

***Performance of Stevia (Stevia Rebaudiana Bertoni)
For Morphological And Economic Traits Under
Different Ecologies of Ethiopia***

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ABSTRACT

Stevia [Stevia rebaudiana Bertoni] is a perennial, calorie free natural sweet herb that belongs to the family Asteraceae. Its fresh leaves and leaf extracts are 15-30 and 250-330 times sweeter than common sugar, respectively. Despite its amazing characteristics, the plant has not been evaluated for its performance in morphological and economic traits in different ecologies of Ethiopia. For assessing the adaptability of the plant in different ecologies, data on morphological and chemical traits were collected from four locations for two years and summarized on mean basis. The performance of *Stevia* varied across the testing locations for all the studied parameters. In the first year, the highest fresh leaf yield of 2332 kg/ha and dry leaf yield 600 kg/ha was recorded at Chench, and the respective lowest values of 992 and 225 kg were recorded at Awada. In the second year, the respective highest value of 3034 kg and lowest 1891 kg fresh leaf yield/ha was recorded at Wondo Genet and Debre Zeit. Whereas, the highest dry leaf yield of 769Kg/ha was at Awada and lowest yield of 541 kg/ha at Chench. The respective percent increase values of 4.5, 14.7 and 45.2% were observed in second year compared to the first year for plant height, fresh leaf yield/ha and dry leaf

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yield/ha. In the second year, number of branches/plant, fresh leaf yield/plant and dry leaf yield/plant had decreased by 46.74, 99.94 and 34.38%, respectively. The stevioside content varied from 9.07-10.35%, which is within the ranges of world reports. Therefore, Stevia can be cultivated for its intense sweet leaves and stevioside in selected areas of the country.

Key words: leaf yield, Stevia, stevioside content

Running title: Adaptability of Stevia in different places of Ethiopia.
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INTRODUCTION

Stevia (*Stevia rebaudiana* L. Bertoni) is a perennial herb that belongs to the family Asteraceae (Ahmed *et al.*, 2007; Ojha *et al.*, 2010). It is native to South and Central America (Debnath, 2008; Sumon *et al.*, 2008; Jackson *et al.*, 2009). The genus *Stevia* contains more than 154 species and the most widely utilized ones are *Stevia eupatoria*, *S. ovata*, *S. plummerae*, *S. salicifolia*, *S. serrata* and *S. rebaudiana* (Soejarto *et al.*, 1983; Jackson *et al.*, 2009). However, *S. rebaudiana* is known for its significant sweetening property (Ingle and Venugopal, 2009). The first commercial cultivation of Stevia was started in Paraguay around 1964 (Sumida, 1968; Katayama *et al.*, 1976). Currently it is cultivated in Japan, Taiwan, Philippines, Hawaii, Malaysia and overall South America for food and pharmaceutical products (Ahmed *et al.*, 2007; Debnath, 2008; Sumon *et al.*, 2008).

The property of the species that called attention to the plant was the intense sweet tests of the leaves and aqueous extracts (Ahmed *et al.*, 2007; Jackson *et al.*, 2009). Dry leaves of Stevia are 15 to 30 times sweeter than table sugar with zero calories (Ojha *et al.*, 2010). The leaves of stevia contain

sweetening compounds namely stevioside, rebaudioside A, rebaudioside B and rebaudioside C and six other compounds which have insulin balancing properties (Moraes and Machado, 2001). These sweeteners impart 250 times sweetness than table sugar and 300 times more than sucrose (Ingle and Venugopal, 2009). These glycosides are extracted from the Stevia leaf as all-natural zero caloric sweeteners (Jackson *et al*, 2009); hence, Stevia is a calorie-free bio-sweetener of high quality with non-fermentable and non-discoloring qualities that maintains heat stability at 100°C and has a long shelf life (Brandle, 1999).

The product has been added to tea and coffee, cooked or baked goods, processed foods, beverages (Ahmed *et al*, 2007), used safely in herbal medicines, tonics, for diabetes and in the daily usage products like mouthwashes and toothpastes (Sumida, 1980). It can be used in chocolates and candies not only to meet the market demand by the diabetes, but also to harvest the added advantages of this herb that it does not encourage tooth decay due to its anti-microbial property, unlike sugar (Sumida, 1980). In the Pacific Rim countries like China, Korea and Japan, Stevia is regularly used in preparation of food and pharmaceutical products and currently Stevia production is centered in China with major market in Japan (Lewis, 1992). No negative clinical reports have papered in any of the countries where Stevia is readily available (Brandle and Rosa, 1992).

The present scenario is that people are more inclined towards products advertised with a brand name "all natural and low carbohydrate" (Nishyama *et al*, 1991). Hence, Stevia will also have wider potential utilizations. Apart from this, Stevia is nutrient-rich, containing substantial amount of protein, calcium and phosphorous (Brandle, 1999). Basing the potential uses of Stevia, there are a number of entrepreneurs showing interest to cultivate it in

a considerably large scale for herbal production in Ethiopia for export to European market (EIAR, 2009).

Nonetheless, there is little research and information available to support the interest and to stimulate the potential for local and national economic roles of the species in Ethiopia. This study is, therefore, designed to contribute to the knowledge of stevia by addressing the existing technology gaps and bringing the crop for cultivation and utilization. Thus, the objective of the study is to test the morphological and economic traits of Stevia under different ecologies of Ethiopia.

MATERIALS AND METHODS

The study was conducted in Oromia and SNNPRS regions of Ethiopia at Wondo Genet, Awada, Chenchu and Debrezeit sites for two years from 2010/2011 to 2011/2012. The ecological descriptions of the testing locations are summarized in table I.

Experimental setup: Soft top stem cuttings of Stevia with 5 nodes were taken from 6 month old disease free mother plants from Wondo Genet Agricultural Research Center botanical garden for seedling preparation. Seedlings were raised in the nursery for three months in polyethylene bags before being transplanted to the field experimental plots. At each location, the seedlings were transplanted on an area of 36 m (6 m length and width). A spacing of 50 cm was maintained between plants and rows. During experimentation, two hoeing and three weeding were done prior to each harvesting. In dry seasons, irrigation was applied weekly to its field capacity. Harvesting was made at the time when the plants start to produce flowering buds (at the time of flower bud initiation).

Table1. Summary of site descriptions used for testing the adaptability testing of Stevia (*Stevia rebaudiana* Bertoni) for morphological and economic traits in Ethiopia

Locations	Latitude	Longitude	Altitude (m.s.s.l)	Soil pH	Soil type	MAR (mm)	Annual average temperature (°C)	
							Mini mum	Maximum
Wondo Genet	7°19' N	38°38' E	1876	6.4	Sandy clay loam (Nitosol)	1000	12.0	26.72
Chencha	6°13' N	37°34' E	2618	4.9	Mollic nitisol	873	6.0	16.3
Awada	6°03' N	38°10' E	1750	5.9	Clay loam (Nitosol)	1267.2	11.0	28.4
Deberezeit	8°44' N	38°58' E	1891	6.9	Black heavy clay (Vertisol)	851	12.2	25.72

Data collection and stevioside content analyses: Data on plant height, number of branches/plant, fresh leaf yield/plant, dry leaf yield/plant, fresh leaf yield/ha and dry leaf yield/ha were collected separately for two years and presented on mean values for comparing the adaptability of the plant over the different testing locations. Data on percent stevioside content was collected following laboratory analysis. Percent stevioside content was determined on dry weight (w/w) basis from 650 g of dry composite leaves harvested from the middle row plants of a plot. Laboratory analysis was done at Ethiopian Health and Nutrition Research Institute (EHNRI) using High Pressure Liquid Chromatography (HPLC) instrument.

RESULTS AND DISCUSSION

Mean values from first year and second year data for six traits of *Stevia* tested over four locations of Ethiopia are summarized in table 2. Performance difference was observed on plant height, number of branches/plant, fresh leaf yield/plant, dry leaf yield/plant, fresh leaf yield/ha and dry leaf yield/ha over the testing locations and years. This indicates that these traits are influenced by environmental and plant age factors. Stevioside content was also varied among the testing locations indicating the performance inconsistency of the plant due to the variation of the plant. Hence identification of suitable growing locations is very important before planning for large scale cultivation of the plant.

Table 2. Mean performance of morphological and economic traits of *Stevia rebaudiana* Bertoni) tested over four locations during the years 2010/2011 and 2011/2012.

Locations	Plant height (cm)	Number of branches/plant	Fresh leaf yield/plant (g)	Fresh leaf yield/ha (kg)	Dry leaf yield/plant (g)	Dry leaf yield/ha (kg)	Percentage of Stevia content
Year I (2010/2011)							
Chencha	31.37	24.62	24.99	2332.01	10.74	599.57	-
Wondo genet	34.62	9.18	34.04	1748.05	11.92	429.80	-
Awada	31.03	11.78	41.52	992.50	5.63	225.18	-
Debrezeit	31.97	34.20	31.60	1891.30	14.98	477.03	-
Over all mean	32.25	19.94	33.04	1740.96	10.82	432.89	-
Year II (2011/2012)							
Chencha	31.25	14.70	29.35	2608.90	6.08	540.50	13.35
Wondo genet	42.30	14.40	34.13	3034.20	7.09	630.31	9.07
Awada	41.45	4.80	23.91	2155.30	8.65	768.97	10.15
Debrezeit	19.82	8.60	21.28	1891.00	6.58	585.26	-
Over all mean	33.70	10.62	27.17	1996.85	7.10	631.26	10.86

Performance of *Stevia* for morphological traits

Plant height: Plant height seem to be influenced by testing locations and years. Plant height varied from 31.03 to 34.62 cm in the first year and 19.82 to 42.3 cm in the second year. In both years, highest plant height was recorded at Wondo Genet. When compared with the second year, the overall plant height had increased by 4.5% during the first year. The result obtained

in the present study agrees with the reports of Mohd *et al.* (2009) who reported a plant height range between 29.77 and 35.77 cm.

Branch number: Branch number was also influenced by testing locations and years. The highest value of branch number (34) was recorded during the first year at Debrezeit and the lowest value (5) was recorded at Awada during the second testing year. In agreement with the present study, a very similar range of number of shoots/plant from 8-34 was reported by Satpathy and Das (2010). But maheshwar (2005) had reported 12 to 17 branches/plant. As the reports of the present study on morphological traits of Stevia are within the range of the different reports, it can be said that the plant is well adapted in the country.

Performance of Stevia for economic traits

Leaf Yield: The mean performance of Stevia for economic traits including fresh leaf yield/plant, dry leaf yield/plant, dry leaf yield/ha and stevioside content are summarized in table 2. Compared with the first testing year, a respective increased percent value of 14.69 and 45.82% was recorded for fresh leaf yield/ha and dry leaf yield/ha during the second testing year. On the other hand, a percent decrease value of 9.5 and 34.38% was recorded on the first testing year as compared with second testing year for fresh leaf yield/plant and dry leaf yield/plant, respectively. The respective overall average fresh leaf yield/plant and dry leaf yield/plant were 33.04 and 10.82 g for first year and 27.17 and 7.10 g for second year. In the first year, fresh leaf yield/ha ranged from 992.5 to 2332.01 kg and the highest was recorded at Chenchu and lowest at Awada. In the second year, however, a fresh leaf yield/ha ranges between 1891.00 and 3034.20 kg was recorded at Debre Zeit and Wondo Genet, respectively. During the two year testing, a higher total fresh leaf yield of 4940.91 kg and dry leaf yield/ha of 1140.07 kg was

obtained at Chenchu. A dry leaf yield range between 225.18 and 599.57 kg/ha for first year and from 540.50 to 768.97 kg/ha was recorded for second year. A range of 7.1-13.8 t/ha fresh leaf yield and 1.91 to 3.54 t/ha dry leaf yield was reported by Maheshwar (2005). Study conducted in Malaysia by using 60 accessions (59 originated from Canada and one from Russia) demonstrated an average fresh leaf yield and dry leaf yield of 10 t/ha and 2.8 t/ha, respectively (Tan *et al*, 2008). The variation in the performance of economic traits in Stevia compared with different reports may be due to the existence of variation of different factors. Allard (1960) and Poehlman and Sleper (1995) also reported that the occurrence of performance variation in any plant is due to hereditary differences in the plants, difference in the environments in which the plants are grown, or a combination of both.

Percent Stevioside Content: The overall mean stevioside content of Stevia over the testing locations was 10.86%. It varies from 9.07 to 13.35% and the highest being recorded at Chenchu and the lowest at Wondo Genet. A relatively lower range of stevioside content, from 4.1 to 9.1%, was reported for Indian clones (Prakash *et al*, 2012), 5-10% for Paraguay and 3.78-6.98% for China cultivars (Midmore and Rank, 2002), 3-8% (Melis, 1992) and 5-10% (Nirpendra *et al*, 2011). An average value of 9.1% stevioside content was also reported for wild Stevia (Goyal *et al*, 2010). As the stevioside content obtained in this study is comparable and even higher than other countries, it can be said that Stevia is adaptable to some locations in Ethiopia and hence it can be cultivated for the production of stevioside.

CONCLUSION AND RECOMMENDATION

Generally, the value recorded for morphological and economic traits in the present study are within the ranges of world reports, indicating the adaptability of Stevia for morphological and yield traits in the country and possibly for large scale commercial cultivation. As shown in the study, the performance of Stevia for morphological and economic traits were not consistent at different locations, thus, identification of favorable location is very much important for Stevia production. Therefore, Stevia can be cultivated for production of leaves and for the extraction of stevioside in highland environment of the country having similar topological and climatic conditions with Chench and Wondo Genet. As this research is the first to its kind, different studies including spacing requirement, irrigation scheme and rate, fertilizer rate and type, cultural management, crop protection, cost benefit analysis and post harvest handling and processing technologies need to be investigated in the future to maximize the yield and quality.

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