm above ground level, according to 1% slope; b) 12 plastic tanks holding 220 I; c) 12 submerged pumps of 90 watt unit power; d) 24 delivery and return overhead lines. The sowing was performed on 15 January in pots (\emptyset = 10 cm) filled with peat and lapil (1:1 in volume), placed in the gullies through a pierced white PE film and spaced 10 cm along and between the rows (18 pots per m²). They were fed by nutritive solutions (Table 1), with 1-6 interventions per day of 5 minutes each, which were never adjusted but they were completely changed at the end of each weekly cycle. Besides, during the crop, an insecticide application against aphids was practiced.

Table 1 - Chemical composition of the soilless nutrient solutions

The crop cycles ended when each plants set reached the scheduled size for pot commercialization, i.e. the plants had four fully expanded leaves couples, and at that time the following determinations were made on plant samples obtained by 8 pots per plot: fresh and dry weight of whole plants and of leaves; leaf area; stems thickness and height. Moreover, water consumption was calculated assessing the volume of nutritive solution in each tank at the beginning and at the end of each weekly cycle. Concurrently, nutritive solution samples were collected in order to assess nutrient consumption through laboratory analyses of nitrogen, phosphorus, potassium, calcium, magnesium and iron, using the same methods as described below for leaf cation and anion determinations.

At the crop cycles end, leaves samples were also collected from 8 pots per plot, in order to perform laboratory analyses. In this respect, two hundred grams of leaves per plot were homogenized in a 1.0 L Waring blender (Waring Laboratory, Torrington, CT, USA) and aliquots of this raw homogenate were used for the analyses of cations. The raw homogenate was centrifuged at 10,000 x g for 30 min at 6°C in an 5810R Eppendorf refrigerated centrifuge (Eppendorf HQ, Hamburg, Germany). The resulting supernatant was passed through a 0.45 µm Acrodisc filter (Gelman Sciences, MI, USA). Samples of this filtered leaves extract were used for assessing anion, sugar and ascorbic acid contents.

The laboratory determinations were performed as follows:

- the dry residue was assessed in an oven at 70°C with a vacuum;
- the soluble solids content or SSC (in °Brix) was measured at 20°C with a Bellingham & Stanley, model RFM 81 digital refractometer on the supernatant obtained from raw homogenate centrifugation;
- anions, sugars and ascorbic acid were determined by high performance liquid chromatography (HPLC) as previously described (Caruso et al., 2011);

Nutritive solution EC (mS·cm ⁻¹)	macronutrients (mmol·L ⁻¹)						micronutrients (m mol·L ⁻¹)						
	Ν	Р	К	Ca	Mg	S	Cl	Fe	Cu	Mn	Zn	В	Mo
2.2	12.3	1.6	5.3	3.8	2.2	1.9	1	35	1	15	5	35	1
2.5	13.8	1.7	5.9	4.3	2.5	2.5	1	35	1	15	5	35	1
2.8	15.7	2.0	6.7	4.8	2.8	2.8	1	35	1	15	5	35	1
3.1	17.5	2.2	7.6	5.4	3.0	3.3	1	35	1	15	5	35	1

For all treatments pH was adjusted to 6.0 and NH₄/NO₃ ratio was 1/9.

- titratable acidity of the leaves homogenate was determined as previously described (Caruso *et al.*, 2014) and it was expressed as grams of anhydrous citric acid per 100 g of leaf fresh weight;
- cations (Ca, Mg, K) content in the leaves homogenate was determined by atomic adsorption spectrophotometry as previously described (Caruso *et al.*, 2011).

Data statistical processing was performed by analysis of variance using the SPSS software version 21, referring to 0.05 probability level, and Duncan multiple test was used for mean separation.

3. Results and Discussion

From the data statistical processing, the year of research resulted to have no significant effect either as a main effect or as an interaction with the two experimental factors, therefore in the following tables the mean values of the experimental data of the years 2007 and 2008 are reported.

As for yield results relevant to the comparison among the nutritive solution electrical conductivities (Table 2), the 2.8 mS·cm⁻¹ EC resulted in the highest basil yield, both as whole plants and of leaves, and in the shortest crop cycle; the lowest nutrient solution strength (2.2 mS·cm⁻¹) showed the worst performances.

Table 2 - Basil yield results

		Yield	Crear avala	
Nutritive solution EC (mS·cm ⁻¹)	Whole plants (g·m⁻²)	Leaves (g·m⁻²)	Leaves/ Plant (%)	duration (days)
2.2	579.1 d	418.7 d	72.3 c	64.0 a
2.5	733.9 c	538.7 c	73.4 b	62.7 b
2.8	958.9 a	713.4 a	74.4 a	61.3 c
3.1	845.5 b	629.9 b	74.5 a	61.0 c
No. plants per pot				
9	761.9 b	570.7 b	74.9 a	61.2 c
12	931.1 a	695.5 a	74.7 a	61.4 c
15	776.3 b	569.8 b	73.4 a	62.6 b
18	648.5 c	465.6 c	71.8 c	63.9 a

Among the plant densities, the 12 plants per pot treatment resulted in the highest yields and both the 9 and 12 plants per pot led to the shortest crop cycle (Table 2).

In terms of growth indexes and biometrical parameters (Table 3), the 2.8 mS·cm⁻¹ EC also produced the highest values of plant dry matter, leaf area and stem thickness, though the latter was not statistically different from that obtained with the highest EC level

Table 3 - Basil growth and biometrical parameters

Nutritive solution EC (mS·cm ⁻¹)	Plant dry matter (g·m ⁻²)	LAI (m²·m⁻²)	Leaf area (cm²·pt⁻¹)	Plant height (cm)	Stem thickness (mm)
2.2	45.0 d	0.66 c	28.5 c	17.2 a	2.76 b
2.5	64.8 c	0.92 b	39.3 b	17.0 a	2.90 b
2.8	90.0 a	1.11 a	47.1 a	16.7 b	3.11 a
3.1	82.8 b	0.98 b	41.6 b	16.6 b	3.16 a
No. plants per pot					
9	73.6 b	0.86 c	51.9 a	16.8 b	3.33 a
12	88.2 a	1.06 a	47.6 b	16.9 ab	3.12 b
15	67.9 b	0.93 b	33.1 c	16.9 ab	2.85 c
18	52.7 c	0.83 c	24.3 d	17.1 a	2.60 d

(3.1 mS·cm⁻¹); conversely, nutritive solution dilution caused the internodes extension and, indeed, the two lowest electrical conductivities enhanced plant height.

With regard to plant density (Table 3), the 12 plants per pot treatment resulted in the highest dry weight and LAI, whereas the lowest density produced the thickest stems; the 15 and 18 plants per pot densities proved excessive and, in particular, the highest one caused the most unbalanced growth of plants. In fact, the plants grown under the 18 plants per pot treatment showed thinner stems and smaller leaves compared to the other experimental treatments, and they were also taller than the most spaced ones.

In contrast with our findings, in previous research (Tesi et al., 1995) the 1.6 mS·cm⁻¹ EC showed the best effect on basil yield. Moreover, other authors reported that doubling the nutrient availability did not affect basil yield (Raimondi et al., 2006), but it led to the increase of leaf dry matter percentage and LAI (Chen et al., 2004). In our research, the depressing effects of salt stress caused by the 3.1 mS cm⁻¹ EC on plant vegetative growth and in particular on leaf area corresponds to the rapid plant adaptation to water deficit (Munns, 2002). Moreover, in our investigation, the density increase presumably caused the light conditions worsening within the canopy and, accordingly, the reduction of plant photosynthetic efficiency. Chang et al. (2007) also reported that basil plant weight is adversely correlated with canopy shading. Consistently, Tesi et al. (1995) recorded the plant weight increase per soil unit area up to a critical density value, over which a decrease occurred; however, they also found that a doubled density, compared to our best treatment of 12 plants per pot provided with the best results using 10 cm diameter pots. Moreover, Raimondi et al. (2006) found that plant density increase from 66 to 100 plants per m² results in total yield increase. Further, in studies on canopy dynamics simulation (Van Oosteron et al., 2001), leaf area index showed increasing trend with the plant density enhancement.

The highest water and nutrient absorptions were recorded in the last crops week, when plant leaf area reached the highest expansion and the greenhouse temperature showed the highest value of 26.4°C (as an average of the two research years). As reported in Table 4, plant water consumption showed a similar trend to the yield one, with the highest values corresponding to 2.8 mS cm⁻¹ EC, whereas the highest nutrients consumption was assessed under the 2.8-3.1 mS cm⁻¹ EC range; the most diluted nutritive solution always resulted in the lowest absorption rates.

As for the comparison among the plant densities (Table 4), the highest daily values of both water and nutrient absorption occurred in the 12 plants per pot treatment, which also resulted in the best yield (Table 2); the highest plant density (18 plants per pot) always showed the lowest consumptions.

Compared to our research findings, a similar plant response to water deficit was recorded in previous investigations (Savvas *et al.*, 2007), where the increase of the nutrient solution strength caused the reduction of plant water absorption. The latter represents a salinity adaptation mechanism, consisting of leaf area and stomata decrease which in turn contributes to reducing transpiration and increasing water use efficiency (Chartzoulakis and Klapaki, 2000).

The quality indicators were significantly affected by the nutritive solution strength (Table 5), as their trends were always increasing with the electrical conductivity raise from 2.2 to 2.8 mS cm⁻¹, whereas no further increases were recorded in the last 0.3 mS cm⁻¹ rise.

The quality parameters showed decreasing trends as a function of the plant density increase (Table 5), with the 9 and 12 plants per pot treatments generally attaining the highest values and the 18 plants per pot treatment displaying the worst performances.

In previous investigation (Adams and Ho, 1989), an increase in sugar content and titratable acidity was reported as a consequence of salinity increase or water deficit. Moreover, Raimondi *et al.* (2006) found that nutrient solution EC interacted with the cultivars in modifying leaf antioxidant content: i.e. Napoletano leaves showed an ascorbate increase with the EC enhancement, whereas Genovese displayed opposite trend. The same authors also recorded that the plant density increase from 66 to 100 plants per m² did not

Nutritive solution EC - (mS·cm ⁻¹)	Maximum daily absorptions								
	Water (L·m ⁻²⁾	Nitrogen (g·m⁻²)	Phosphorus (g·m ⁻²)	Potassium (g·m⁻²)	Calcium (g·m ⁻²)	Magnesium (g⋅m⁻²)	lron (mg·m⁻²)		
2.2	1.6 d	0.35 c	0.11 c	0.42 c	0.29 c	0.11 c	5.76 c		
2.5	2.0 c	0.51 b	0.15 b	0.61 b	0.42 b	0.16 b	7.20 b		
2.8	2.6 a	0.76 a	0.21 a	0.92 a	0.64 a	0.23 a	9.00 a		
3.1	2.4 b	0.78 a	0.22 a	0.93 a	0.65 a	0.24 a	8.46 a		
No. plants per pot									
9	2.2 b	0.62 b	0.19 b	0.74 b	0.53 b	0.19 b	7.56 b		
12	2.6 a	0.73 a	0.21 a	0.90 a	0.58 a	0.23 a	9.00 a		
15	2.1 b	0.58 b	0.17 c	0.70 b	0.49 c	0.17 c	7.38 b		
18	1.7 c	0.47 c	0.13 d	0.54 c	0.40 d	0.15 d	5.94 c		

Table 4 - Basil water and nutrient absorptions

Table 5 - Basil leaves quality indicators

Nutritive solution EC (mS·cm ⁻¹)	Dry residue (%)	Soluble solids (°Brix)	Titratable acidity (g · 100 g⁻¹ d.w.)	Glucose (g · 100 g⁻¹ d.w.)	Fructose (g · 100 g⁻¹ d.w.)	Sucrose (g · 100 g ⁻¹ d.w.)	Ascorbic acid (mg·100 g⁻¹ d.w.)
2.2	9.5 c	3.2 c	0.76 c	1.80 c	2.25 c	0.40 c	508.4 c
2.5	9.7 bc	3.4 bc	0.84 b	2.12 b	2.52 b	0.52 b	585.7 b
2.8	10.0 ab	3.6 ab	0.96 a	2.34 a	2.83 a	0.63 a	703.8 a
3.1	10.0 a	3.7 a	1.02 a	2.47 a	2.98 a	0.67 a	744.6 a
No. plants per pot							
9	10.0 a	3.6 a	0.98 a	2.30 a	2.86 a	0.64 a	708 a
12	10.0 a	3.6 a	0.96 a	2.24 a	2.78 a	0.62 a	689.5 a
15	9.8 ab	3.4 ab	0.88 b	2.14 ab	2.54 b	0.52 b	617.3 b
18	9.6 b	3.3 b	0.78 c	2.02 b	2.40 b	0.45 c	527.4 c

affect fresh produce quality, but it just lowered the soluble solids content.

As reported in Table 6, mineral nutrient concentrations were significantly affected by the nutritive solution strength, as their trends were always increasing with the electrical conductivity raise from 2.2 to 2.8 mS·cm⁻¹, whereas no further increases were recorded in the last 0.3 mS·cm⁻¹ rise.

With regard to plant density (Table 6), the 9 and 12 plants per pot treatments always resulted in the highest nutrient accumulation in the leaves, except for the nitrates which showed an opposite trend, increasing from the lowest to the highest density. The decreasing trend of the leaves mineral ion concentrations as a function of the pot plant density increase resulted in relation with the plant nutrient

4. Conclusions

From research carried out in southern Italy on soilless pot-grown basil, it can be inferred that the 2.8 mS·cm⁻¹ nutritive solution resulted in the best product yield and quality, whereas a further increase to 3.1 mS·cm⁻¹ caused the reduction of the water and nutrient efficiency use. Moreover, the 12 plants per pot density showed the optimal compromise between the individual plants and the pot plants set performances, providing with the highest production and leaves quality. Indeed, density intensification to 15 and further to 18 plants per pot caused the reduced efficiency use of water and nutrients and accordingly the plant growth worsening, as well as the crop cycle extension up to 2.7 days.

Nutritive solution EC (mS·cm ⁻¹)	Nitrates (g∙kg⁻¹ d.w.)	Phosphates (g·kg⁻¹ d.w.)	Sulphates (g·kg⁻¹ d.w.)	Calcium (g·kg ⁻¹ d.w.)	Magnesium (g·kg ⁻¹ d.w.)	Potassium (g∙kg⁻¹ d.w.)
2.2	3.2 c	3.6 c	1.5 c	5.9 c	3.3 c	45.3 c
2.5	3.7 b	4.1 b	1.8 b	6.4 b	3.8 b	48.8 b
2.8	4.4 a	5.0 a	2.2 a	7.1 a	4.3 a	53.4 a
3.1	4.8 a	5.3 a	2.3 a	7.3 a	4.4 a	55.0 a
No. plants per pot						
9	3.4 d	4.8 a	2.1 a	7.1 a	4.2 a	54.2 a
12	3.8 c	4.7 ab	2.1 a	7.0 ab	4.1 ab	52.4 ab
15	4.2 b	4.4 bc	1.9 ab	6.5 bc	3.8 bc	49.7 bc
18	4.7 a	4.1 c	1.7 b	6.1 c	3.6 c	46.2 c

Table 6 - Basil leaves chemical composition

absorptions (Table 4).

Notably, the increase of mineral cations concentration in the plant tissues is caused by salt ion accumulation in the rizhosphere (Sonneveld, 2002) as a consequence of the plant active exclusion in response to salt occurrence in the external solution (Bethke and Drew, 1992). Moreover, the nitrate concentration increase in response to nutritive solution strength raise recorded in our research is consistent with the reports of previous investigations (Tesi et al., 1997; Raimondi et al., 2006). As for plant density, the increasing nitrate accumulation corresponding to the enhancement of the plants number per pot was presumably caused by the gradual light conditions worsening. Contrastingly, in previous research (Tesi et al., 1995) a decreasing trend of nitrate accumulation as a function of plant density increase was reported. Interestingly, in our research the nitrate concentration was always very low and, accordingly, basil leaves consumption not exceeding 563 g per day complies with the Acceptable Daily Intake for nitrate (222 mg·d⁻¹ for 60 kg adult) (Authority EFS, 2008).

Acknowledgements

The authors wish to thank Mr. Rosario Nocerino and Mr. Luigi Sannino for their valuable assistance with the greenhouse equipments and farming practices.

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