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Different species of basil need different ammonium to nitrate ratio in hydroponics' system

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ABSTRACT

Basil is a very important medicinal plant and culinary spice, and is marketed fresh, dried or frozen. In crop nutrition, nitrogen is essential for plant growth and as a macro-element, is part of the proteins' structure and participates in the metabolic processes involved in the synthesis and energy transfer. It has been shown that a balance between ammonium and nitrate favors plant growth and that the degree of benefit varies among crops. This study was conducted to evaluate the growth of two varieties of basil in function of four nutrient solutions containing different NH_4^+/NO_3^- ratios. Results showed that different variety response differently to nutrient solution. Although the highest yield in both varieties (sweet and purple) was obtained when fed by nutrient solution without ammonium but their response on quality indices were different due to nitrate ammonium ratio in nutrient solutions. The highest total phenol content of sweet and purple basil was 92 and 100 mg gallic acid equivalent per gram of dry weight respectively, while the highest antioxidant capacity was obtained in purple variety grown in nutrient solution 2 (NH_4^+:1/NO_3:4) and the lowest value were related to sweet variety with the same nutrient solution. Moderate content of total nitrogen can be suitable for sweet variety while for purple variety nutrient solution with low amount of ammonium can be more suitable.

Key words: NFT, antioxidant, phenol, vitamin C, chlorophyll, yield

IZVLEČEK

RAZLIČNI BIOTIPI BAZILIKE POTREBUJEJO V RAZMERAH HIDROPONSKEGA GOJENJA RAZLIČNA RAZMERJA NITRATA IN AMONIJA

Bazilika je zelo pomembna zdravilna rastlina in začimba, ki se prodaja sveža, suha ali zmrznjena. V prehrani rastlin je dušik esencialni makroelement, potreben za rast, sintezo beljakovin, druge presnovne procese in pretvorbo energije. Znano je, da ravnovesje med amonijsko in nitratno obliko dušika izboljšuje rast in, da se njuno ugodno razmerje spreminja glede na posamezne rastline. V raziskavi smo ovrednotili rast dveh biotipov širokolistne bazilike (zeleno- in vijoličnolistno) v štirih hranilnih raztopinah, ki so vsebovale različna razmerja NH_4^+/NO_3^-. Rezultati so pokazali različen odziv obeh biotipov na razmerja hranil v raztopinah. Čeprav je bil pridelek obeh (zelene in vijolične) največji v hranilni raztopini z amonijsko obliko dušika, so kvalitetni kazalci variirali glede na razmerja nitrata in amonija. Največja vsebnost celokupnih fenolov v zeleni in vijolični baziliki je bila 92 in 100 mg ekvivalenta galne kisline na gram suhe mase, vendar je bila največja antioksidativna sposobnost dosežena pri vijolični baziliki, ki je rastla v hranilni raztopini 2 (NH_4^+:1/NO_3:4) in najmanjša vrednost pri zeleni baziliki, ki je uspevala v isti hranilni raztopini. Zmerna vsebnost celokupnega dušika je primerna za gojenje zelene širokolistne bazilike, medtem ko je za gojenje vijolične primernejša hranilna raztopina z manjšo vsebnostjo amonija.

Ključne besede: NFT hranilna raztopina, antioksidanti, fenoli, vitamin C, klorofil, pridelek

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1 INTRODUCTION

Among the various medicinal and culinary herbs, some herbs such as basil (*Ocimum basilicum* L.) are of particular interest because they may be used for the production of raw materials or preparations containing phyto-chemicals with significant antioxidant capacities and health benefits (Exarchou *et al*. 2002). Basil is a very important medicinal plant and culinary spice, and is marketed fresh, dried or frozen. Traditionally, basil has been used as a medicinal plant in the treatment of headache, cough, diarrhea and kidney malfunctions (Simon *et al*. 1984), against insect bites, acne (Waltz, 1996), and it has long been used to flavor foods, as well as dental and oral products (Simon *et al*., 1984).

In crop nutrition, nitrogen is essential for plant growth, and is the fourth most abundant element found in plant tissues after carbon, oxygen and hydrogen. As a macro-element, it is part of the proteins' structure and participates in the metabolic processes involved in the synthesis and energy transfer. It is absorbed by the plant roots in the form of ammonium ($NH4^+$) or nitrate (NO_3^-) ions. The form in which N is supplied influences directly the absorption and the rhizosphere pH (Trejo *et al*. 2008). In hydroponics, both nitrate and ammonium forms are used in nutrient solutions. It can be argued that either of the two is beneficial or that either may equally cause growth problems or imbalances in the nutrient solution. It has been shown that a balance between ammonium and nitrate favors plant growth and that the degree of benefit varies among crops (Mengel and Kirkby, 1987).

For most plant species, NO_3^- supply combined with low quantities of NH_4^+ favors growth, but the response depends on the species and the age of the plant. Mengel and Kirkby (1987) reported plant species that grow better when nitrogen is administered as NO_3^- instead of NH_4^+. Reports have indicated that the incorporation of nitrogen in N-NH_4^+ form is toxic for many species, even in low concentrations (Salsac *et al*. 1987).

In other hands the yield depends to a large degree on the content of photosynthetically active pigments. Authors of numerous papers showed a close correlation between the level of these pigments and nitrogen content in leaves determined by the dose and time of fertilization (Baghour *et al*. 2000; Biczak *et al*. 1998; Smith 1999; Swiader and Moore 2002).

Hydroponic systems have consented to improve the cropping management and to reach higher produce quality standards in leafy vegetables (Santamaria *et al*. 2002). However, the identification of optimal soilless practices such as plant density and nutrient solution composition are critical factors in order to increase yield, reduce pathogen incidence, enhance dry matter production, improve antioxidant activity, and reduce nitrate content (Chen et al. 2004; Fravel and Larkin 2002).

This study was conducted to evaluate the growth of two variety of basil in function of four nutrient solutions containing different NH_4^+/NO_3^- ratios to test basil species response to nutrient solution prepared with NO_3^- combined with NH_4^+.

2 MATERIALS AND METHODS

In order to determine, the effect of different nutrient solution on the yield and quality indices of sweet and purple basil in a NFT culture, an investigation was conducted in a PVC greenhouse in 2010 in a completely randomized experimental design with three replications.

Basil seeds were sown in May 2010 and transferred to NFT system after two weeks. Harvesting took place during July to September 2010 (four times). Nutrient solutions (Tables 1-2) were prepared with municipal tap water and delivered to plants by a mist irrigation system.

Yield, dry mater and ash percent, total soluble solid, total acidity, total carotenoid, total phenol, vitamin C and antioxidant capacity of basil leaves were determined.

2.1 Determination of dry matter, total soluble solids, and ash:

the dry matter of samples was determined by drying at 75±5 °C until they reached constant weight (AOAC 1984) and then ashes in electrical oven. Titrable acidity (TA) was measured by the titrimetric method (AOAC 1984).

2.2 Determination of ascorbic acid:

Ascorbic acid was quantitatively determined according to 2, 6- dichlorophenolindophenol dye method (Ranganna 1997). The ascorbic acid of fresh samples (10 g) was extracted by grinding in a suitable medium with a small amount of sand and using 3% metaphosphoric acid (v/v) as a protective agent. The extract was made up to a volume of 100 ml mixed and centrifuged at 3000 g for 15 min at room temperature. Ten ml was titrated against standard 2, 6-dichlorophenolindophenol dye, which was already standardized against standard ascorbic acid. Results were expressed as mg/100 g on fresh weight (FW) basis.

2.3 Determination of total chlorophyll:

Total chlorophyll mg/100g was determined by a modified method of Ranganna (1997) using acetone and petroleum ether as extracting solvents and measuring the absorbance by spectrophotometer.

2.4 Determination of total phenolic compound:

the methanol extracts of basil were used for the determination of total phenolics. Total phenolic content was evaluated by colorimetric analyses using Folin-Ciocaltaue's phenol reagent (Singleton and Rossi, 1965). The content of total phenolics was expressed as mg galic acid equivalent per 100 g of leaves.

2.5 2-Diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging activity:

the free radical-scavenging activity against DPPH radical was evaluated according to the method of Leong and Shui (2002) and Miliauskas et al. (2004) with minor modification. According to principle of this method, in the presence of an antioxidant, the purple color intensity of DPPH solution decays and the change of absorbance are followed spectrophotometrically at 517 nm. The scavenging activity was expressed as IC_{50} (mg/ml).

2.6 Nutrient content and nitrate analysis:

Phosphorus, Calcium and magnesium in leaves were measured by spectrometry (JENWAY 6105 U.V/V). Nitrate was measured in following to Humphries (1956).

The resultant data were subjected to analysis of variance using SAS statistical program. Means were separated by Tukey's Multiple Range Test.

3 RESULTS AND DISCUSSION

Due to significant interaction between nutrient solution and variety we are not able to propose a significantly better nutrient solution for both varieties so we discuss only about interaction.

Interaction between variety and nutrient solution on total phenol showed that the highest amount of phenol was obtained in purple variety with nutrient solution 2 (NH_4^+:1/NO_3:4) and the lowest value were related to sweet variety with the same nutrient solution (NH_4^+:1/NO_3:4). This variety show the highest phenol content when fed with nutrient solution 3 (NH_4^+:0.5/NO_3:4). These values were significantly different from other values (figure 1).

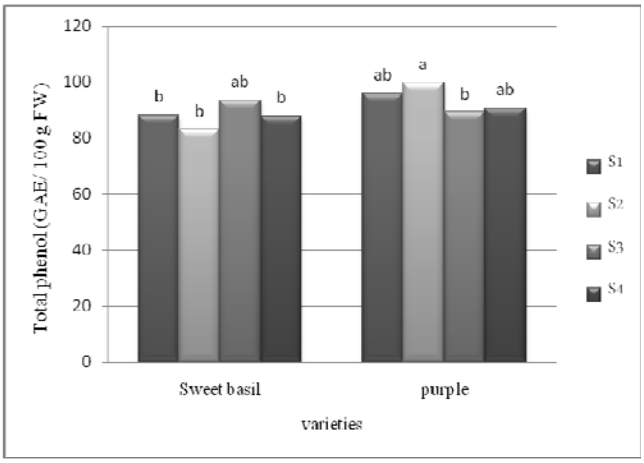

Figure 1: Nutrient solution and variety interaction on basil total phenol (GAE: Gallic acid equivalent)

The highest total phenol content of sweet and purple basil was 92 and 100 mg gallic acid equivalent per gram of dry weight of basil leaves, respectively. The highest value in this recent study was higher than the value (51.1 mg gallic acid equivalent per gram of dry weight) reported by Juliani and Simon (2002). These results were not doubtful because phenolic compounds in plant foods are largely influenced by genetic factors and environmental conditions including nutrition condition (Bravo, 1998). The difference in phenolic content could affect the antioxidant capacity of plants, because many phenolic compounds in plants are good sources of natural antioxidants (Amiot *et al.* 1997; Ho 1992).

The highest number of antioxidant capacity was obtained in purple variety with nutrient solution 2 (NH_4^+:1/NO_3:4) and the lowest value were related to sweet variety with the same nutrient solution. This variety showed the highest antioxidant capacity when fed with nutrient solution 3 (NH_4^+:0.5/NO_3:4) and 4 (NH_4^+:0/NO_3:5) (figure 2).

Antioxidants, including vitamin C, carotenoid, phenols and chlorophyll, are responsible for the level of antioxidant capacity of a leaf extract. Thus, enhanced amounts of the biologically active reduced form of antioxidants can be indicative of an increased antioxidant capacity, i.e. the healthy properties of basil leaves extracts (Sgherri *et al.*, 2010).

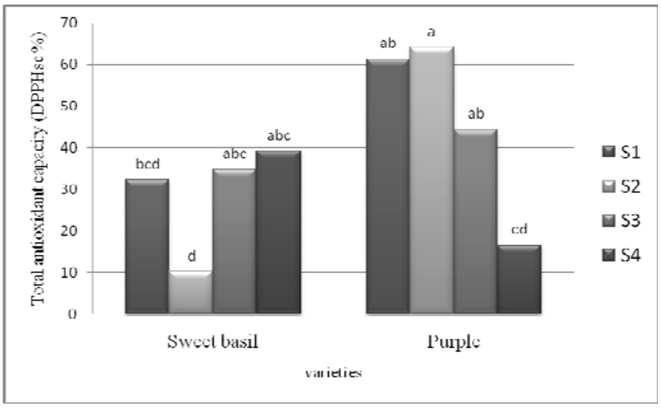

Figure 2: Nutrient solution and variety interaction on basil antioxidant capacity

Interaction between variety and nutrient solution on total chlorophyll showed that the highest chlorophyll was obtained in purple variety with nutrient solution 3 (NH_4^+:0.5/NO_3:4) but nutrient solution did not affect sweet basil total chlorophyll (figure 3). Nutrient solution has influenced plant biochemical characteristics such as concentrations of photosynthetic pigments. Surprisingly sweet and purple varieties total chlorophyll was similar in spite of their different color.

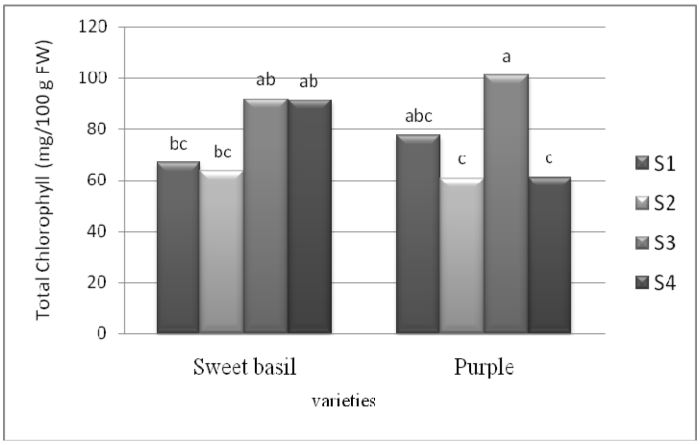

Figure 3: Nutrient solution and variety interaction on total chlorophyll content in fresh basil

Interaction between variety and nutrient solution on Mg, P and Ca content showed that the highest level of these elements in purple variety was obtained in plants that were feed on with nutrient solution 3 (NH_4^+:0.5/NO_3:4), while the highest level of Mg and Ca in sweet basil was obtained in plants that were feed on with nutrient solution 4 (NH_4^+:0/NO_3:5). On the other hand the highest level of P was obtained when the sweet basil was feed on with nutrient solution 2 (NH_4^+:1/NO_3:4).

Interaction between variety and nutrient solution on yield showed that the highest yield was obtained in purple and sweet variety when they were grown in nutrient solution 4 (NH_4^+:0/NO_3:5) and the lowest value were related to sweet variety with nutrient solution 2 (NH_4^+:1/NO_3:4) (figure 7). For most plant species, an $NO3^-$ supply combined with low quantities of $NH4^+$ favors growth, but the response depends on the species and the age of the plant. Mengel and Kirkby (1987) reported plant species that grow better when nitrogen is administered as $NO3^-$ instead as $NH4^+$. Reports have indicated that the incorporation of nitrogen in N-$NH4^+$ form is toxic for many species, even in low concentrations (Salsac *et al* 1987). A positive response was also observed when basil plants were fed with both nitrogen sources (NH_4^+ and NO_3^-). This coincides with Zornoza et al. (1988), who found that sweet peppers increased their yield for 10% when they were fertigated with a nutrient solution containing 80/20 (NO_3^-/NH_4^+) compared to the fertigation with NO_3^- as the only source of nitrogen in the nutrient solution. Goyal et al. (1982) and Xu et al. (1992) also stated that plant growth is notably favored when treated with both forms of nitrogen, but it is needed to find the optimal nitrate/ammonium ratios for each crop.

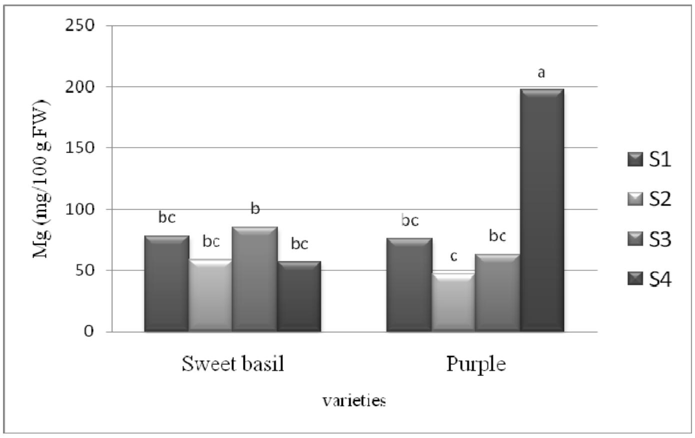

Figure 4: Nutrient solution and variety interaction on Mg content in fresh basil

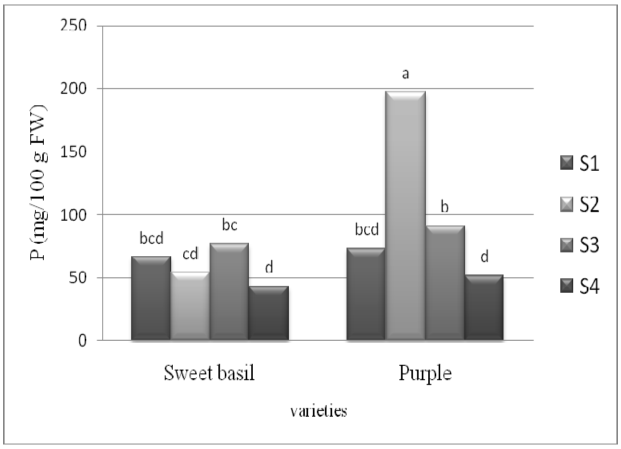

Figure 5: Nutrient solution and variety interaction on P content in fresh basil

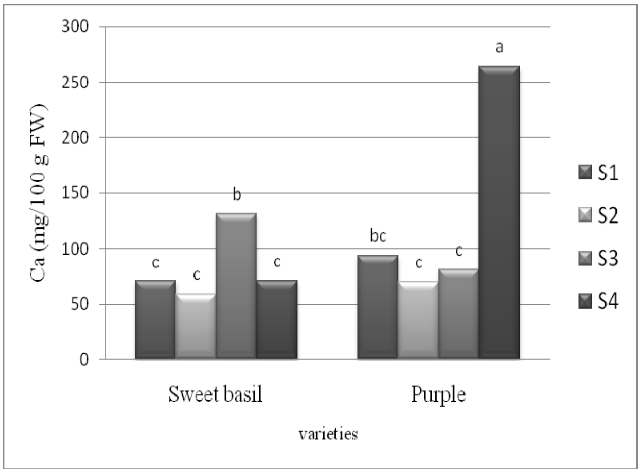

Figure 6: Nutrient solution and variety interaction on Ca content in fresh basil

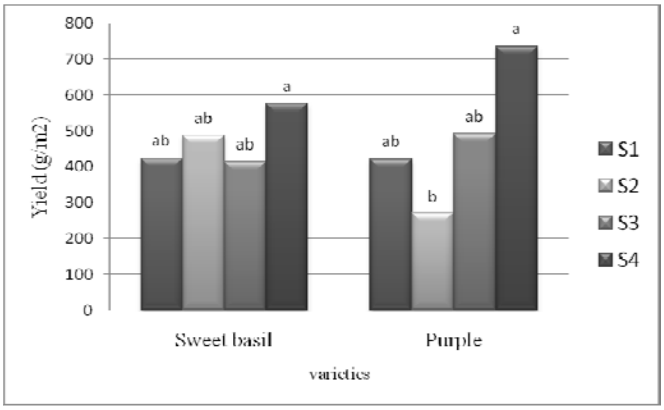

Figure 7: Nutrient solution and variety interaction on basil yield in g/m^2

Interaction between variety and nutrient solution on ash showed that the highest ash was obtained in purple variety with nutrient solution 4 (NH_4^+:0/NO_3:5) and the lowest value were related to sweet variety with nutrient solution 2 (NH_4^+:1/NO_3:4). This variety showed the highest ash content when fed with nutrient solution 3 (NH_4^+:0.5/NO_3:4). These values were significantly different from other values (figure 8).

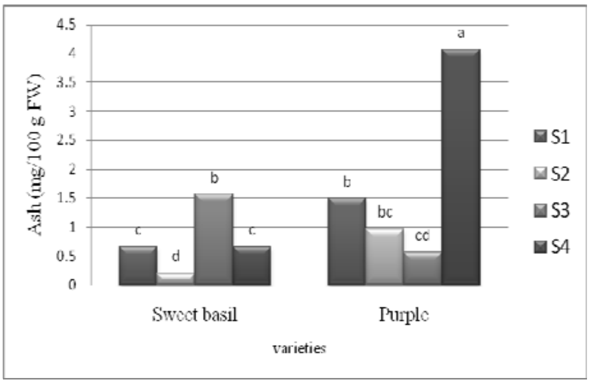

Figure 8: Nutrient solution and variety interaction on basil ash

Interaction between variety and nutrient solution on vitamin C content showed that the highest vitamin C was obtained in purple and sweet variety with nutrient solution 3 (NH_4^+:0.5/NO_3:4) and 1 (NH_4^+:1.5/NO_3:3.5) respectively (figure 9).

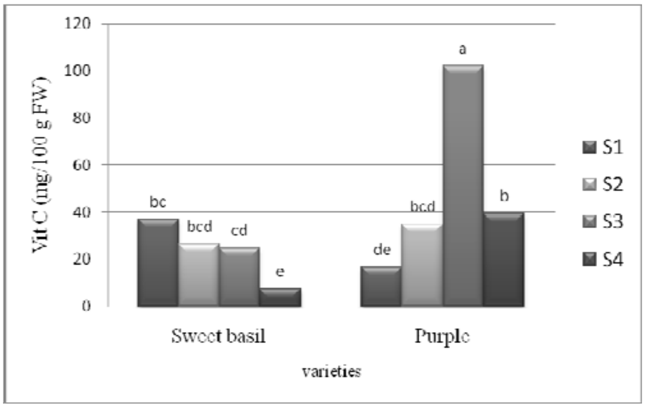

Figure 9: Nutrient solution and variety interaction on basil vit. C

Interaction between variety and nutrient solution on nitrate accumulation showed that the highest and the lowest nitrate accumulation was obtained in both varieties with nutrient solution 4 (NH_4^+:0/NO_3:5) and 1 (NH_4^+:1.5, NO_3:3.5) respectively (figure 10).

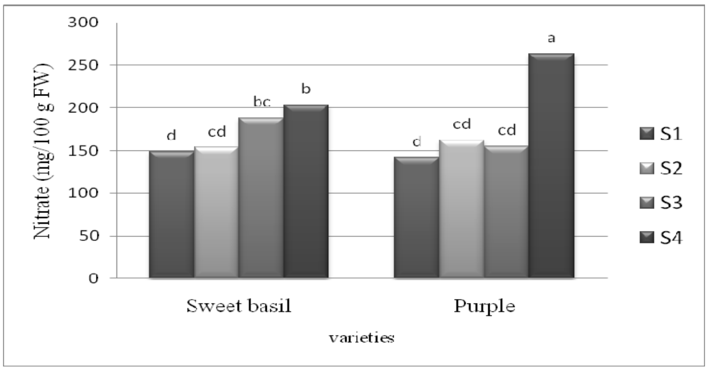

Figure 10: Nutrient solution and variety interaction on basil Nitrate

4 CONCLUSION

Results indicated that sweet basil that have green color and high chlorophyll content responded not significantly to ammonium content in nutrient solution on yield and some other indices while nitrate accumulation is a big problem when this variety irrigated with high nitrogen content nutrient solution so a moderate content of total nitrogen such as nutrient solution 2 or 3 can be suitable for this variety cultivation. In purple variety with lower amount of chlorophyll NO3:NH4 ratio is important. Nutrient solution with low amount of ammonium such as nutrient solution 3 or 4 can be more suitable for this variety in soilless culture.

Table 1: Macronutrients used in nutrient solutions.

Nutrient solution	Meq/l										
	KNO_3	KH_2PO_4	K_2SO_4	NaCl	$CaNO_3$	$MgSO_4$	$MgHPO_4$	NH_4NO_3	$CaSO_4$	SO_4	NO_3
1	1	1.5	2	0.1	1	0.5	1.5	1.5	1.5	4	3.5
2	2	1.5	1	0.1	1	0.5	1.5	1	1.5	3	4
3	2	1.5	1	0.1	2	0.5	1.5	0.5	0.5	3	4
4	3	1.5	0	0.1	2	0.5	1.5	0	0.5	1	5

Table 2: Micronutrient used for nutrient solution preparation.

Compound	Mg/l Irrigation solution
(NH4) 6Mo 7O2/4H2O	0.1
H3BO3	1.5
MnSO4/4H2O	2
CuSO4/5H2O	0.25
ZnSO4/7H2O	1
Sequesteren Fe 136	10

5 REFERENCES

Amiot M.J., Fleuriet A., Cheynier V., Nicolas J. 1997. Phenolic compounds and Oxidative mechanisms in fruit and vegetables. In Tomas-Barberan FA, Robins RJ. (eds.). Phytochemistry of fruit and vegetables. Clarendon Press, Oxford. P: 51-85.

AOAC .1984. Officials methods of analyses (14th ed.). Arlington, VA, USA. Association of official analytical chemist.

Baghour M., Ruiz J.M., Romero L. 2000. Metabolism and efficiency in nitrogen utilization during senescence in pepper plants: Response to nitrogenous fertilization. J. Plant Nutrition, 23(1): 91-101. DOI: 10.1080/01904160009382000

Biczak R., Gurgul E., Herman B. 1998. The effect of NPK fertilization on yield and content of chlorophyll, sugars and ascorbic acid in celery. Folia Hort. 10(2): 23-34.

Bravo L. 1998. Polyphenols: chemistry, dietary sources, metabolism, and nutritional significance. Nutrition. Review. 56(11): 317-333. DOI: 10.1111/j.1753-4887.1998.tb01670.x

Chen B.M., Wang Z.H., Li S.X., Wang G.X., Song H.X., Wang X.N. 2004. Effects of nitrate supply on plant growth, nitrate accumulation, metabolic nitrate concentration and nitrate reductase activity in three leafy vegetables. Plant Science. 167: 635–643. DOI: 10.1016/j.plantsci.2004.05.015

Exarchou V., Nenadis N., Tsimidou M., Gerothanassis I.P., Troganis A., Boskou D. 2002. Antioxidant activities and phenolic composition of extracts from Greek oregano, Greek sage and summer savory. Journal of Agricultural and Food Chemistry. 50(19): 5294–5299. DOI: 10.1021/jf020408a

Fravel D.R., Larkin R.P. 2002. Reduction of fusarium wilt of hydroponically grown basil by *Fusarium oxysporum* L. strain CS-20. Crop Protection. 21: 539–543. DOI: 10.1016/S0261-2194(01)00143-0

Goyal S.S., Huffaker R.C., Lorenz O.A. 1982. Inhibitory effects of ammoniacal nitrogen on growth of radish plants. II. Investigations on the possible causes of ammonium toxicity to radish plants and its reversal by nitrate. J. American Society Horticulture Science. 107: 130-135.

Ho C.T. 1992. Phenolic compounds on food: an overview. In Ho, C.T., C.Y. Lee M.T. Huang. Phenolic compounds in food and their effects on health I: analysis, occurrence, & chemistry. American Chemical Society, New York. P: 2-7.

Humphries E.C. 1956. Mineral components and analysis in “Modern methods” of plant analysis. 1:468-502. in Peach, K and M.V. Tracy. springer. Verlag. Berlin.

Juliani H.R., Simon J.E. 2002. Antioxidant of basil. In Janick, J and Whipkey, (eds.). Trend in new crops and new uses. ASHS Press, Alexandra, VA. P: 575-579.

Leong L.P., Shui G. 2002. An investigation of antioxidant capacity of fruits in Singapore markets. Food chemistry. 76: 69-75. DOI: 10.1016/S0308-8146(01)00251-5

Mengel K., Kirkby E.A. 1987. Nitrogen. p.347-374. In: Mengel, K and E.A. Kirkby (eds.), Principles of Plant Nutrition. 4th ed. International Potash Institute. WorldblaufenBern. Switzerland.

Miliauskas G., Venskutonis P.R., Van T.A. 2004. Screening of radical scavenging activity of some medicinal and aromatic plant extracts. Food chemistry. 85: 231-237. DOI: 10.1016/j.foodchem.2003.05.007

Ranganna S. 1997. In Manual of analysis of fruit and vegetable products. 9th edition, Tata Mc Graw Hill, New Delhi. Salsac L., Chaillou S., Morot J.F., Lesaint C., Jolivet E. 1987. Nitrate and ammonium nutrition in plants. Plant Physiology and Biochemical. 25: 805-812.

Santamaria P., Elia A., Serio F. 2002. Effect of solution nitrogen concentration on yield, leaf element content, and water and nitrogen use efficiency of three hydroponically-grown rocket salad genotypes.

J. Plant Nutrition. 25:245-258. DOI: 10.1081/PLN-100108833

Sgherri C., Cecconami S., Pinzino C., Navari-Izzo F., Izzo R. 2010. Levels of antioxidants and nutraceuticals in basil grown in hydroponics and soil. Food chemistry. 123: 416–422. DOI: 10.1016/j.foodchem.2010.04.058

Simon J.E., Chadwick A.F., Craker L.E. 1984. Herbs: An indexed bibliography,1971–1980. The scientific literature on selected herbs and aromatic and medicinal plants of the temperate zone. Hamden, CT: Archon Books.Singleton V.L., Rossi J.A. 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagent. American Journal of Enology Viticuture. 16(3): 144-158.

Smith D.L. 1999. Field evaluation of the chlorophyll meter to predict yield and nitrogen concentration of switchgrass. J. Plant Nutrition. 22(6): 1001-1010. DOI: 10.1080/01904169909365689

Swiader J.M., Moore A. 2002. SPAD-chlorophyll response to nitrogen fertilization and evaluation of nitrogen status in dryland and irrigated pumpkins. J. Plant Nutrition. 25(5): 1089-1100. DOI: 10.1081/PLN-120003941

Trejo L.I., Rodríguez-Mendoza M.N., Fernández-Luqueño F. 2008. Nutrición de cultivos. Manual Ed. Papiro Omega. México D.F.Waltz L. 1996. The herbal encyclopedia. Available from http://www.wic.net/waltzark/herbenc.htm.Xu Q.F., Tsai C.L., Tsai C.Y. 1992. Interaction of potassium with the form and amount of nitrogen nutrition on growth and nitrogen uptake of maize. J. Plant Nutrition. 15(1): 23-33.

Zornoza P., Caselles J., Carpena O. 1988. Influence of Light and NO3-:NH4+ ratio on nutrient uptake by pepper plant in sand culture. Soilless Culture. 4: 65-74.