

Effect of organo-mineral fertilizers on growth, biomass, and mineral nutrition of natural non-calorie sweetener plant (*Stevia rebaudiana* Bertoni) in northwestern Morocco

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Abstract

To maintain consistently high biomass productivity, soil nutrient management is essential, and fertilization is the only way to supply soil nutrients. This study investigated the effect of organo-mineral fertilizers treatments on growth, biomass, and mineral nutrition of stevia (*Stevia rebaudiana* Bertoni) plants under northwestern Moroccan conditions. Four treatments of organo-mineral fertilizers were tested: T0 (absolute control), T1 (300:100:240 kg nitrogen (N), phosphorus (P), and potassium (K) ha⁻¹), T2 (40 t ha⁻¹ of the organic fertilizer), and T3 (75:60:20 kg NPK ha⁻¹ + 10 t ha⁻¹ of the organic fertilizer). Two harvests were taken during the study. Cumulative results showed that significantly higher fresh biomass (168.30 g plant⁻¹), fresh leaf yield (125.77 g plant⁻¹), and dry leaf yield (28.93 g plant⁻¹) were obtained with T1 as compared to other fertilized treatments and control. This superior

combination also resulted in considerably greater mean contents of N (1.80%), phosphorus P (0.17%), and K (2.27%) in leaf. Plant height, stem diameter, number of leaves plant⁻¹, and NPK uptake data showed the same trend as dry leaf yield and NPK content. Also, the results indicated that all the above parameters were influenced by time of harvest. The greater growth, biomass, and mineral nutrition were obtained at the first harvest (85 days after transplanting) compared with the second harvest (56 days after first harvest) in all treatments. Consequently, 300:100:240 kg ha⁻¹ NPK combination level was appeared to be optimal for higher biomass and mineral nutrition of stevia grown during summer season.

Keywords: Nitrogen, Phosphorus, Potassium, Organic fertilizer, Stevia, Biomass.

1. Introduction

The use of mineral and organic fertilizers has its advantages and disadvantages in the context of nutrient supply, crop growth and environmental quality. Mineral fertilizers are soluble and immediately available to the plants, and are relatively inexpensive. However, over-application of mineral fertilizer can result in negative effects such as nutrient loss, pollution of water resources, soil acidification or basification, reductions in useful microbial communities, and increased sensitivity to harmful insects [1]. Organic fertilizer has a number of disadvantages, comprising low nutrient content, slow release of nutrient, and the major plant nutrients may not exist in sufficient quantity. However, organic fertilizer has multiple advantages due to the balanced supply of nutrients, provide micronutrients and plant-growth-promoting molecules, increased soil nutrient availability due to increased soil microbial activity, the decomposition of harmful elements, soil structure improvements and root development, and increased soil water availability [2].

The success of sustainable agriculture depends upon the availability of good quality organic fertilizers. Among the sources of available organic fertilizers, farmyard manures and vermicompost are easily available and are potential sources due to the presence of readily

available plant nutrients, growth-enhancing substances, and a number of beneficial microorganisms such as nitrogen-fixing, phosphorus-solubilizing, and cellulose-decomposing organisms. However, the simultaneous use of mineral and organic fertilizers has revealed diverse results relative to the plant types and soil characteristics [2, 3, 4].

Stevia (*Stevia rebaudiana* Bertoni) is a natural non-calorie sweetener plant (family *Asteraceae*), originating from the Amambay region in the north-east of Paraguay. Dry leaves are the economic part in stevia plant, with a high concentration of steviol glycosides (stevioside, rebaudioside A, B, C, D, steviol, dulcoside A, steviolbioside), which are many times sweeter than sugarcane and sugarbeet but importantly without any calories [5]. Due to the non-calorie nature of this crop, stevia leaves and their compounds are used in many therapeutic applications such as diabetes, obesity, plague retardant, hypoglycemia, indigestion, dental health, yeast infection, oral health, skin toning and healing burns and wounds [6].

2. Material and methods

2.1. Experimental details and growth conditions

A pot experiment was carried out during two successive stevia growing seasons: summer season and fall season of 2016, in open field at the Regional Centre of Agronomic Research of Rabat in Morocco (INRA) (34.21 N, 6.40 E, 10.5 m above mean sea-level). The area is located in sub humid region of north-western Morocco, with mean maximum temperature of 27.1 °C in August and mean minimum of 8°C in January. The average annual rainfall received is about 554 mm, of which about 74 percent is received during November to March.

The INRA stevia variety was sown into plug trays filled with soil and commercial substrate on March 25th, 2016 in the greenhouse. Two-month-old uniform seedlings were transplanted in the plastic pots on May 27th, 2016, with two plants per pot. The 20 L pots were filled with 2

kg of gravel at bottom for drainage and 15 kg of sandy soil to avoid any residual effect of the fertilizers. The soil contained 4.3% clay, 11.4% silt, and 81.3% sand. The organic matter content was 2.2 %, the pH was 8.10 and the nitrogen (N), phosphorus (P), and potassium (K) contents were 31.7, 6.9, and 19.5 ppm, respectively. Soil moisture at field capacity was 13.22 % and soil moisture at permanent wilting point was 4.67 %. Soil density (ρ) was 1.3 g cm^{-3} , which was used to convert doses of NPK from kg ha^{-1} to g pot^{-1} . There were total four treatments including control, three replications and three pots per replicate of stevia plants. Totally 36 experimental pots arranged according to a randomized complete block design. The organo-mineral fertilizers treatments were as follows: T0, used as control; T1, the combination (300:100:240 kg NPK ha^{-1}) who gave the best results in earlier research [7]; T2, recommended level of organic fertilizer (40 t ha^{-1}); and T3, the best mixture (75:60:20 kg NPK ha^{-1} + 10 t ha^{-1} of the organic fertilizer) selected through a preliminary plots study.

Organic manure used in this study was cattle manure. The main characteristics of the organic manure were: pH water, 7.11; EC at 25°C , 8.21 dS m^{-1} ; total N, 0.81%; total C, 13.78%; available P, K, Mg, Ca, and Na were 2.31%, 3.68%, 1.51%, 1.88%, and 1.86% respectively. The soil and cattle manure were analysed in laboratory of Research unit on Environment and Conservation of Natural Resources INRA, RCAR of Rabat. The required amount of organic manure as per treatment was weighed, added and well mixed in the soil before transplanting stevia. The NPK fertilizers were applied in the form of ammonium nitrate (33% of N), triple superphosphate (45% of phosphorus pentoxide (P_2O_5)), and muriate of potash (50% of potassium oxide (K_2O)), respectively. A half dose of N and full dose of P and K as per treatment were applied at the time of transplanting, while remaining half dose of N was applied at 45 days after transplanting. But for the second ratoon crop entire P and K nutrients along with 50% of N was applied two days after harvest and the remaining 50% of N was applied at 45 days after harvest of the previous crop. The pots were irrigated manually to field

capacity (FC). The first crop was harvested at 85 days after transplanting on August 17, 2016, whereas the second crop was harvested at 56 days after first cutting on October 12, 2016 with the help of sickles. The plants of the whole pots were cut 10 cm above the base of the stem.

2.2. Measurements

2.2.1. Growth measurements

The growth parameters viz., plant height, stem diameter, and number of leaves plant⁻¹ were recorded in each harvest. The plant height was measured with a meter ruler from ground to the base of the fully opened leaf and the stem diameter was measured with slide calipers up to 0.01 mm accuracy. Fully opened leaves were counted in each plant and the mean was computed to get the average number of leaves plant⁻¹.

2.2.2. Yield analysis

Data on fresh biomass, fresh leaf yield, and dry leaf yield per plant were determined in each harvest. These parameters were chosen as important yield parameters of stevia because the leaves are the economic part of the plant. We estimated the fresh biomass, fresh and dry leaf yield per plant using one digital scale with precision of 0.01 g. The leaves were dried at 50°C temperature in hot air dryer for 6 hours and stored in clean gunny bags. At this temperature, the quality of dried leaves produced, in terms of colour, sweetness and nutrient content, was better compared with drying at 70°C [8].

2.2.3. Determination of NPK in leaf

The dried stevia leaf samples were prepared with a laboratory grinder having a sieve spacing of 2 mm to determine total nitrogen (N), phosphorus (P), and potassium (K) content in the leaf. Total N content was determined by using the Macro Kjeldahl digestion and distillation method [9], while total P and K were determined using a colorimetric method [10] and flame

photometer (model CL378) [11], respectively. NPK uptake was estimated by multiplying dry leaves yield and NPK content.

2.3. Statistical analysis

Data obtained were analyzed by the analysis of variance (ANOVA) using Statistical analysis System ver. 9.1 (SAS Institute Inc., Cary, NC., USA), and means were compared using Duncan's multiple range test (DMRT) at the 0.05 significance level.

3. Results

3.1. Growth responses

Plant height, stem diameter, and number of leaves plant⁻¹ were significantly affected by different organo-mineral fertilizers treatments in each harvest and in mean values (Table 1). Results showed that plants treated with fertilizers were more vigorous, had higher plant height, wider stem diameter, and more number of leaves than those of the control plants. Treatment T1 (300:100:240 kg NPK ha⁻¹) recorded significantly higher plant height (58.83 cm) followed by T2 (40 t ha⁻¹ of the organic fertilizer) (56.25 cm) and T3 (75:60:20 kg NPK ha⁻¹ + 10 t ha⁻¹ of the organic fertilizer) (50.05 cm) in mean values. The lowest plant height was with absolute control (28.94 cm). Also maximum stem diameter and number of leaves plant⁻¹ were obtained in the T1 treatment (6.54 mm and 323.05, respectively) as compared to T2 (5.85 mm and 278.83, respectively), T3 (5.21 mm and 206.27, respectively), and the control (3.78 mm and 35.83, respectively) in mean data. The control had significantly decreased plant height, stem diameter, and number of leaves plant⁻¹ until 50.81%, 42.20%, and 88.90%, respectively, compared to T1 in mean values. The plant height, stem diameter, and number of leaves plant⁻¹ in each harvest varied significantly due to fertilizers which followed the similar trend as in mean values. At the first harvest, significantly higher plant height, stem diameter, and number of leaves plant⁻¹ were obtained in comparison to the

control. However, for the second harvest, all parameters were found to be low in all the treatments.

Table 1. Effect of organo-mineral fertilizers on growth parameters of stevia.

Treatment	Plant height (cm)			Stem diameter (mm)			Number of leaves plant ⁻¹		
	Harvest 1	Harvest 2	Mean	Harvest 1	Harvest 2	Mean	Harvest 1	Harvest 2	Mean
T0	31.44 ^D	26.44 ^D	28.94 ^D	5.20 ^D	2.37 ^D	3.78 ^D	47.22 ^D	24.44 ^D	35.83 ^D
T1	70.44 ^A	47.22 ^A	58.83 ^A	9.96 ^A	3.12 ^A	6.54 ^A	341.33 ^A	304.77 ^A	323.05 ^A
T2	67.44 ^B	45.05 ^B	56.25 ^B	8.71 ^B	2.98 ^B	5.85 ^B	315.22 ^B	242.44 ^B	278.83 ^B
T3	61.06 ^C	39.05 ^C	50.05 ^C	7.66 ^C	2.77 ^C	5.21 ^C	262.78 ^C	149.77 ^C	206.27 ^C

* Means followed by different letters in each column are significantly different (Duncan multiple range test at the 5 % significance level).

T0 (Control), T1 (300:100:240 kg NPK ha⁻¹), T2 (40 t ha⁻¹ of the organic fertilizer), and T3 (75:60:20 kg NPK ha⁻¹ + 10 t ha⁻¹ of the organic fertilizer).

3.2. Yield responses

Data presented in Table 2 reveals that fresh biomass yield, fresh leaf yield, and dry leaf yield were significantly influenced by all fertilized treatments compared to the control in all harvests as well as in cumulative total. Plants grown at 300:100:240 kg NPK ha⁻¹ (T1) produced the highest total fresh biomass (168.30 g plant⁻¹), which was significantly higher compared to other fertilized treatments and control. The increase in the fresh biomass yield at treatment T1 was 83.89%, 16.82%, and 38.73% higher over control, treatments T2 (40 t ha⁻¹ of the organic fertilizer), and T3 (75:60:20 kg NPK ha⁻¹ + 10 t ha⁻¹ of the organic fertilizer), respectively, in cumulative total. Similar trend of fresh biomass yield was observed in each harvest. Fresh leaf yield and dry leaf yield were significantly influenced by the organo-mineral fertilizers in each harvest and in cumulative total. Data on cumulative total fresh and dry leaf yields revealed that treatment T1 recorded higher fresh leaf yield (125.77 g plant⁻¹) and dry leaf yield (28.93 g plant⁻¹) which was followed by treatments T2 (98.35 and 23.88 g plant⁻¹, respectively) and T3 (80.28 and 17.41 g plant⁻¹, respectively). The control resulted in significantly lower fresh leaf yield and dry leaf yield of 18.87 and 5.27 g plant⁻¹, respectively.

Also, the results indicated that yield parameters of stevia were influenced by time of harvest. The greater fresh biomass yield, fresh and dry leaf yield were obtained at the first harvest compared with the second harvest in all treatments.

Table 2. Effect of organo-mineral fertilizers on yield parameters of stevia.

Treatment	Fresh biomass (g plant ⁻¹)			Fresh leaf yield (g plant ⁻¹)			Dry leaf yield (g plant ⁻¹)		
	Harvest 1	Harvest 2	Total	Harvest 1	Harvest 2	Total	Harvest 1	Harvest 2	Total
T0	18.32 ^D	8.79 ^D	27.11 ^D	12.12 ^D	6.74 ^D	18.87 ^D	3.47 ^D	1.80 ^D	5.27 ^D
T1	96.37 ^A	71.93 ^A	168.30 ^A	69.35 ^A	56.42 ^A	125.77 ^A	19.46 ^A	9.47 ^A	28.93 ^A
T2	83.92 ^B	56.06 ^B	139.98 ^B	54.63 ^B	43.71 ^B	98.35 ^B	16.48 ^B	7.39 ^B	23.88 ^B
T3	65.95 ^C	37.16 ^C	103.11 ^C	50.55 ^C	29.72 ^C	80.28 ^C	12.48 ^C	4.92 ^C	17.41 ^C

* Means followed by different letters in each column are significantly different (Duncan multiple range test at the 5 % significance level).

T0 (Control), T1 (300:100:240 kg NPK ha⁻¹), T2 (40 t ha⁻¹ of the organic fertilizer), and T3 (75:60:20 kg NPK ha⁻¹ + 10 t ha⁻¹ of the organic fertilizer).

3.3. Nutrient (NPK) content in leaf

The effects of different treatments of organo-mineral fertilizers on nitrogen (N), phosphorus (P), and potassium (K) contents in dry leaf of stevia in each harvest and in mean values are presented in the [Table 3](#). These parameters were significantly influenced by different fertilization treatments compared to the control. Significantly maximum mean N content (1.80%) was observed on treatment T1 with 300:100:240 kg ha⁻¹ NPK, followed by T2 (40 t ha⁻¹ of the organic fertilizer) (1.69%) and T3 (75:60:20 kg NPK ha⁻¹ + 10 t ha⁻¹ of the organic fertilizer) (1.38%). Minimum mean N content (0.35%) was observed on control free of organo-mineral fertilizers. Also, treatment T1 recorded significantly greater P content (0.17%) as compared to the control (0.07%) and T3 (0.14%) but remained statistically at par with T2 (0.17%) in mean values. Also maximum mean K content (2.27%) was obtained with treatment T1 as compared to the control (0.77%) and other treatments. The treatment T1 increased the mean N, P, and K contents by 82.77%, 58.82%, and 66.08%, respectively,

compared to the control. The NPK contents in each harvest varied significantly due to fertilizers which followed the similar trend as in mean data.

Table 3. Effect of organo-mineral fertilizers on NPK (%) content in dry leaf of stevia.

Treatment	N (%)			P (%)			K (%)		
	Harvest 1	Harvest 2	Mean	Harvest 1	Harvest 2	Mean	Harvest 1	Harvest 2	Mean
T0	0.37 ^D	0.31 ^D	0.35 ^D	0.08 ^C	0.05 ^C	0.07 ^C	0.82 ^D	0.71 ^D	0.77 ^D
T1	1.80 ^A	1.80 ^A	1.80 ^A	0.17 ^A	0.17 ^A	0.17 ^A	2.27 ^A	2.27 ^A	2.27 ^A
T2	1.68 ^B	1.68 ^B	1.69 ^B	0.17 ^A	0.17 ^A	0.17 ^A	1.90 ^B	1.90 ^B	1.90 ^B
T3	1.38 ^C	1.38 ^C	1.38 ^C	0.13 ^B	0.14 ^B	0.14 ^B	1.41 ^C	1.41 ^C	1.41 ^C

* Means followed by different letters in each column are significantly different (Duncan multiple range test at the 5 % significance level).

T0 (Control), T1 (300:100:240 kg NPK ha⁻¹), T2 (40 t ha⁻¹ of the organic fertilizer), and T3 (75:60:20 kg NPK ha⁻¹ + 10 t ha⁻¹ of the organic fertilizer).

3.4. NPK uptake in leaf

The uptake of nitrogen (N), phosphorous (P), and potassium (K) by stevia showed significant differences due to different fertilization treatments during both harvests and in total data (Table 4). Significantly higher NPK uptake was recorded with T1 in total data (52.17, 5.04, and 65.73 g plant⁻¹, respectively) as against T2 (40.31, 4.15, and 45.50 g plant⁻¹, respectively) and T3 (24.10, 2.35, and 24.61 g plant⁻¹, respectively). Absolute control recorded significantly lowest NPK uptake in total data (1.89, 0.38, and 4.16 g plant⁻¹, respectively). Similar trend was observed with respect to NPK uptake during first harvest and second harvest. Treatment T1 recorded the highest N uptake (35.06 and 17.10 g plant⁻¹), P uptake (3.38 and 1.65 g plant⁻¹), and K uptake (44.22 and 21.50 g plant⁻¹) in the first and second harvests, respectively and followed by T2 and T3. Whereas, the lowest values were recorded from the control during both harvests. Actually, all organo-mineral combinations produced significantly better NPK uptake than the control. Among both cuttings in general the higher NPK uptake was seen in all the treatments of first cutting as compared to second cutting.

Table 4. Effect of organo-mineral fertilizers on NPK uptake (g plant^{-1}) in dry leaf of stevia.

Treatment	N uptake (g plant^{-1})			P uptake (g plant^{-1})			K uptake (g plant^{-1})		
	Harvest 1	Harvest 2	Total	Harvest 1	Harvest 2	Total	Harvest 1	Harvest 2	Total
T0	1.32 ^D	0.57 ^D	1.89 ^D	0.28 ^D	0.10 ^D	0.38 ^D	2.87 ^D	1.28 ^D	4.16 ^D
T1	35.06 ^A	17.10 ^A	52.17 ^A	3.38 ^A	1.65 ^A	5.04 ^A	44.22 ^A	21.50 ^A	65.73 ^A
T2	27.85 ^B	12.46 ^B	40.31 ^B	2.86 ^B	1.28 ^B	4.15 ^B	31.41 ^B	14.08 ^B	45.50 ^B
T3	17.30 ^C	6.79 ^C	24.10 ^C	1.64 ^C	0.70 ^C	2.35 ^C	17.65 ^C	6.96 ^C	24.61 ^C

* Means followed by different letters in each column are significantly different (Duncan multiple range test at the 5 % significance level).

T0 (Control), T1 (300:100:240 kg NPK ha^{-1}), T2 (40 t ha^{-1} of the organic fertilizer), and T3 (75:60:20 kg NPK ha^{-1} + 10 t ha^{-1} of the organic fertilizer).

4. Discussion

In this study, all treatments of organo-mineral fertilizers gave higher growth parameters compared to the control. The higher plant height with higher levels of nitrogen (N) (300 kg ha^{-1}), phosphorus (P) (100 kg ha^{-1}), and potassium (K) (240 kg ha^{-1}) nutrient combination increased the number of leaves plant^{-1} . This combination recorded significantly higher plant height, wider stem diameter, and more number of leaves as against the highest level of organic fertilizer (40 t ha^{-1}), lower levels of NPK and organic fertilizer combination (75:60:20 kg NPK ha^{-1} + 10 t ha^{-1} of the organic fertilizer), and the absolute control. This may be attributed to more proliferation of root system resulting in more absorption of nutrients and water from the soil leading to production of higher vegetative biomass [12]. Similar findings were reported by [13]. [14] also showed that plant height and number of leaves were higher with higher dose of NPK in comparison with lower doses of NPK at harvest. Increased plant height and number of leaves plant^{-1} with increased levels of N, P and K fertilizers was also reported by [15] in India, and by [16] in Brazil. [17] at Japan had also reported the increased number of leaves plant^{-1} of stevia with higher N nutrition, and [18] reported a positive response to higher levels of K application. [19] showed that the increasing level of N from 60 to 105 kg ha^{-1} could increase height of stevia for about 16.01% (from 48.96 to 56.80 cm). It is

known that sufficient level of N allows to the formation of new cells in plants but insufficient N levels cause small stems in plants [20].

The highest fresh and dry leaf yields in stevia were obtained with higher number of leaves resulted from the higher N, P, and K application. Similar increase in fresh and dry leaf yields of stevia with same NPK combination was showed in earlier research [7]. Full dose of fertilizer application also helped to produce maximum biomass yield [13]. Also, fertilizer dose of 300:150:100 kg NPK ha⁻¹ has been considered as an optimum level of nutrients for stevia in vertisols of Dharwar, Karnataka [15]. Similarly, many reports have shown a positive correlation between NPK fertilization and biomass production [19, 21, 22, 23]. In another study, the highest dry leaf yield was obtained from 275:112.5:172.5 kg NPK ha⁻¹ applications [14]. Whereas, application of 50:60:50 kg NPK ha⁻¹ in western Himalaya region recorded significantly higher dry leaf yield as compared to other treatments [24]. Plants grown at 60 kg N ha⁻¹ produced significantly higher dry leaf yield compared compared to lower nitrogen levels and control [25]. Similarly, [26] concluded that the production of 1 ton of dry leaves of stevia required 64.6 kg N, 7.6 kg P, and 56.1 kg K ha⁻¹. [27] also reported increase in leaf yield with moderate application of N, P, and K fertilizers in Korea. [28] reported that the applications of 90 kg N, 40 kg P, and 40 kg K ha⁻¹ are the best nutritional conditions in terms of dry leaf yield.

In the present investigation, the growth and yield parameters were highest in plants grown at the highest level of organic fertilizer (40 t ha⁻¹) as compared to control plants. It could be due to the increased the nutrient availability and the water holding capacity of the soil by organic matter. Availability of moisture and nutrients during early growth helped to good vegetative growth, which might have enhanced better light utilization resulting in high economic yield [29]. [25] have also reported that the total fresh biomass production of stevia was greatest with 45 ton of farmyard manure ha⁻¹. The trend of increasing the number of leaves per plant

with the application of farmyard manure was also recorded by [30]. [31] showed that plants treated with cow manure were superior in development to control plants.

The higher content of NPK nutrients in stevia leaf at harvest may be caused to the adequate quantity and higher availability of these nutrients in the root zone during plant growth period. [32] in India recorded higher NPK content with higher availability of NPK nutrients. [13] also reported higher K content in stevia leaf at harvest with full dose of fertilizer. These findings are in accordance with the results reported by [33] wherein higher nutrients content in stevia plant was attributed to higher availability of nutrients in the root zone. [25] have also reported that increased supply of N resulted in increased plant N content by stevia. Whereas, [28] reported that applied N, P, and K had little effect in altering the concentration of N, P and, K in stevia plant. The highest level of organic fertilizer (40 t ha^{-1}) recorded the highest N, P, and K concentrations in stevia leaf at harvest than the absolute control; this could be due to the release of NPK nutrients from organic manure. A study on tomatoes (*Lycopersicon esculentum*) and corn (*Zea mays*) in acidic soil by [3] showed that organic manure increases crop productivity, nitrogen utilization efficiency, and soil health compared to chemical fertilizer. Also, [34] found that increased macronutrients in the leaves of rice and maize were due to organic manure application that confirms our results.

The NPK uptake varies positively with NPK content in the leaf and dry leaf production at harvest. Since NPK uptake is the product of dry leaf and NPK content. The uptake of NPK increased probably because it was being used for plant growth. Similar results were reported by [13]. [25] also reported that increased supply of N resulted in increased dry biomass and N uptake by stevia. The results are in accordance with the findings of [32], who reported that the application of $300:150:100 \text{ kg NPK ha}^{-1}$ produced higher dry leaf yield and recorded higher nutrient uptake by stevia plant. In general the NPK uptake was low in all the treatments in second harvest as compared to first harvest. This is ascribing to the small growth of stevia

with lower number of leaves at harvest due to lower temperature and short day lengths prevailed during fall season. The sensitivity of stevia to day length, photoperiod and temperature was also reported by [35] in Morocco and [36] in Egypt. [37] illustrated that the time of harvest is closely related to yield.

The effect of organo-mineral fertilizers has revealed diverse results relative to the plant. [4] have reported that the mixed use of NPK chemical fertilizer and livestock organic manure increases the mean growth of mint (*Mentha arvensis*) and mustard (*Brassica juncea*) by 46%. [38] compared the use of chemical fertilizer treatment only and mixed chemical fertilizer and organic manure treatment in farmland rotating sorghum (*Pennisetum glaucum*) and wheat (*Triticum aestivum*), and found that organic manure increased the soil concentrations of organic carbon and NPK.

Conclusion

Plant nutrients are essential for the production of crops and healthy food for the world's expanding population. Plant nutrients are therefore a vital component of sustainable agriculture. Application of 300:100:240 kg ha⁻¹ NPK combination recorded significantly higher plant height, number of leaves plant⁻¹, and fresh and dry leaf biomass plant⁻¹ as compared to other fertilized treatments and control. This superior combination also resulted in considerably greater contents of nitrogen (N), phosphorus (P), and potassium (K) in stevia leaf. The NPK uptake data showed the same trend as dry leaf yield and NPK content.

Also, our results indicated that all the parameters were influenced by time of harvest. The higher growth, biomass, and mineral nutrition were obtained at the first harvest (85 days after transplanting) compared with the second harvest (56 days after first harvest) in all treatments. Consequently, 300:100:240 kg ha⁻¹ NPK combination level was appeared to be optimal for higher biomass and mineral nutrition of stevia grown during summer season under north-western Moroccan conditions.

References

1. Chen JH. The combined use of chemical and organic fertilizers and/or biofertilizer for crop growth and soil fertility. Proceedings of International Workshop on Sustained Management of the Soil-Rhizosphere System for Efficient Crop Production and Fertilizer Use; 2006.
2. Han SH, An JY, Hwang J, Kim SB, Park BB. The effects of organic manure and chemical fertilizer on the growth and nutrient concentrations of yellow poplar (*Liriodendron tulipifera* Lin.) in a nursery system. Forest Science and Technology. 2016; 12 (3): 137-143.
3. Murmu K, Swain DK, Ghosh BC. Comparative assessment of conventional and organic nutrient management on crop growth and yield and soil fertility in tomato-sweet corn production system. Aust J Crop Sci. 2013; 7 (11):1617-1626.
4. Chand S, Anwar M, Patra DD. Influence of long-term application of organic and inorganic fertilizer to build up soil fertility and nutrient uptake in mint mustard cropping sequence. Commun Soil Sci Plant Anal. 2006; 37(2):63-76.
5. Reis M, Coelho L, Santos G, Kienle U, Beltrão J. Yield response of stevia (*Stevia rebaudiana* Bertoni) to the salinity of irrigation water. Agri Water Manag. 2015; 152: 217-221.
6. Maiti RK, Purohit SS. Stevia: a miracle plant for human health. Agrobios. 2008; 71.
7. Benhmimou A, Ibriz M, Douaik A, Lage M, Al Faiz C, Chaouqi S, Zouahri A. Effect of NPK Fertilization on the Growth, Yield, Quality and Mineral Nutrition of New Sweet Plant in Morocco (*Stevia rebaudiana* Bertoni). American Journal of Biology and Life Sciences. 2018; 6 (3): 36-43.
8. Samsudin A, Aziz IA. Drying of stevia leaves using laboratory and pilot scale dryers. J. Trop. Agric. Food. Sci. 2013; 41: 137-147.
9. Pomeranz Y, Clifton ME. Food analysis: Theory and practice. 2nd edition Van Nostrand Reinold. New-York. 1987: 797.
10. Olsen SR. Estimation of available phosphorous in soils by extraction with sodium bicarbonate. Cir. U.S. Dep. Agr. 1954; 939: 1-19.
11. Van Rast E, Verloo M, Demeyer A, Pauwels JM. Manual for the Soil Chemistry and Fertility Laboratory; 1999.
12. Harrier LA, Sawczak J. Detection of the 3-phosphoglycerate kinase protein of *Glomus mosseae*. Mycorrhiza. 2000; 10: 81-86.
13. Behera MS, Verma OP, Mahapatra PK, Singandhupe RB, Kumar A, Kannan K *et al.* Effect of fertigation on stevia (*Stevia rebaudiana*) under drip irrigation. Indian Journal of Agronomy. 2013; 58 (2): 243-250.
14. Nevase PV, Bafna AM, Shinde KA. Effect of N, P, K and FYM on growth and TSS of stevia. Crop Res. 2011; 42: 131-135.
15. Aladakatti YR, Palled YB, Chetti MB, Halikatti SI, Alagundagi SC, Patil PL *et al.* Effect of nitrogen, phosphorus and potassium levels on growth and yield of stevia (*Stevia rebaudiana* Bertoni). Karnataka. J. Agric. Sci. 2012; 25 (1): 25-29.
16. Buana L, Goenadi DH. A study on the correlation between growth and yield of stevia. Menara Perkebunan. 1985; 53:68-71.
17. Kawatani T, Kaneki Y, Tanabe T. On the cultivation of Kaa-hee *Stevia rebaudiana* (Bert). Japanese. J. Tropical Agri. 1977; 20: 137-142.
18. Kawatani T, Kaneki Y, Tanabe T, Takahashi T. On cultivation of Kaa-He-E (*Stevia rebaudiana* Bert).VI. Response of stevia to potassium fertilization rates and to the three major elements of fertilizer. Nettai Nogyo. 1980; 24: 105-112.
19. Maheshwar HM. Effect of different levels of nitrogen and gates of planting on growth and yield of stevia (*Stevia rebaudiana* Bert.) [MSc thesis]. Dharwad: Department of Horticulture College of Agriculture, Dharwad University of Agricultural Sciences; 2005.
20. Lavres J, Dos Santos JDG, Monteiro FA.. Nitrate reductase activity and spad readings in leaf tissues of guinea grass submitted to Nitrogen and potassium rates. Revista Brasileira De Ciencia Do Solo. 2010; 34 (3): 801-9.
21. Chalapathi MV, Thimmegowda S, Rao GGE, Devakumar N, Chandraprakash J. Influence of fertilizer level on growth, yield and nutrient uptake of ratoon crop of stevia (*Stevia rebaudiana*). J. Med. Aromatic Plant Sci. 1999; 21: 947-949.
22. Uçar E, Turgut K. The effect of different nitrogen levels on yield and quality of stevia (*Stevia rebaudiana* bert.). JOURNAL OF PLANT NUTRITION. 2018; 41 (9): 1130-1137.
23. Tavarini S, Pagano I, Guidi L, Angelini LG. Impact of nitrogen supply on growth, steviol glycosides and photosynthesis in *Stevia rebaudiana* Bertoni. Plant Biosystems. 2015; 150 (5):953-62.

24. Kumar R, Sharma S, Ramesh K, Prasad R, Pathania V, Singh B, Singh RD. Effect of agrotechniques on the performance of natural sweetener plant-stevia (*Stevia rebaudiana* Bertoni) under western Himalayan conditions. *Ind. J. Agron.* 2012; 57: 74-81.
25. Rashid Z, Rashid M, Inamullah S, Rasool S, Ah Bahar F. Effect of different levels of farmyard manure and nitrogen on the yield and nitrogen uptake by stevia (*Stevia rebaudiana* Bertoni). *Afr. J. Agric. Res.* 2013; 8: 3941-3945.
26. Lima FOF, Malavolta E. Nutritional interactions in stevia (*Stevia rebaudiana* Bert). Bertoni.). *Arquivos de Biologia e Tecnologia Curitiba.* 1997; 40: 351-357.
27. Lee JI, Kang KH, Park HW, Ham YS, Park CH. Studies on new sweetening source plant stevia (*Stevia rebaudiana*) in Korea. II. Effects of fertilizer rates and planting density on dry leaf yields and various agronomic characteristics of *Stevia rebaudiana*. *Research Reports of the office of Rural Development (Crop Suwon).* 1980; 22 138-144.
28. Pal PK, Kumar R, Guleria V, Mahajan M, Prasad M, Pathania V *et al.* Crop-ecology and nutritional variability influence growth and secondary metabolites of *Stevia rebaudiana* Bertoni. *BMC Plant Biol.* 2015; 15: 67.
29. Krishnamurthy K. Stevia the herb sweetener than sugar. *Kisan World.* 2001; 28(10): 31-32.
30. Goenadi DH. Effect of FYM, NPK and liquid fertilizers on *Stevia rebaudiana*. *Menara Perkebunan.* 1985; 53:23-30.
31. Hoseini RZ, Goltapeh EM, Kalatejari S. Effect of bio-fertilizer on growth, development and nutrient content (leaf and soil) of *Stevia rebaudiana* Bertoni. *J. Crop Prot.* 2015; 4: 691-704.
32. Aladakatti YR. Response of stevia (*Stevia rebaudiana* Bertoni.) to irrigation schedule, planting geometry and nutrient levels. *Doctoral Thesis., Univ. of Agric. Sci., Dept of Agron., Dharwad, Bangladesh;* 2011.
33. Angkapradipta P, Warsito T, Faturachim P. The N, P, and K requirements of *Stevia rebaudiana* on latosolic soil. *Menara Perkebunan.* 1986; 54: 1-6.
34. Tejada M, Gonzalez J. Effect of foliar application of beet vinasse on maize yield. *Biological Agriculture and Horticulture.* 2006; 24: 197-214.
35. Benhmimou A, Ibriz M, Al Faiz C, Gaboun F, Douaik A, Amchra FZ *et al.* Effects of Planting Density and Harvesting Time on Productivity of Natural Sweetener Plant (*Stevia rebaudiana* Bertoni.) in Larache Region, Morocco. *International Journal of Plant Research.* 2017; 7(4): 83-89.
36. Allam AI, Nassar AM, Besheit SY. Nitrogen fertilizer requirements of *Stevia rebaudiana* Bertoni under Egyptian condition. *Egyptian J. Agric. Res.* 2001; 79: 1005-1018.
37. Ramesh K, Singh V, Ahuja PS. Production potential of *Stevia rebaudiana* (Bert.) Bertoni under intercropping systems. *Arch. Agron. soil Sci.* 2007; 53(4): 443-58.
38. Kaur K, Kapoor KK, Gupta AP. Impact of organic manure with and without mineral fertilizers on soil chemical biological properties under tropical conditions. *J Plant Nutr Soil Sci.* 2005; 168 (1):117-122.