International Journal of
Environmental Sciences & Natural Resources
ISSN: 2572-1119

Juniper PUBLISHERS key to the Researchers

Research Article
Volume 22 Issue 3 - November 2019
DOI: 10.19080/IJESNR.2019.22.556090

Int J Environ Sci Nat Res

The Response of Stevia (*Stevia rebaudiana L.*) to Nitrogen and Phosphorous Fertilizer Rates at Menagesha, West Ethiopia

Nebret Tadesse*

Wondo Genet Agricultural Research Center, Ethiopia

Submission: October 04, 2019; **Published:** November 14, 2019

***Corresponding author:** Nebret Tadesse, Wondo Genet Agricultural Research Center, EIAR, P.O. Box: 198, Shashemene, Ethiopia

Abstract

A field trial was conducted at Menagesha Cheshire Rehabilitation center to investigate the effect of nitrogen and phosphorus application on growth and yield of stevia during 2014/15 and 2015/16 cropping season in supplementary irrigation. The treatments consisted of four levels of N (0, 23, 46, and 96kg ha-1) and four levels of P (0, 10, 20 and 30kg ha^{-1}) arranged in Randomized Complete Block Design with three replications. The main effect of nitrogen had a significant influence on leaf and above-ground biomass yield of Stevia. The main effect of phosphorous had no significant effect on all of the parameters. The interaction effect of nitrogen and phosphorus were significantly affected number branch per plant, fresh leaf weight and above-ground biomass yield of Stevia. Significantly higher fresh leaf yield (17.18t ha^{-1}) was obtained from the combined application of 23/30NPkg $ha^{-1}$and 46/0NPkg ha^{-1}. The maximum above-ground biomass (28.3t ha^{-1}) was recorded from the combined application of 23/30NP kg ha^{-1}. In conclusion, the results revealed that nitrogen and phosphorous at moderate rates led to a significant increment in growth components and leaf yield of Stevia. Thus, be realized that the application of nitrogen and phosphorous noticeably increases the production of Stevia in the area.

Keywords: Stevia; Nitrogen; Phosphorous; Growth and Stevia leaf yield

Information

Stevia rebaudiana Bertoni, known as the "sweetest plant of the world", belongs to the family Asteracea and is native to the South American center of diversity. The world of sweetness has seen a sweeter change in the recent past with the introduction of stevia sugar obtained from leaves of stevia containing compounds about 250 to 300 times sweeter than the table sugar [1]. The substance of interest is stevioside, rebaudioside - A and at least six other compounds that have glucosidal groups attached to a three-carbon ring central structure. Stevioside could be equivalent to the sweetening power of 28 tons per acre of sucrose sugar [2]. Though these sugars have sweetening qualities, they have been found to contribute to calories and are not advised for the consumption by diabetic patients.

Growth, yield, and quality of Stevia are not affected by genetic factors, but also environment and one of them is nutrient availability for the plant. The main nutrients required by the plant were nitrogen and phosphorous. Nitrogen is an essential nutrient that could improve growth during the vegetative phase and protein synthesis. According to Hardjowigeno [3] nitrogen is applied in the plant, which is going to be taken the leaves, because it would make the foliar grow well. Sufficient nitrogen in the soil would make the plants look greener, which means that nitrogen plays its role in forming chlorophyll for the photosynthetic process. Phosphorus is needed for normal growth and development of the plants due to its vital role in chlorophyll synthesis and involvement in various physiological and metabolic processes of the plant. Based on this information, it is presumed that the production of Stevia in Ethiopia is very important however there is lack of cultivation technology on Stevia, therefore this trial aimed to improve Stevia productivity.

Materials and Methods

The experimental site, design, and treatments

The experiment was conducted at Menagesha Cheshire Rehabilitation center, Ethiopia. Menagesha Rehabilitation Center-Cheshire is located 20km west of Addis Ababa at 09°03′N latitude and 38°34′ 60′′E longitude with an altitude of 2812masl. The site receives a mean annual rainfall of 1056mm with minimum and maximum temperatures of 6 ^{0}C and 22 ^{0}C, respectively. The soil is clay with an average pH of 5.4. Experimental plots were established in October, 2014/15. The experiment comprised a factorial combination of four levels of N (0, 23, 46, and 96kg ha^{-1}) and four levels of P (0, 10, 20 and 30kg ha^{-1}) arranged in Randomized Complete Block Design (RCBD) with three replications. The sources of N and P were urea (46% N) and triple superphosphate (TSP)

(46% P_2O_5), respectively. Stevia (*rebaudiana* Bertoni) was used as a test crop. For seedling preparation, soft stem cutting of 15cm length was taken from one-year-old disease-free Stevia mother plants maintained at Wondo Genet Agricultural Research Center botanical garden. Seedlings were raised in the nursery for three months in polyethylene pots. Transplanting was done in October 2015. A spacing of inter-row 50cm and intra-row 30cm were used for transplanting. Nitrogen fertilizer was applied in three equal splits in each year's (following the treatment), 1/3 during transplanting, and the remaining N was applied on top dressed in two equal splits at subsequent ratoon plant immediately after the first and second harvest done. The full dose of phosphorus fertilizer was applied at transplanting following the treatment. This was followed and maintained for each cutting throughout the crop season during both the years. First cycle harvesting was done four months after transplanting; the plants were cut uniformly 15cm above the ground level whereas the succeeding crops were harvested at a regular interval of three months for two consecutive years. All pertinent data were collected from net plot five selected plants excluding plants from either end of the rows.

Soil analysis

Soil samples were collected from a depth of 30cm before planting at random, and one complete composite sample was prepared to determine the particle size distribution and chemical properties of the soil. Soil texture was determined by Bouyoucos hydrometer method [4], Organic matter determined based on the oxidation of organic carbon with acid dichromate medium following the Walkley and Black method And total N determined by Kjeldahl method [5] and available soil P determined using Olsen & Dean [6] method. Soil pH determined in 1:2.5 soil: water ratio using a glass electrode attached to a digital pH meter

Data on plant height, number of branches per plant, were recorded from each harvesting cycle and each year and the average value of year was used for analysis. In addition to this, the cumulative value of fresh leaf weight, fresh aboveground biomass, and dry leaf weight in kg ha^{-1} was taken three times each year and the values summed up and the average value of two years was used for analysis.

Data analysis

All data were subjected to analysis of variance (ANOVA) using Statistical Analysis Software [7]. The difference among significant treatment means was tested using the least significant difference (LSD) at 5% level of significance.

Result and Discussion

Major physico-chemical properties of the soil

The result of the physical and chemical analysis of experimental soil revealed that the textural class of the surface soil (0-30cm) was clay with a particle size distribution of 14% sand, 28% silt and 58% clay. According to Tekalign [8] and Landon [9], the chemical analysis showed that the soil was moderately acidic with relatively low nitrogen, low organic matter, and low organic carbon and high available phosphorus. The available P, total N and organic carbon of the soil were 24mg kg^{-1}, 0.12%, and 2.3% respectively. Such soil often responds to P and N application according to Amar [10] and Landon [9] respectively (Table 1).

Table 1: Pre- transplanting chemical properties of soil (0-30cm depth).

Property	Values/Type
Sand (%)	14
Silt (%)	28
Clay (%)	58
Texture class	Clay
pH	5.41
CEC micros/m	27.6
Total Nitrogen (%)	0.12
Organic carbon (%)	2.3
Available Phosphorous (mg kg-1)	24
C: N ratio	15.2

Plant height and Number of primary branches per plant

Table 2: Pre- transplanting chemical properties of soil (0-30cm depth).

Source of Variation	DF	PH	NBPP	FLW (t ha^{-1})	DLW (t ha^{-1})	FAGB (t ha^{-1})
Replication	2	0.11	2.31	6680777	560003.6	13275052
Nitrogen	3	2.45ns	34.97ns	13765279.7*	2169765.7*	32976995.3*
Phosphorous	3	7.81ns	13.92ns	2671301.6ns	564407.4ns	19712119.4ns
N x P	9	9.71ns	54.57*	11437225.3*	622186.54ns	27809165.8*
Error	30	4.96	13.6	4903090	298157.2	8300903
CV%		7.04	9.8	14.6	13.2	12.92

*Significant at p<0.05, ns=Not Significant at p<0.05, Ph = Plant height, NBPP= Number of branches per plant, FLW = fresh leaf weight, FAGB =Fresh aboveground biomass weight, and DLW= Dry leaf weight.

The main effect, as well as the interaction effect of N and P, did not show a significant difference (P>0.05) on the plant height of Stevia (Table 2). The interaction effect of nitrogen and phosphorous on the number of primary branches per plant were significant (P<0.05); however, the main effect of nitrogen and Phosphorous didn't affect the number of primary branch per plant (Table 2). Maximum number of primary branch per plant (44 $plant^{-1}$) was obtained in a response of combined application of 69/20 NP kg ha^{-1} which was statistically parity with number of primary branch per plant (39),(39),(42),(41) & (42) obtained at the combined

How to cite this article: Nebret Tadesse. The Response of Stevia (*Stevia rebaudiana L.*) to Nitrogen and Phosphorous Fertilizer Rates at Menagesha, West Ethiopia. Int J Environ Sci Nat Res. 2019; 22(3): 556090. DOI: 10.19080/IJESNR.2019.22.556090

application of 23/10NP kg ha^{-1}, 23/20 NPkgha^{-1}, 23/30 NPkgha^{-1}, 46/0 NPkgha^{-1} , 46/10 NPkgha^{-1} respectively, followed by the combined application of 0kg ha^{-1}+30 Pkg ha^{-1}, 69 Nkgha^{-1}+10Pkg ha^{-1} , 69Nkgha^{-1}+30Pkgha^{-1}.

Table 3: Main effects of nitrogen and phosphorous on average plant height and Dry leaf weight of Stevia over the 2014/15 and 2015/16 cropping season.

Treatments	Plant Height (cm)	Dry leaf weight (kg ha^{-1})
Nitrogen (kg ha^{-1})		
0	31.4	3548b
23	32.2	4511a
46	31.2	4115a
69	31.7	4371a
LSD	NS	455.3
Phosphorous (kg ha^{-1})		
0	30.4	3844
10	31.9	4244
20	32	4113
30	32.2	4344
LSD 5%	NS	455.3
Nitrogen * phosphorous	NS	NS
CV%	7.04	13.2

Means followed by the same letter within a column are statistically non-significant at p≤0.05 probability level; CM = Cent meter; CV=Coefficient of Variance; LSD= Least Significant Difference.

Increasing nitrogen and phosphorus fertilizer on the Stevia plant enhance the number of primary branch per plant over the control. This could be an adequate supply of nitrogen is associated with vigorous vegetative growth and more efficient use of other nutrients. The results are in accordance with the findings of Aladakatti et al. [11], who reported the increased number of branches and leaves plant^{-1} of Stevia with higher nitrogen and phosphorus. Chalapathi et al. [12], who also reported an increased number of branches plant^{-1} with nutrient levels of 40:30:45 kg NPK ha^{-1} in sandy loam soils at Bangalore.

Fresh leaf weight (t ha^{-1}) and dry leaf weight (t ha^{-1})

The application of nitrogen was significantly (P<0.05) affected fresh leaf weight; while no significant variation in fresh leaf weight was observed due to the application of phosphorus. The interaction effect of nitrogen and phosphorus was also found to be significant on fresh leaf weight (Table 2). Although the results were inconsistent, the interaction of nitrogen with phosphorous significantly influenced the fresh leaf weight of stevia. Maximum fresh leaf weight (19.3t ha^{-1}) was recorded from the combined application of 69/20kg ha^{-1} which was in statistical parity with the fresh leaf weight obtained in response to the combined application of 23/10, 23/20, 23/30, 46/0 and 69/10NP kg ha^{-1}. Increasing the rates of nitrogen rates from 0 to 23N kg ha-1 with phosphorous rates resulted in higher fresh leaf yield; beyond that showed decreasing trend except for the combined application of 69/10 and 69/20NP kg ha^{-1}. Increasing nitrogen and phosphorous rates from 0 to 23/30NP, and 69/20NP kg ha^{-1} resulted in about 39.3%, and 56.4% additional increased in fresh leaf weight. The significant increase in fresh leaf weight in response to the increased rates of nitrogen and phosphorous application might be ascribed to the increased availability of nitrogen in the soil for uptake by plant roots, which may have sufficiently enhanced vegetative growth through increasing cell division and elongation. The results are in accordance with the findings of Aladakatti et al. [11], who reported that the highest fresh leaf weight of Stevia with the application of 400N kg ha^{-1} with 200Pkg ha $^{-1}$ in medium black, clay soil, India. Inugraha et al. [13] at Malang also reported applying 200N kg ha^{-1} with 225kg K_2O ha^{-1} obtained higher fresh leaf yield of Stevia.

Table 4: Average number of branch per plant, Fresh leaf weight and Fresh Above ground-biomass of Stevia as affected by the interaction effect of nitrogen and phosphorous over the 2014/15 and 2015 /16 cropping season.

Nitrogen (kg ha^{-1})	Phosphorous (kg ha^{-1})	Number of Branch per plant	Fresh Leaf Weight (t ha^{-1})	Fresh Above Ground Biomass (t ha^{-1})
0	0	35c-f	12.33cd	17.95e
	10	33ef	13.97bcd	17.90e
	20	33ef	14.27bcd	19.06e
	30	38b-f	14.53bcd	21.71cde
23	0	33ef	14.06bcd	19.34de
	10	39a-d	15.91abc	24.69abc
	20	39a-d	15.94abc	23.37bcd
	30	42ab	17.18 ab	28.34a
46	0	41abc	17.19 ab	24.73abc
	10	42ab	15.43 cd	22.68b-e
	20	32f	11.91 d	21.78cde
	30	35c-f	15.85 bc	22.66b-e
69	0	35c-f	14.57cd	20.75cde
	10	38b-f	16.15 ab	23.36bcd

How to cite this article: Nebret Tadesse. The Response of Stevia (*Stevia rebaudiana L.*) to Nitrogen and Phosphorous Fertilizer Rates at Menagesha, West Ethiopia. Int J Environ Sci Nat Res. 2019; 22(3): 556090. DOI: 10.19080/IJESNR.2019.22.556090

	20	44a	19.29 a	27.20ab
	30	38b-f	14.92bcd	21.36cde
LSD0.05		6.15	3.69	4.8
CV%		9.8	12.9	14.6

Means followed by the same letter within a column are statistically non-significant at p≤0.05 probability level; CV=Coefficient of Variance; LSD= Least Significant Difference.

Applying phosphorous rates with absences of nitrogen resulted in lower fresh leaf weight; this result confirmed that nitrogen fertilizer responded to the crop due to low nitrogen nutrients found in the soil (Table 1 & 4).

Significant differences were observed due to the main effect of nitrogen on the dry leaf weight of Stevia. However, the main effect of phosphorous, as well as its interaction with nitrogen, did not affect dry leaf weight. Increasing the rate of nitrogen application from 0 to 23kg N ha^{-1} resulted in about a 27.1% additional increase in the dry leaf yield. However, the dry leaf yields obtained at 23, 46 and 69 kg N ha^{-1} were in statistical parity (Table 3). Confirming the results of this study, Rashid et al. [14] reported that dry leaf yield increased significantly at 60 kg N ha^{-1} compared to the control treatment. Conduct experiment at Egypt also showed a significant increase in dry leaf biomass yields of Stevia when nitrogen fertilizer was increased from 10 to 30kg N ha^{-1} wherein the dry leaves yield increased by 64 percent compared to a lower dose [15]. This result is in contradictory with that of Aladakatti et al. [11] who reported that dry leaf yield increased significantly from 9.5 to 11.42t ha^{-1} due to increased nitrogen application from 200 to 400kg N ha^{-1}. Increased dry leaf yield was also reported by Shock [2] in Japan with a moderate application of nitrogen, phosphorus, and potassium.

Fresh above-ground biomass (t ha^{-1})

Fresh aboveground biomass was significantly (P<0.5) influenced by the main effect of nitrogen and its interaction with phosphorous. However, the main effect of phosphorous didn't affect fresh above-ground biomass weight (Table 2). Significantly maximum fresh above-ground biomass (28.34t ha^{-1}) was obtained with the combined application of 23/30NP kg ha^{-1}, which was in statistical parity with the fresh above-ground biomass obtained in response to the combined application of 23/10, 46/0, 69/20NP kg ha^{-1}. This could be attributable to the significantly maximum number of branches per plant and fresh leaf weight obtained that contribute to the higher fresh above-ground biomass weight. The lower fresh above-ground biomass weight was recorded at nil rates of NP kg ha^{-1} and 0N kg ha^{-1} combined with all rates of phosphorous application as well as at the combined application rates of 23/0,46/10, 46/20,46/30, 69/0, 69/30NP kg ha^{-1}. Increasing rates of nitrogen from 23 to 46 and 69N kg ha^{-1} the result was not constant. With increasing nitrogen application rates, the fresh aboveground biomass increased, and this increase was observed only at the combined application of 46/0NP kg ha^{-1} and with the combined application of 69/20NP kg ha^{-1} the results were inconsistent. The higher rate of nitrogen might have resulted in more vigorous plant growth, particularly information phase or vegetative growth, such as stem, and leaf. A similar result was reported by Aladakatti et al. [11] increased fresh above-ground biomass with the nutrient level of 400N kg ha^{-1} and 200Pkg ha^{-1}. Maheshwar [16] also reported that the application of 105:30:45kg NPK/ha recorded significantly higher above-ground biomass due to a maximum number of leaves per plant and branches per plant as compared to lower doses of nitrogen and phosphorous (Figure 1).

Figure 1: Field status during experimental period.

How to cite this article: Nebret Tadesse. The Response of Stevia (*Stevia rebaudiana L.*) to Nitrogen and Phosphorous Fertilizer Rates at Menagesha, West Ethiopia. Int J Environ Sci Nat Res. 2019; 22(3): 556090. DOI: 10.19080/IJESNR.2019.22.556090

Conclusion

It could be concluded that the two consecutive study years result showed that nitrogen and phosphorus application significantly increased growth and biomass yield of Stevia. The optimum economic fresh leaf yield (19.29t ha-1) and dry leaf yield (4511kg ha^{-1}) were obtained in response to the combined application of 23N kg ha^{-1} with 10Pkg ha^{-1}. Therefore; around Menagesha and similar agroecology, soil nutrients condition should apply 23kg ha^{-1} nitrogen + 10kg ha^{-1} phosphorus to optimize the economic yield of Stevia.

Acknowledgment

We would like to acknowledge Land and Water Resource Research Directorate and Wondo Genet Agricultural Research Center for providing all the necessary facilities and supports during the entire experimentation. Our sincere appreciation goes to Belist Lulie Begna Bekele, Abush Yoseph, Zerihun Jonba and, for their help in field and laboratory works.

References

1. Kumar VR (2002) Medicinal and Aromatic Plants. Green Cross Media International, Bangalore. pp. 90-91.
2. Shock CC (1982) Rebaudi's natural non-caloric sweeteners. Cal Agric 36: 4-5.
3. Hardjowigeno S (2010) Soil science. Akademika Pressindo, 7th copies, Jakart.
4. Day PR (1965) Hydrometer method of particle size analysis. In: Black CA (Ed.), Methods of soil analysis. American Society of Agronomy, Wisconsin Argon 9(2): 562-563.
5. Dewis J, Freitas P (1970) Physical and chemical methods of soil and water analysis. FAO Bull. No.10, Rome, p. 271.
6. Olsen SR, Dean LA (1965) Phosphorus. Methods of soil analysis. Agronomy No 9. American Society of Agronomy, Madison, USA.
7. SAS (2002) Version 9.0 Institute inc., Cary, NC, USA.
8. Tekalign Tadesse (1991) Soil, plant, water, fertilizer, animal manure and compost analysis working document. No 13. International Livestock Research Center for Africa (ILCA). Addis Ababa.
9. Landon JR (1991) Booker Tropical Soil Manual. A handbook for soil survey and agricultural land evolution in the tropics and subtropics. Longman Scientific and Technical. Essex, New York, p. 474.
10. Amar B (1999) Diagnosis and prescription of phosphate fertilization within the Mediterranean region countries. In: African Crop Science Society Proceedings. Casablanca, 10-14 October 1999, pp. 99-106.
11. Aladakatti YR, Palled YB, Chetti MB, Halikatti SI, Alagundagi SC, et al. (2012) Effect of nitrogen, phosphorus and potassium levels on growth and yield of stevia (Stevia rebaudiana Bertoni). Karnataka. J Agric Sci 25(1): 25-29.
12. Chalapathi MV, Kumar ND, Rao GGE, Jayaramaiah R (1999) Comparative performance of plant and ratoon crops of stevia. Indian Agril 43: 211-212.
13. Inugraha, Maghfoer MD, Widaryanto E (2014) The response of Stevia (*Stevia rebaudiana* Bertoni M) To Nitrogen and Potassium Fertilization. J Agric and Vet Sci 7(10): 47-55.
14. Rashid Z, Rashid M, Inamullah S, Rasool S, Bahar F Ah (2013) Effect of different levels of farmyard manure and nitrogen on the yield and nitrogen uptake by stevia (*Stevia rebaudiana* Bertoni) Punjab, India. Afr J Agric Res 8(29): 3941-3945.
15. Allam AI, Nassar AM, Besheit SY (2001).
16. Maheshwar HM (2005) Effect of different levels of nitrogen and dates of planting on growth and yield of stevia (*Stevia rebaudiana*). M.Sc Thesis. Department of horticulture, University of agricultural sciences, Dharwad, Karnataka, India, p. 100.

How to cite this article: Nebret Tadesse. The Response of Stevia (*Stevia rebaudiana L.*) to Nitrogen and Phosphorous Fertilizer Rates at Menagesha, West Ethiopia. Int J Environ Sci Nat Res. 2019; 22(3): 556090. DOI: 10.19080/IJESNR.2019.22.556090