academicresearchJournals

Vol. 6(2), pp. 70-76, February 2018 DOI: 10.14662/ARJASR2017.066 Copy©right 2018 Author(s) retain the copyright of this article ISSN: 2360-7874 http://www.academicresearchjournals.org/ARJASR/Index.htm

Academic Research Journal of Agricultural Science and Research

Full Length Research

Agronomic Characterstics and Essential Oil Yield Of Java Citronella (*Cymbopogon Winterianus Jowitt*) as Affected by Harvesting Age and Plant Population Density

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Accepted 25 September 2017

A study was conducted to evaluate the influence of population density and harvesting age on agronomic and chemical characteristics of Java Citronella (*Cymbopogon winterianus Jowitt*) at Wondo Genet, Southern Ethiopia for three years. The experiment consisted of three levels of plant population density and five levels of harvesting age arranged in split plot design with three replications. Population density was assigned to main plot and harvesting age to sub-plot. Data on number of tiller hill⁻¹, number of leaf hill⁻¹, fresh leaf weight hill⁻¹, dry leaf weight hill⁻¹, fresh biomass yield ha⁻¹, dry biomass yield ha⁻¹, essential oil content and essential oil yield ha⁻¹ were collected and analyzed. Population density significant influence on all of the parameters except essential oil content. Harvesting age also exerted a significant influence on all of the studied parameters except on number of tiller hill⁻¹. Interaction effect of population density and harvesting age was significant on all of the parameters except on dry biomass yield ha⁻¹, essential oil content and essential oil yield ha⁻¹. The highest fresh biomass yield ha⁻¹, dry biomass yield ha⁻¹ and essential oil yield ha⁻¹ was recorded at plant population density of 27,778 and for harvests made at every six months after planting.

Key Words: Biomass, citronella, essential oil, Ethiopia, yield.

Cite this article as: Zigene ZD, Kassahun BM, Degu B (2018). Agronomic Characteristics and Essential Oil Yield Of Java Citronella (*Cymbopogon Winterianus Jowitt*) as Affected by Harvesting Age and Plant Population Density. Acad. Res. J. Agri. Sci. Res. 6(2): 70-76

INTRODUCTION

The *Cymbopogon* genus is composed of more than 100 species found in tropical countries (Lorenzi and Matos, 2003). About 56 species are aromatic and some of them are medicinal, pharmacological and industrial important (Morais *et al.*, 2005; Singi *et al.*, 2005). Two species of *Cymbopogon* are known according to their region. *Cymbopogon* nardus (Jamarosa) and *Cymbopogon* winterianus (Java citronella). Have similar volatile oil

scent and medicinal uses, but show different citronellal content. Of these Java Citronella (*Cymbopogon winterianus* Jowitt) is a perennial, aromatic, up to 2m in height, long and narrow leaf grass. It belonging to the Poaceae family and native to India and Sri Lanka; whereby the plant can be found growing wild in most tropical Asian countries including Malaysia (Hazwan *et al.*, 2012).

Java Citronella is a valuable herb as it produces a very important essential oil named Citronella oil or java citronella oil. The oil is reach in geraniol (36.0%) and citronellal (42.7%) (Medicinal and Aromatic Plant Series, 1999), and has a fresh, powerful, lemon like scent (Katiyar et al., 2011). The oil is extensively used in soap, perfumery, cosmetic, detergents, household cleansers, technical products, insecticides, and flavoring industries throughout the world (Wany et al., 2013). It also shows repellent, antimycotic, and acaricide activities (Blank et al., 2007). The oil is also used as an air freshener (Guenther, 1992; Matos, 2000). Citronellal, the constituent of Java citronella oil is used to prepare Hydroxycitronellal, which is a key ingredient in compounding. Hydroxycitronellal is one of the most frequently used floralizing perfume materials it finds its way into almost every type of floral fragrance and great in many non-floral ones (Medicinal and Aromatic Plant Series, 1999). In addition Citronella oil has significant anti-bacterial and anti-fungal properties. Traditionally citronella oil is used for treatment of fever, intestinal parasites, digestive and menstrual problems. In Chinese medicine, it is used for rheumatic pain (Katiyar et al., 2011).

There are many factors that affect agronomic characteristics, biomass and essential oil yield of aromatic plants (Khazaie et al., 2007). Harvesting time, plant age and crop density are among the most important factors that influence the yield of aromatic plants (Marotii et al., 1994). Even though the biosynthesis of secondary metabolites of citronella is controlled genetically, it is strongly affected by environmental, harvest and post harvest factors (Blank et al., 2007). Seasonal change, harvesting time and age of harvest also had a significant influence on essential oil yield and content of Java Citronella (Sarma et al., 2001). The influence of harvesting age and spacing on agronomic characteristics, biomass, essential oil content and essential oil vield was also reported by Beemnet et al. (2011) in Lemon grass and Pepper mint, by Zewdinesh and Beeemnet (2012) in palmarosa and by Solomon and Beemnet (2011) in spear mint and Japanis mint.

Despite the crop has huge market demand and population density and harvesting age has a great impact on agronomic characteristics, biomass and essential oil yield of the crop, there is no research conducted on their effect in Ethiopia. Therefore, this experiment is designed to investigate the impact of Plant population density and harvesting age on yield and yield components of Java Citronella.

MATERIALS AND METHODS

The experiment was conducted in Southern regions of Ethiopia at Wondo Genete Agricultural Research Center

experimental site starting from 2010 up to 2013 for three consecutive years. The site is located at 7°192 N latitude and 38° 382 E longitudes with an altitude of 1876 m.a.s.l. The area receives mean annual rainfall of 1000 mm with minimum and maximum temperature of 12.02 and 26.72°C, respectively. The experiment was consisted of three levels of population densities (27,778, 18518 and 12,345 plants ha⁻¹) and five levels of harvesting ages (3, 4, 5, 6 and 7 months after planting) laid out in split-plot design with three replication. Plant population density was assigned to main plot while harvesting age was assigned to sub plots. Each plot had an area of 12.96 m² (3.6 m length and 3.6 m width). A respective spacing of 1 m, 1.5 m and 2 m were maintained between plots, subplots and replications. A healthy planting material of citronella grass was taken and planted on experimental field on 26 June, 2010. Proper hoeing, weeding and irrigation of the experimental field were carried out uniformly whenever required. The experiment was completed on 26 may, 2013.

To determine the yield and agronomic characteristics of citronella grass under the different treatments used, data on number of tiller hill⁻¹, number of leaf hill⁻¹, fresh leaf weight hill⁻¹, dry leaf weight hill⁻¹, fresh biomass yield ha⁻¹, dry biomass yield ha⁻¹, essential oil content and essential oil yield ha⁻¹were collected. Throughout the experimental period, each parameter was collected five times for all of the harvesting age used. Average values of the five cycles over the three years were taken to estimate number of tiller hill⁻¹, number of leaf hill⁻¹ and essential oil content. For the rest parameters, the total values over the three years were taken to estimate the yield of citronella grass throughout its life cycle under Ethiopian condition. Percent EO content was determined on dry weight (w/w) basis from 250g of composite leaves harvested from the five 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).

Experimental data was statistically analyzed 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 (Snedecor and Cochran, 1990).

RESULT AND DISCUSSION

Variation in Agronomic and Chemical Characteristics of Citronella Grass

Mean square from the analysis of variance tested for evaluating the agronomic and chemical characteristics of citronella under different population density and harvesting age are summarized in Tables 1. Analysis of variance showed that the existence of a very highly

Source	Df	THi	LHi	FWHi	FBYH	DWHi	DBYH	EOC	EOY
Replication(RP)	2	170.56 ^{ns}	1503.79 ^{ns}	0.47 ^{ns}	371.38 ^{ns}	0.04 ^{ns}	30.06 ^{ns}	0.09 ^{ns}	31204.54 ^{ns}
Population Density (PD)	2	6682.14***	152594.19***	52.49***	827.31*	4.55***	99.01*	0 ^{ns}	68646.28*
RPxPD (error a)	4	167.22 ^{ns}	2148.71 ^{ns}	1.71*	828.07*	0.18*	82.87*	0.01 ^{ns}	49631.51*
Harvesting age (HG)	4	163.73 ^{ns}	12218.7***	2.53**	866.19*	0.51***	158.54***	0.2**	64650.42**
HGxPD	8	233.24*	4435.44**	1.77**	553.59*	0.16*	42.49 ^{ns}	0.05 ^{ns}	25403.51 ^{ns}
Error	16	73.9	1239.46	0.45	202.55	0.04	19.7	0.04	14472.8
CV (%)		11.2	9.7	12.34	14.7	12.8	15	7.5	15.45
*** Cignificant at D . 0.001. *	* Cianifi	append of $D = 0.011 *$	Cignificant at D	. 0 0 Et ma	lon cignificon		TIL Tiller bi	11-1 I I I I I	

Table 1. Mean squares of agronomic and chemical traits of citronella as affected by harvesting age and population density

***= Significant at P < 0.001; **= Significant at P < 0.01; *= Significant at P < 0.05; ns= Non significant at P < 0.05, THi= Tiller hill⁻¹, LHi= Leaf hill⁻¹, FWHi= Fresh leaf weight hill⁻¹, FBYH = Fresh biomass yield ha⁻¹, DWHi = Dry weight hill⁻¹, DBYH = Dry biomass yield ha⁻¹, EOC= Essential oil content, EOY= Essential oil yield ha⁻¹

significant (P<0.001) influence of plant population density on number of tiller hill⁻¹, number of leaf hill⁻¹ ¹, fresh leaf weight hill⁻¹ and dry leaf weight hill⁻¹ and a significant influence (P<0.05) on fresh biomass yield ha-1, dry biomass yield ha-1 and essential oil yield ha⁻¹. The influence of population density was non significant (P>0.05) on essential oil content. Harvesting age exerted a very highly significant (P<0.001) influence on number of leaf hill⁻¹, dry weight hill⁻¹, dry biomass yield ha⁻¹ and a highly significant (P<0.01) influence on fresh weight hill⁻¹, essential oil content and essential oil vield ha⁻¹. Effect of harvesting age was significant (P<0.05) on fresh biomass yield ha⁻¹ but its effect was not significant (P > 0.05) on number of tiller hill⁻¹. Interaction effect of harvesting age and population density was highly significant (P < 0.01) on number of leaf hill⁻¹ and fresh weight hill⁻¹; and significant (P < 0.05) on number of tiller hill⁻¹, fresh biomass yield ha⁻¹ and dry weigh hill⁻¹. Interaction effect of plant population density and harvesting age was not significant (P > 0.05) on dry biomass yield ha⁻¹, essential oil content and essential oil yield ha⁻¹.

Effect of Plant Population Density on Dry biomass yield ha⁻¹ and Essential oil yield ha⁻¹ of Citronella

As showed in table 2, dry biomass yield ha⁻¹ and essential oil yield ha-1 of citronella increased with increasing plant population density and reached at the maximum value for the highest population density used. The value ranged from 27.76 t ha⁻¹ to 32.43 t ha⁻¹ and from 733.35 kg ha⁻¹ to 856.24 kg ha⁻¹ for dry biomass yield ha⁻¹ and essential oil vield ha⁻¹, respectively. Percent increase from the lower plant population density (12,345 plants ha⁻¹) to the highest plant population density (27,778 plant ha⁻¹) was 16.82% for dry biomass yield ha⁻¹ and 16.76% for essential oil yield ha⁻¹. In agreement to the current finding, an increase in dry biomass yield ha⁻¹ with increasing plant population density was reported by Zewdinesh and Beemnet (2012) for palmarosa (Cybopogon martinii) and by Solomon and Beemnet (2011) for Pepper mint (Mentha pepriata L.). The higher leaf yield ha⁻¹ of rose-scented Geranium (Pelargonium graveolens) at higher plant population density

reported by Beemnet et al. (2012) was also support the current finding. Al-Ramamneh (2009) also reported maximum dry leaf yield ha⁻¹ at higher density than lower density for thyme (Tyhmus vulgaris.). The higher dry biomass yield ha⁻¹ at highest plant population density (narrower spacing) is due to the occurrence of more number of plants per unit area of land. Similar findings were also reported by Charles et al. (1990) in Artemisia (Artemisia annua), by Nekonam and Razmjoo (2007) in Plantago ovate and by Aflatuni (2005) in Pepper mint (Mentha pepriata). In accordance with the present study, an increase in essential oil yield with increasing plant population density was reported by Linares et al. (2005) for Lemon grass (Cymbopogon citrates), by Khorshidi et al. (2009) for Fennel (Foeniculum vulgare) and by Simon et al. (1990) for Artemisia (Artemisia annua). The results reported by Zewdinesh and Beemnet (2012) for palmarosa (Cymbopogon martinii) by Beemnet et al. (2011) for Pepper mint and by Solomon and Beemnet (2011) for Japanis mint were also support this finding.

Treatments			
HG (Months)	Dry biomass yield ha ⁻¹ (t)	Essential oil content (%)	Essential oil yield ha ¹ (kg)
3	23.14 ^c	3.00 ^a	670.9 ^c
4	29.3 ^b	2.67 ^b	726.65 ^{bc}
5	29.17 ^b	2.75 ^b	803.82 ^{ab}
6	34.75 ^a	2.68 ^b	894.75 ^a
7	31.02 ^{ab}	2.61 ^b	796.19 ^{ab}
LSD 0.05	4.43	0.2	120.2
Spacing			
27,778	32.43 ^a	2.73	856.24 ^a
18,518	28.24 ^b	2.75	745.79 ^b
12,345	27.76 ^b	2.74	733.35 ^b
LSD 0.05	3.43	ns	93.12

Table 2. Performance of agronomic and chemical traits of Citronella as affected by harvesting age and population density

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

Effect of harvesting age on dry biomass yield ha^{-1} , essential oil content and Essential oil yield ha^{-1} of Citronella

Performance of dry biomass yield ha⁻¹, essential oil content and essential oil yield ha-1 under the different harvesting age used is summarized in table 2. The minimum dry biomass yield ha⁻¹ (23.14 t) and essential oil yield ha⁻¹ (670.9 kg) were recorded for harvests made at every three months interval and the value of these parameters were increased with increasing harvesting age and reached at the maximum values (34.75 t ha⁻¹ and 894.75kg ha⁻¹, respectively) for harvests made at every six month. The value of dry biomass ha⁻¹ and essential oil yield ha⁻¹ were started to decline when going from six to seven months of harvesting age. When going from three months to six months of harvesting age, dry biomass yield ha⁻¹ and essential oil yield ha⁻¹ were increased by 50.2% and 33.37%, respectively. This showed that dry biomass yield and essential oil yield ha⁻¹ of Citronella increased with increasing harvesting age up to six months after planting. In line with this finding, Solomon and Beemnet (2011) reported an increase in essential oil yield with increasing harvesting age for Japanis mint. Higher biomass yield at later stage than earlier stage was also reported by Zheljazkov and Cerven (2009) in Pepper mint. In contrary to the present result Zewdinesh and Beemnet (2012) and Zewdinesh et al. (2011) reported a decreasing trained of dry biomass yield with increasing harvesting age for palmarosa and Artemisia annua, respectively. Concerning essential oil content, the value varied from 2.61% to 3%. The highest essential oil content was recorded for harvests made at every three months interval and the lower value was recorded for harvests made at every seven months interval. In going from three to seven months of harvesting age essential oil content decreased by 13%. Similar to this finding, Zewdinesh and Beemnet (2012) reported highest essential oil content at three months of harvesting age and lower value at seven months of harvesting age for Palmarosa. Higher essential oil content at bud initiation stage than flowering stage of Pepper mint reported by Zheljazkov and Cerven (2009) also support the current finding.

Interaction effect of Plant Population Density and Harvesting age on Agronomic Traits of Citronella

Interaction effect of plant population density and harvesting age on number of tiller hill⁻¹, number of leaf hill¹, fresh weight hill¹, fresh biomass yield ha¹ and dry weight hill⁻¹ are summarized in Table 3, 4 and 5. An increasing trend was observed in going from the highest plant population density to the lower at all of the harvesting age used for number of tiller hill¹, number of leaf hill¹, fresh leaf weight hill¹ and dry leaf weight hill¹. The highest number of tiller hill⁻¹ was recorded at the lowest plant population density for harvest made at every 4 months interval. The highest values for number of leaf hill⁻¹, fresh leaf weigh hill⁻¹ and dry leaf weight hill⁻¹ were obtained for harvest made from 4 up to 7 months at the same population density. As it is shown, the parameters taken as individual plant base recorded the highest values at the lower plant population density (wider spacing). This is due to the reason that at a wider spacing there is less inter-plant competition for available

Treatments	N	umber of tiller hi	II ⁻¹	Number of leaf hill ⁻¹		
HG (Months)	27,778 plants ha ⁻¹	18,518 plants ha ⁻¹	12,345 plants ha ⁻¹	27,778 plants ha ⁻¹	18,518 plants ha ⁻¹	12,345 plants ha ⁻¹
3	55.48 ^h	79.91 ^{de}	82.73 ^{cd}	231.29 ^e	322.03 ^{bcd}	360.58 ^b
4	53.53 ^h	71.79 ^{defg}	111.19 ^a	268.64 ^{de}	344.91 ^{bc}	513.73 ^ª
5	60.87 ^{gh}	75.79 ^{def}	108.55 ^{ab}	276.31 ^{de}	374.39 ^b	533.13 ^ª
6	66.71 ^{efgh}	74.49 ^{defg}	95.44 ^{bc}	336.67 ^{bc}	363 ^b	473.65 ^ª
7	54.07 ^h	61.21 ^{fgh}	100.67 ^{ab}	274.68 ^{de}	294.85 ^{cd}	493.04 ^a

Table 3. Performance of agronomic traits of citronella as affected by harvesting age and population interaction

Means followed by the same letter with in the same column and row are statistically non significant at P < 0.05 according to least significant difference (LSD) test

 Table 4. Performance of agronomic traits of citronella as affected by harvesting age and population interaction

Treatments	<u>Frest</u>	n leaf weight hill	⁻¹ (Kg)	Fresh	biomass yield h	a⁻¹ <u>(t)</u>
HG (Months)	27,778 plants ha ⁻¹	18,518 plants ha⁻¹	12,345 plants ha ⁻¹	27,778 plants ha ⁻¹	18,518 plants ha ⁻¹	12,345 plants ha ⁻¹
3	3.13 ^d	5.4 ^b	5.5 ^b	86.9 ^{bc}	99.45 ^b	68.16 ^c
4	3.5 ^{cd}	4.99 ^b	8.6 ^a	97.67 ^b	92.48 ^{bc}	106.23 ^b
5	3.49 ^{cd}	5.1 ^b	7.5 ^a	96.86 ^b	94.08 ^b	92.69 ^b
6	5.0 ^b	5.2 ^b	8.1 ^a	139.43 ^a	95.95 ^b	100.17 ^b
7	3.8 ^{cd}	4.5 ^{bc}	7.6 ^a	106.27 ^b	82.76 ^{bc}	93.8 ^b

Means followed by the same letter with in the same column and row are statistically non significant at P < 0.05 according to least significant difference (LSD) test

Treatments	Dry weight/hi(kg)				
Harvesting Age (Months)	27,778 plants hill ⁻¹	18,518 plants hill ⁻¹	12,345 plants hill ⁻¹		
3	0.88 ^e	1.45 ^{bc}	1.46 ^b		
4	1.04d ^e	1.53 ^b	2.48 ^a		
5	1.09cd ^e	1.58 ^b	2.26 ^a		
6	1.52 ^b	1.61 ^b	2.6 ^a		
7	1.3 ^{bcd}	1.45 ^b	2.43 ^a		

Means followed by the same letter with in the same column and row are statistically non significant at P < 0.05 according to least significant difference (LSD) test

resource, this leads the plat to spread and grow more. In consistent to this finding, higher fresh and dry leaf weight plant⁻¹ of rosemary at wider spacing was reported by Zewdinesh *et al.* (2012) and by Mishra *et al.* (2009). The finding of Simon *et al.* (1990) and Zewdinesh *et al.* (2011) in *Artemisia annua* and by Hussein *et al.* (2006) in *Dracocephalum moldavica* also support the present result. Higher number of branches plant⁻¹, leaf number plant⁻¹ and leaf weight plant⁻¹ at wider spacing in rose-scented Geranium (*Pelargonium graveolens*) reported by Beemnet *et al.* (2012) is also in accordance with this finding.

Fresh biomass yield ha⁻¹ of citronella was varied with varying harvesting age and spacing. The minimum fresh biomass yield plant⁻¹ (68.16 t) was obtained at harvesting age of three months for the lowest population density (12,345 plant plant⁻¹). The value of this parameter increased with increasing plant population density and the maximum value (139.43 t) was obtained at the highest plant population density (27,778 plants plant⁻¹) for harvest made at every six months of interval. Increasing plant population density from 12,345 plants plant⁻¹ to 27,778 plants plant⁻¹ increased fresh biomass yield plant⁻¹ by 51%. In agreement with this finding, Zewdinesh and

Beemnet (2012) reported higher fresh biomass yield plant⁻¹ of palmarosa at a population density of 27,778 for harvests made at every six months of interval. Similarly, Linares et al. (2005) and Patra et al. (2004) reported higher herbage yield plant⁻¹ at higher plant population density in lemongrass and Palmarosa, respectively. Similar results were also reported by Yasin et al. (2003) and Saeed et al. (1996) for Mot Elephant grass (Pennisetum purpurem Schum). An increase in fresh yield under narrow spacing was also reported by Solomon and Beemnet (2011) for Japanis mint, by Aflatuni (2005) for Pepper mint and by Beemnet et al. for rose-scented Geranium (2012)(Pelargonium graveolens). A higher biological yield of Plantago ovate at higher plant population density reported by Nekonam and Razmjoo (2007) and by Najafi and Moghadam (2002) also support the current finding.

As it is discussed above, the maximum yield for the most economically important parameters of citronella (Fresh biomass yield, dry biomass yield and essential oil yield) were obtained for plant population density of 27,778 plants ha-1 and harvests made at every six months interval. Therefore planting citronella at a spacing of 60cm x 60cm and harvests at 6 months after planting and consecutively can be recommended. In addition the author would like to recommend further investigation to see the effect of higher population density than the current higher density used and also consecutive harvesting of less than six months after the first harvest is made at six months.

AKNOWELDEMENT

We would like to acknowledge Wondo Genet Agricultural Research Center and National Aromatic and Medicinal plant project for providing all the necessary facilities and financial support for the experiment. Our sincere thanks also go to Wondimu Manebo, Teshome Tesfaye, Zerihun Jomba and Birtukan Yanera for their help in field and laboratory data collection. Our heartfelt thanks also go to all who made input for this study.

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