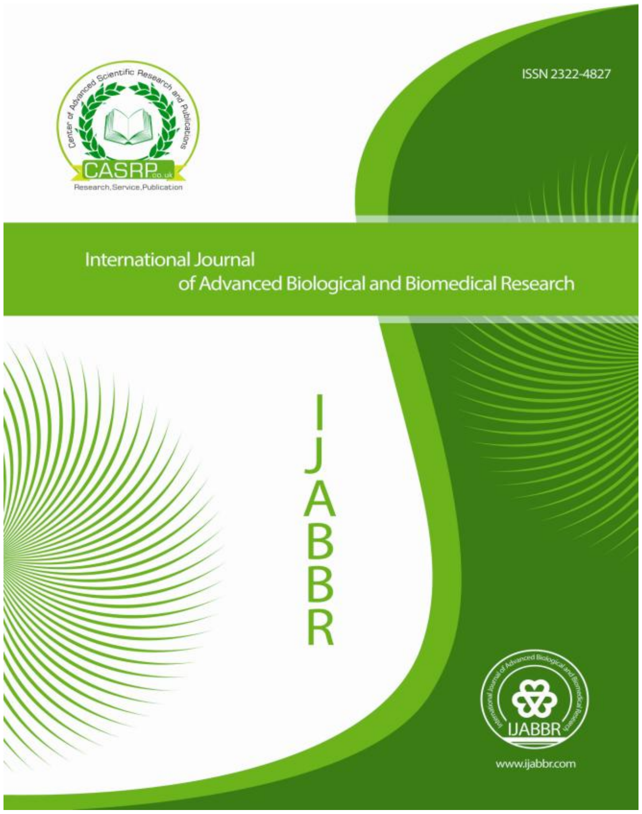
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Original Article **Open Access**

Performance of sage (*Salvia officinalis* L.) for morpho-agronomic and chemical traits in different agro-ecologies of Ethiopia

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Abstract

This experiment was conducted with the objective of evaluating the performance sage (*Salvia officinalis* L.) in different agroecology of Ethiopia for morpho-agronomic and chemical traits at Wondo genet, Hawassa, Qoqa and Holleta for two years from 2011/2012 to 2012/2013 cropping seasons. Data on plant height, branch number/plant, fresh leaf weight/plant, fresh stem weight/plant, above ground biomass/plant, fresh leaf weight/ha, essential oil (EO) content and EO yield/ha were collected from four locations arranged in randomized complete block design with four replications. Growing location demonstrated a significant influence (P<0.05) on the performance of all the parameters considered. With increasing year, the performance of Sage was significantly affected (P<0.05) only on three of the nine traits. The interaction between location and yea was significant for seven of the nine traits. The overall average value of plant height, branch number/plant, leaf number/plant, fresh leaf weight/plant, fresh stem weight/plant, above ground biomass/plant, fresh leaf weight/ha EO content and EO yield/ha of sage across the tested location and years ranged from 47.7-66.9 cm, 70.13-262.07, 619.7-4046.2, 159.13-506.31 g, 63.77-279.93 g, 221.6-743.8 g, 2772-14064 kg, 1.11-1.70% and 30.67-240.03 kg, respectively. The maximum for all the characters were recorded at Hawassa and Wondo Genet except for branch number which was found tall at Holleta. Most of the characters are statistically the same over the testing years; however, plant height, branch number and leaf number/plant demonstrated a respective percent increase value of 21.3%, 50.19% and 71.98% during the first testing year.

Keywords: Essential oil, Ethiopia, Herb, Sage (*Salvia officinalis* L.).

1. Introduction

Sage is one of the most important species of the genus Salvia which comprises nearly 1000 species throughout the world, and represents one of the largest genera in the Lamiaceae family (Lakusic et al., 2013). As its Latin name Salvia means ‘to cure’ and species name ‘officinalis’ means medicinal, it is clear that sage has a historical reputation of promoting health and treating ailments and even in ancient Rome, it was even called the sacred plant (Kamatou et al., 2008). Among the different species of the genus Salvia, *Salvia officinalis* is economically most important species being widely used in medicine, cosmetic and food industry (Al-Tawaha et al., 2013; Grzegorcazyk and Wysokinska, 2008). It was first found northern Mediterranean countries (Lakusic et al., 2013) and eventually spread to England, France and Switzerland in the fourteenth century (Miller, 1976). It has been grown as a perennial medicinal and culinary herb for thousands of years and can now be found in gardens everywhere indicating the significance of the crop in diversified communities of the world.

Sage is one of the most appreciate herbs for its rich essential oil and its plethora of biologically active compounds extensively used in folk medicine (Aziz et al., 2013). It can be used as herbal tea, for food flavoring, in cosmetics, perfumery and the pharmaceutical industries throughout world (Chalchat et al*.,* 1998). It is generally known for their multiple pharmacological effects including their antibacterial (Delamare et al., 2007; Kamatou et al., 2008), antiviral (Loizzo et al., 2008), antioxidative (Kelen and Tepe, 2008), antimalarial (Kamatou et al., 2008), anti-inflammatory (Baricevic et al., 2001), antidiabetic (Eidi et al., 2009), cardiovascular, antitumor and anticancer (Loizzo et al., 2007)**.** Recent studies show that some plants from the lamiaceae family are very rich in phenolic compounds, such as flavonoids, phenolic acids and phenolic diterpenes and possess high antioxidant activities (Alizadeh et al., 2010; Lu and Foo, 2001; Zheng and Wang, 2001). Phenolic compounds are plant secondary metabolites and naturally present in all plant materials (Gülçin, 2005; Psomiadou et al., 2002). These compounds can delay or inhibit the oxidative damage caused by free radicals (Velioglu et al., 2007) and can protect human beings against major diseases such as coronary heart disease and cancer in human (Kris-Etherton et al., 2002). Due to the presence of sufficient phenolic compounds and its antioxidative properties, the leaves of Sage are reported to be used widely in the food processing industries (Farag et al., 1989; Lamaison et al., 1990; Cuvelier et al., 1994; Hohmann et al., 1999). There is increasing evidence to suggest that many degenerative diseases, such as brain dysfunction, cancer, heart diseases, and weakened immune system, could be the result of cellular damage caused by free radicals, and antioxidants present in human diet may play an important role in disease prevention (Aruoma, 1998; Nees and Powles, 1997; Steinmetz and Potter, 1996). There are some reports that sage has been recommended through the centuries as restoratives of lost or declining mental functions (Tildesley et al., 2003; Tildesley et al., 2005).

Morphological characters, yield and quality of Sage are influenced by season, geographic origin, environmental factors, extraction methods, plant organ (Santos-Gomes et al., 2001), phonological stage (Mirjalili et al., 2006), sampling techniques (Putievsky et al., 1986) and genetic differences (Perry et al., 1996). Therefore it is important to understand to what extent environment affects the adaptability of Sage for Morph-Agronomic and chemical characteristics in different locations of Ethiopia.

Despite Sage is very useful in food, cosmetic and pharmaceutical industries (Al-Tawaha et al., 2013; Grzegorcazyk and Wysokinska, 2008); introduced to Ethiopia long time ago and availability of increasing interest of farmers and investors for its cultivation in Ethiopia (EIAR, 2009) and existence of diverse ecological conditions in the country (NMSA, 1996; Kebebew, 2003; Andargachew, 2007), there exist scanty information about the production, processing and utilization technologies of Sage in Ethiopia. This lack of information is the major hindrance to exploit the potential of the plant. Currently Wando Genet Agricultural Research Center of Ethiopia is doing a number of awareness creations about the production, processing, marketing and utilization of prioritized aromatic and medicinal plant at national level. From this priority sage is one. Therefore, in order to contribute in addressing the existing technology gaps and bringing the crop for cultivation and utilization, this activity was designed with the objective to evaluate the performance of Sage for morphological, agronomical and chemical traits under different ecologies of Ethiopia.

2. Materials and methods

Planting material maintained at the botanic garden of Wondo Genet Agricultural Research was used for the study. The experiment was conducted in Oromia and SNNPRS regions of Ethiopia at Wondo Genet, Hawassa, Koka and Holleta for two years between 2011/2012 and 2012/2013. The ecological descriptions of the testing locations are summarized under table 1. Top cuttings having a length of 9 cm were taken from 6 months old disease free mother plants for seedling preparation. The lower two pairs of leaves were removed and planted in properly prepared soil mixtures in polyethylene bags of 10 cm diameter and 12 cm height. Seedlings were raised in the nursery for three months in polyethylene bags before being transplanted to the field experimental plots.

A spacing of 60 cm was maintained between rows and plants. Six rows each having six plants were maintained in the plot. No fertilizer and chemical was applied during evaluation activity. All cultural practices and watering through flooding irrigation were done as required. Harvesting was done 6 months after transplanting to the main experimental field. Harvesting was done by cutting the plant 10 cm above the ground level with the help of sickles as soon as the night dew has evaporated from the plants.

Data on plant height, number of branch/plant, fresh leaf weight/plant, fresh stem/plant, above ground biomass/plant, fresh leaf weight/ha, essential oil content and essential oil yield/ha were collected from four locations arranged in randomized complete block design with four replications according to the procedures of Gomez and Gomez (1984). Percent EO content was determined on fresh weight (w/w) basis from 250g of fresh composite leaves harvested from the three middle row plants of a plot. The laboratory analysis was done at Wondo Genet Agricultural Research Center. EO was extracted by hydro distillation as illustrated by Guenther (1972). Hydro distillation is a distillation method in which the plant material to be distilled (in this case the coriander seeds) comes in direct contact with the boiling water. Heat was provided by electro mantle. The emerging vapor from the flask containing the volatile essential oil was led to a condenser for condensation and collected in the oil separate unit. Essential oils that float on top of the separator funnel were collected and their weight was measured (W_{oil}). Then, essential oil (EO) content was calculated as follow.

$$EOcontent(W/W\%) = \frac{W_{oil}}{W_{freshleaf}} X100$$

To statically analyze the differences in agronomic and chemical characteristics caused by the growing locations and years, five samples were taken from the central rows of each plot replicated four times. Statistical analysis of experimental data was performed by analysis of variance (ANOVA) using SAS PROC GLM (2002) at P<0.05. Differences between means were assessed using the least significance difference (LSD) test at P<0.05 according to the procedures of Snedecor and Cochran (1990).

3. Results and discussion

3.1. Variation in morpho-agronomic characteristics and chemical traits of sage

Mean squares from the analysis of variance for morphological, agronomic, economic and chemical traits are summarized in table 2. Analysis of variance reveled that testing year exerted a highly significant influence (P<0.01) on plant height and branch number/plant and a significant influence (P<0.05) on number of leaves/plant and statistically non significant influence (P>0.05) on fresh leaf weight/plant, fresh stem weight/plant, above ground biomass/plant, essential oil content and fresh leaf weight/ha. Interaction effect between testing location and testing years also excreted a significant influence on plant height/plant, branch number/plant, fresh leaf weight/plant, fresh stem weight/plant, aboveground biomass/plant, fresh leaf weight/ha and essential oil yield/ha. Thus, indicating performance inconsistency of these characters to varied environments. Testing location also exerted a statistically significant influence (P<0.05) on above ground biomass per plant and essential oil yield/ha, highly significant influence (P<0.01) on plant height, number of leaves/plant, fresh leaf weight/plant, fresh stem weight/plant, fresh leaf weight/ha and essential oil content (P<0.01) and a very highly significant influence (P<0.001) on branch number/plant. This indicates, these traits were influenced by a change in the growing environment. The significance of location effect was expected because Hawassa, Wondo Genet, Koka and Holleta vary in their soil type, rainfall and temperature (Table 1).

In agreement to the present study, Fehr (1991) reported that every factor that is a part of the environment of a plant has the potential to cause differential performance. Likewise, Frankel et al. (1994) and IRRI (1996) reported that fluctuating features of the location such as rainfall, relative humidity, temperature, etc. are some of the environmental factors that cause performance variation in plants. The influence of location on agronomic and chemical traits of aromatic and medicinal plants were also reported for *Oreganum vulgaris* (Beemnet et al., 2014a), *Matricharia chamomile* L. (Beemnet et al., 2015), *Mentha spicata* L. (Beemnet et al., 2014b), *Aloysia triphylla* L. (Beemnet et al., 2013), *Coriandrum sativum* L. (Beemnet and Getinet, 2010), *Cymbopogon citratus* L. (Beemnet et al., 2014c; Beemnet et al., 2011), *Artemisia annua* L. (Belay, 2007), *Stevia rebaudiana* Bertoni (Beemnet et al., 2012) and Aflatuni (2005) for *Mentha arvensis* L. and *M. piperiata* L., indicating the importance of knowing optimum growing locations before intending production of oregano in Ethiopia.

3.2. Performance of sage in morpho-agronomic and chemical traits in different agro-ecologies of Ethiopia

Analysis of variance revealed that there existed a performance variation in morpho-agronomic characters (plant height, branch number, leaf number/plant, fresh leaf yield/plant, fresh stem weight/plant, aboveground biomass yield/plant and leaf yield/ha) and chemical characters (percent EO content and EO yield/ha) of sage across testing locations and years (Table 2). The respective mean performances of morpho-agronomic and chemical characters of due to location effects and testing years are summarized in tables 3 and 4.

3.2.1. Performance of sage in morphological and agronomic traits in different agro-ecologies of Ethiopia

The overall mean performance of plant height, branch number/plant, leaf number/plant, fresh leaf weight/plant, fresh stem weight/plant, above ground biomass/plant, fresh and leaf weight/ha were 59.3 cm, 197.3, 2626.9, 408.13 g, 184.28 g, 592.075 g and 9951 kg, respectively. The respective plant height, branch number/plant, leaf number/plant, fresh leaf weight/plant, fresh stem weight/plant, aboveground biomass/plant and fresh leaf yield/ha of sage across the tested location ranged from 47.7-66.9cm, 70.13-262.07, 619.7-4046.2, 159.13-506.31g, 63.77-279.93 g, 221.6-743.8 g and 2772-14064 kg, respectively (Table 3). The maximum for all the characters were recorded at Hawassa and Wondo Genet except for branch number which was found tall at Holleta. Most of the characters are statistically the same over the testing years; however, when compared with the second testing year, plant height, branch number and leaf number/plant demonstrated a respective percent increase value of 21.3%, 50.19% and 71.98% during the first testing year (Table 4).

The current study demonstrated a higher value compared with the report of Aziz et al. (2013) who reported a range of values for plant height (11.67-35 cm), branch number/plant (3.67-11), aboveground biomass/plant (3.57-46 g) for studies conducted under different salt stress condition in Egypt. Comparable regent of plant height range from 42.86-69.53 cm and a relatively lower fresh herb yield range between 4115.2-8190.88 kg/ha was reported by Agha et al. (2011) for exotic sages tested at Quetta and Kalat locations in Pakistan. The variation in morpho-agronomic characters may be due to the variation in genotype, environment, soil, climatic factors. As the value obtained in this experiment demonstrated relatively higher values for fresh leaf yield, it is possible to cultivate sage in Ethiopian for the production of Sage herbal leaf.

3.2.2. Performance of sage for chemical traits in different agro-ecologies of Ethiopia

The overall mean value of EO content and EO yield tested over three locations and two years were 1.44% and 149.34 kg. The EO content and EO yield ranged from 1.106–1.707% and 30.77–239.46 kg, respectively. The highest values of these characters were obtained at Hawassa and the lowest at Holleta (Table 3). The values on EO content and EO yield were consistent under both testing years (Table 4). Hence, it is the location that affects the production of Sage in Ethiopia under these testing locations. The ranges of values obtained in the present study are in agreement within the range of different reports. The essential oil content range obtained in the present study is within the ranges of values from 1.11 to 2.76% on dry weight basis for six sage accessions grown under North Western Himalayan Region of India (Raina et al., 2013). A wide and lower range of essential oil content between 0.2% and 1.3% was reported by Boszormenyi et al. (2009) for five sages in Hungary. Similarly, a lower essential oil content range from 0.11-0.56% and essential oil yield range between 0.01 and 0.24 ml/plant was reported for common sage by Aziz et al. (2013) for studies conducted under different salt stress condition in Egypt. Comparatively lower essential oil content (0.5-0.8%) and essential oil yield (6.8-24.4 kg/ha) was reported by Ardakani et al (2014) for clary sage in Iran. The difference in essential oil content and essential oil yield may be due to climatic factors such as temperature, rainfall and light. Fahmy (1955) mentioned that climatic factors such as temperature, day length, humidity and rainfall, affected oil content of mint plants.

Table 1
Summary of the site description used for adaptation testing of Sage (*S. officinalis* L.) for yield and yield component.

Test location	Longitude	Latitude	Soil pH	Soil type	Rain fall (mm)	Altitude (M.s.s.l)	Annual average temperature (^{0}c)	
							Minimum	Maximum
Wondo Genet	$7^0$19’N	38^0 38’E	6.4	Sand clay loam (Nitosol)	100	1876	12.02	26.72
Hawassa	$7^0$05’N	39^0 29’E	7.2	Sandy loam (Andosol)	964	1652	12.94	27.34
Qoqa	$8^0$26’N	39^0 1’E	-	Clay soil	830.9	1604	13. 68	28.30
Holleta	$9^0$03’N	38^0 30’E	5.5	Red brown clay loam soil (Nitosol)	1100	2390	6.13	22.20

Table 2

Mean square from the combined analysis of variance for the different traits of Sage (*S. officinalis* L.) tested at Wondo Genet, Hawassa, Koqa, and Holleta during the years 2011/12 and 2012/13.

		Mean squares								
Source of variation	**DF**	**PH**	**BN**	**NL**	**FLWPP**	**FSWPP**	**AGBM**	**EOC**	**FLWPHA**	**EOY(kg)**
Year (Y)	1	784**	37588**	11599551*	12730ns	30379ns	81508.1ns	0.017ns	145012.4ns	60634634 ns
Location (L)	3	435**	79445***	12551433**	169712**	54600**	372452*	0.436**	151184419**	60265649*
Replication (R)	2	4	447	219881	7715	2966	18923	0.025	4589859	9379630
Y*L	3	415**	199369***	4845942ns	154249**	57477*	384874*	0.076ns	144604119**	60095117*
Y*R	2	57ns	4380ns	1126411	9213ns	901ns	15620ns	0.051ns	14502164ns	9251256 ns
L*R	6	80ns	3249ns	1009024	21476ns	3203ns	39748ns	0.025ns	9701247ns	9435997 ns
Cv (%)		7.46	19.52	22	25	25	30.7	13.3	25	18.6

*******,* * and * Significant at P<0.001, P<0.01 and P<0.05 probability level, respectively. Ns= non significant at P>0.05 probability level DF = degree of freedom, REP= Replication, PH= plant height, BN= branch number per plant, NL= number of leaves per plant, FLWPP= fresh leaf weight per plant, FSWPP= fresh stem weight per plant, AGBM= above ground biomass per plant, EOC = Essential oil content, FLWPHA= fresh leaf weight per hectare EO= Essential oil yield per hectare, CV= Coefficient of variation.

Table 3

Mean performance of Sage (*S. officinalis* L.) for its agronomic and chemical traits tested at Wondo Genet, Hawassa, Koqa, and Holleta during the years 2011/12 and 2012/13.

Locations	PH (cm)	BN	NL	FLWPP (g)	FSWPP (g)	AGBM (g)	EOC (%)	FLWPHA (kg)	EOY(kg)
Hawassa	64.20^{ab}	262.07^{b}	4046.2^{a}	506.31^{a}	237.44^{ab}	743.8^{a}	1.7067^{a}	14064^{a}	239.46^{a}
W/Genet	66.933^{a}	135.13^{c}	3093.5^{a}	453.18^{a}	279.93^{a}	733.1^{a}	1.6017^{ab}	12588^{ab}	201.62^{a}
Qoqa	58.733^{b}	70.13^{d}	2748.0^{a}	513.90^{a}	155.97^{bc}	669.9^{a}	1.3417^{bc}	10379^{b}	133.98^{a}
Holleta	47.667^{c}	321.77^{a}	619.7^{b}	159.13^{b}	63.77^{c}	221.6^{b}	1.1067^{c}	2772^{c}	30.77^{b}
LSD $_{(0.05)}$	6.26	54.419	1560.9	145.19	118	257	0.27	3526.5	43.21
Mean	59.38325	197.275	2626.85	408.13	184.2775	592.1	1.4392	9950.75	149.34

Means followed by the same letter with in the same column are statistically non significant at *P* < 0.05 according to least significant difference (LSD) test. PH= plant height, BN= branch number per plant, NL= number of leaves per plant, FLWPP= fresh leaf weight per plant, FSWPP= fresh stem weight per plant, AGBM= above ground biomass per plant, EOC = Essential oil content, FLWPHA= fresh leaf weight per hectare EOY= Essential oil yield per hectare, CV= Coefficient of variation, LSD= Least significant differences.

Table 4
Mean performance of Sage for its agronomic and chemical traits tested at Wondo Genet, Hawassa, Koqa, and Holleta during the years 2011/12 and 2012/13.

Year	PH (cm)	BN	NL	FLWPP (g)	FSWPP (g)	AGBM (g)	EOC (%)	FLWPHA (kg)	EOY(kg)
2012/2013	65.100[a]	236.85[a]	3322.1[a]	431.16[a]	219.86[a]	650.35[a]	1.46583[a]	10029[a]	150.68[a]
2012/2013	53.667[b]	157.70[b]	1931.7[b]	385.10[a]	148.70[a]	533.80[a]	1.41250[a]	9873[a]	147.99[a]
Mean	59.3835	197.275	2626.9	408.13	184.28	592.075	1.439165	9951	149.34
$LSD_{(0.05)}$	4.428	38.48	1103.7	102.67	83.534	181.81	0.1916	2493.6	30.56

Means followed by the same letter with in the same column are statistically non significant at $P < 0.05$ according to least significant difference (LSD) test PH= plant height, BN= branch number per plant, NL= number of leaves per plant, FLWPP= fresh leaf weight per plant, FSWPP= fresh stem weight per plant, AGBM= above ground biomass per plant, EOC = Essential oil content, FLWPHA= fresh leaf weight per hectare EOY= Essential oil yield per hectare, CV= Coefficient of variation, LSD= Least significant differences.

4. Conclusion

Generally, the values obtained in this experiment demonstrated comparable and even higher values in morpho-agronomic and chemical traits with the different reports. Indicating the adaptability of Sage cultivar for morphological, leaf yield, essential oil content and essential oil yield in Ethiopia. Hence, it is possible to use the existing Sage cultivar for the production of fresh herbal, dried leaf as a spice, medicinal leaves and essential oil in Ethiopia.

Conflict of interest:

All the authors of this research papers have no conflict of interest.

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