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Herbage yield and Bio-chemical traits as influenced by harvesting age of lemongrass (*Cymbopogon citratus* (DC) Stapf) varieties at Wondogenet, South Ethiopia

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Lemongrass, a perennial herb widely cultivated in the tropics and subtropics, designates two different species, East Indian, Cymbopogon flexuosus DC (Stapf). and West Indian, Cymbopogon citratus DC (Stapf). This study was carried out on lemongrass (Cymbopogon citratus DC (Stapf)), one of the members of the family Poaceae. The oil of this aromatic herb is one of the most important essential oils. Lemon grass has high citral content (75-85percent), the oil possesses a strong lemon odor. The studies conducted from August 2015 to January 2016 were aimed to assess the effect of harvesting age of lemongrass varieties on agronomic, herbage yield, essential oil content. Planting material was prepared by dividing well grown clumps into slips. The study consisted two varieties of lemongrass: Lomisar-I and WG-Lomisar-UA and four harvesting age: $105(H_1)$, $135(H_2)$, $165(H_3)$ and $195(H_4)$ days after planting (DAP)were factorials arranged in randomized complete block design(RCBD) with three replications. The herbage yield and essential oil content were determined. The result shows fresh herbage per hectare was in the interaction between variety WG-lomisar-UA and harvesting age at 135 days after planting which was higher by 84% from early harvesting at 105 dates after planting. The highest dry herbage per hectare was obtained when WG-Iomisar-UA harvested at 195 DAP. Thus, dry herbage yield higher by 73% from early harvested at 105 days after planting. The highest mean essential oil content was in the interaction between variety lomisar-I, harvested at 195 days after planting. Based on this result, the highest fresh herbage yield obtained when variety WG-lomisar-UA harvested at 135 DAP. The highest essential oil content was obtained from Lomisar-I variety (0.60%); while the lowest was obtained for WG-Lomisar-UA variety (0.47 %). Variety Lomisar-I is better and can be selected for the production of essential oil than WG-Lomisar UA. The best age of harvesting for essential oil production is 195 DAP.

Key word; lemongrass; harvesting age; essential oil

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INTRODUCTION

Lemongrass (*Cymbopogon citratus*) isone of the members of the family Poaceae. The oil of this herb is one of the most important essential oils. Because of its high citral content (75-85%), the oil possesses a strong

lemon odor; hence the name is given as lemongrass. Essential oils usually occur in plants as secondary metabolites and it can be obtained by physical and chemical processes such as water distillation or steam distillation, maceration, solvent extraction and in the case of fruits by a mechanical process (Abderrahmane, 2013).The oil of lemongrass is widely used for scenting soaps, detergents, and many kinds of technical products. The oil is used to cleanse oily skin, and in aromatherapy, it is used as a relaxant (Carlson et al. 2001). Commercially, in the essential oil industries it has been used in perfumes, cosmetics, beverages and pharmaceutical industries as antiseptics and flavoring agents (Geda et al., 2001)

Ethiopia has different climatic zones and soil types. This advantage offers great potential to cultivate different species and varieties of plants. Even though in developing countries most of the medicinal and some of the aromatic plants have been collected from the wild; that gives low quality herbs, cultivation of aromatic grass is an essential prerequisite for producing high quality herbs. In Ethiopia, lemongrass are commonly grown around houses in a very limited scale as an ornamental plant, for its fragrant foliage and can be recognized by the lemon scent of the leaves when crushed. Lemongrass locally used as bedding in some special traditional occasions together with Artemisia spp. The demand for essential oil In Ethiopia is majorly met through imported products. The import volumes of various types of essential oils and extracts have increased almost four fold within the last fifteen years(Ethiopian Revenue and customs Authority, 2011). The demand for the various types of essential oils potentially will increase mainly with the expansion and establishment of industries engaged in production of foods, soft drinks, alcoholic drinks, pharmaceuticals, cosmetic and various chemicals. Therefore, large scale production is at high need which should be coupled with better research and understanding of the varieties and distillation processes.

Some of the factors influencing quantity and quality of the essential oil and citral of the aromatic plants are the harvesting age, drying temperature and period of drying (Jose et al., 2006). The production of higher quality oil with high citral content (\geq 75%) is determined by the proportion of young leaves to older leaves, when harvested at a given point. Research reports showed that overall essential oil production is associated with the early growth stage in plants such as Cymbopogon *flexuosus*and Cymbopogon martini(Singh et al., 1989). According to Rocha et al. (2014) the essential oil content of lemongrass leaves did not influenced by plant age but there wasa significant change in its chemical composition. Cymbopogon flexuosus essential oil content was highest in early stages (10-20 days) of leaf development, and then declined substantially (Rocha et al. (2014).

Consequently, as the demand of herbs containing essential oil continues to increase worldwide, there has been a parallel need in agricultural research to determine the best cultural method at field and crop handling after

harvest to produce high quality herbs/oil. The content of oil and its composition are determined by the quantity and quality of aromatic raw materials which in turn easily affected by agronomic practices. An extensive study has been published on the cultivation of this plant in other countries. In Ethiopia studies are very limited particularly with regard to cultural practices like age of harvesting. It is necessary for farmers or producer to be aware and get the knowledge of proper harvesting age for lemongrass so that they will produce and supply as an input for the industry. The efficiencies of distillation and easy production of essential oils are affected by the quality of herbage yield supplied from the field. Therefore, the objective of this study is to determine the effects of harvesting age on the herbage yield and essential oil content of lemongrass varieties.

MATERIALS AND METHODS

The study was conducted in experimental site of Aromatic and Medicinal plants research center at Wondogenet during the period from September 2015 to March 2016. Geographically, the experimental site is located at 7° 192'N latitude and 38° 382'E longitude with an altitude of 1780m.a.s.l. The site receives a mean annual rainfall of 1000mm. The minimum and maximum average temperature is 12.02 and 26.72°C, respectively. The soil textural class of the experimental site is sandy loam with a pH of 6.4 (Abayneh *et al.*, 2006).

Experimental materials

Lemongrass (*Cymbopogon citratus* cv. Lomisar-I and WG-Lomisar-UA) which was evaluated for adaptability by Wondogenet Agricultural Research Center (WGARC) in 2011 and 2014, respectively, were used for herbage yield determination. WG-Lomisar-UA introduced from India and but Lomisar-I is not known when and from where it is introduced to Ethiopia. The varieties are generally stemmed less perennial grass with numerous stiff tillers arising from short rhizomatous rootstock, making large tussocks. The evergreen leaves are bright bluish to green and release a citrus aroma when crushed. The colors of the varieties are very different. WG-Lomisar-UA is light green and Lomisar-I is green. The WG-Lomisar-UA variety is more resistant to rust disease as opposed with the very rust sensitive Lomisar-I(MOA. 2014).

Treatment structure and experimental design

The treatment structure was two varieties (Lomisar-I and WG-Lomisar-UA) and four levels of harvesting ages (105, 135, 165and 195 days after transplanting)factorials arranged in randomized complete block design (RCBD) with three replications. An experimental area size of 18 ×

56 m was used for planting the lemongrass. A spacing of 60 cm between plants and rows was maintained. The area was divided into three blocks with each eight plot with spacing of 4.8m width and 6m length (28.8 m^2) and each plot has 8 rows with 10 plants. The distance between adjacent blocks was 1.5 m and a spacing of 1 m was maintained between plots.Data collection was carried out by taking random samples from central rows of each treatment. Five plants or hills were randomly selected from each treatment of the central rows for determination of herbage yield. The herbage was harvested early in the morning. The lemongrass was cut manually using sickle from 10 cm-15cm (manually) above ground level. Dried leaves and other foreign material were removed after harvested. The recordeddata were herbage yield, oil content and citral content.

Data analysis

Statistical analysis of quantitative experimental data was performed by analysis of variance (ANOVA) using SAS PROC GLM (2002). Differences between means were assessed using the least significance difference (LSD) test according to the procedures of Snedecor and Cochran (1990). The relationships between herbage yields, essential oil content and citral with harvesting age determined using Pearson correlation analysis according to the procedures given by (Ahlgren *et al.*, 2003)

RESULTS AND DISCUSSION

Herbage Yield and Bio-chemical traits

The analysis of variance revealed that there was a significant variation (P < 0.01) betweenvarieties for fresh herbage yield per hectare, and essential oil content. However, there was no significant difference between varieties for dry herbage per hectare. Harvesting age had a significant influence (P < 0.001) on both fresh and dry herbage yields as well a son essential oil content. The interaction effects of variety and harvesting age were significant for all traits of herbage yields considered in this study.

Herbage Yield

The mean Fresh herbage yield/hectare was 5.11t. Fresh herbage yield of the tested varieties over different harvesting age ranged from 4.69 to 5.53t. WG-Lomisar-UA gives the highest fresh herbage yield/hectare, which is 11.51 percent greater than Lomisar I. The maximum herbage yield of WG-Lomisar-UA was due to its height and produced maximum tiller number per bunch than variety lomisar-I. The two cultivars produced statistically different fresh herbage yield at different age of harvesting

(Table 1). The performance of varieties for fresh herbage yield was found high at 165 DAT over the harvesting age. The highest fresh herbage yield/hectare (6.57t) obtained at165DAT. Compared with earlier harvested at 105 DAT; the respective increase value is nearly77.35%. The slope of increasing in herbage biomass was very rapid at135DAT which is rise about 63 percent from early harvested lemongrass, then it increase with decreasing rate about 12 percent at 165DAT and reach maximum fresh herbage yield. Growing of fresh herbage yield was expected because the increasing in fresh biomass was due to division of the clump. When division of clump stopped at 195DAT resulting decrease fresh herbage yield and Leaves usually produce photosynthetic assimilates in excess of their own needs, exporting them to the rest of the plant. However, because the productivity declines with aging, old leaves may scarcely contribute to their own nutrition and no longer benefiting the plant as a whole. In that instance, it said to be at the compensation point, and those leaves are hypothesized to be targeted to senesce and died.

Dry herbage yield was found high at 195 DAT over the harvesting age. The highest dry herbage yield/hectare (1.47t) obtained at 195DAT.Compared with earlier harvested at 105 DAT, the respective increase value of 78%. Interaction between lemongrass varieties and harvesting age gave highly significant difference on fresh herbage yield per hectare. The highest mean fresh herbage per hectare (7.41t) was in the interaction between variety WG-lomisar-UA and harvesting at 135 days after transplanting, followed by fresh herbage per hectare (6.73t)in the interaction between variety WGlomisar-UA and harvesting age 195davs after transplanting. The lowest mean of fresh herbage per hectare (1.2t) was in the interaction between variety WGlomisar-UA and harvesting at 105 days after transplanting (Table 2).

Interaction between varieties and harvesting age also gave highly significant difference on dry herbage yield per hectare. The highest mean dry herbage per hectare (1.49t) was in the interaction between variety WGlomisar-UA and harvesting at 195 days after transplanting, followed by dry herbage per hectare (1.47t)in the interaction between variety lomisar-I and harvesting age 135days after transplanting (Table 2). The lowest mean of dry herbage per hectare (0.25t) was in the interaction between variety WG-lomisar-UA and harvesting at 105 days after transplanting.

(100, 100, 100 and 100 D/1).				
Treatment	FHH	DHH	EOC (%)	
Varieties				
Lomisar-I	4.69b	1.03	0.60a	
WG-Lomisar-AU	5.53a	1.13	0.47b	
LSD _{0.05}	0.49	ns	0.01	
Harvesting age				
105	1.51b	0.32c	0.44d	
135	5.85a	1.17b	0.49c	
165	6.57a	1.36ab	0.58b	
195	6.51a	1.47a	0.63a	
LSD _{0.05}	0.83	0.21	0.02	
CV (%)	13.17	15.94	6.04	

Table 1 Mean herbage yield and essential oil content of lemongrass varieties as influenced by different harvesting age (105, 135, 165 and 195 DAT).

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; ns= Non significant at P < 0.05. FHH= fresh herbage per hectare; DHH= dry herbage per hectare.

Table 2 Interaction between varieties and harvesting age on herbage yield(fresh herbage/hectare (FHH), dry herbage/hectare (DHH), and essential oil content(EOC)).

Source Of Variation	FHH (t)	DHH (t)	EOC (%)
V1H1	1.82c	0.39c	0.56b
V1H2	4.31b	0.87b	0.54b
V1H3	6.37a	1.39a	0.69a
V1H4	6.29a	1.46a	0.74a
V2H1	1.20c	0.25c	0.40c
V2H2	7.41a	1.47a	0.43c
V2H3	6.77a	1.325a	0.41c
V2H4	6.73a	1.49a	0.58b
LSD _{0.05}	1.18	0.30	0.06
CV (%)	13.17	15.94	6.47

Note: V1= Lomisar-I; V2= WG-lomisar-UA; H1= 105 days after transplanting; H2=135 days after transplanting; H3=165 days after transplanting; H4=195 days after transplanting.

Bio chemical traits

Essential oil content

The essential oil content of tested lemongrass varieties harvested at different age varied from 0.47 to 0.60% (Table 1). The highest essential oil content was obtained from Lomisar-I variety (0.60%); while the lowest was obtained for WG-Lomisar-UA variety (0.47 %). So Lomisar-I had better essential oil content and can be selected for the production of oil than WG-Lomisar-UA variety (Table 1). Harvesting age of lemongrass varieties had a significant influence on essential oil content. The essential oil content ranged from 0.44 to 0.63%. The performance of lemongrass varieties for essential oil content was found high at 195 DAT(0.63%) over the lowest harvesting ade and the recorded at 105DAT(0.44%)(Table 1). The essential oil content obtained at 195 date after planting was higher by 30% from early harvested at 105 date after transplanting. The essential oil content increasing with an increase age of lemongrass may be due to, Plants evolved secondary metabolism to produce wide array of low molecular weight molecules. These functions include vital processes for plant survival and development as plant signaling and control of germination, used as hormone for regeneration etc. Many secondary metabolites were reported to be used in the plant as storage compounds; in cases of nutrient starvation these compounds are mobilized and fluxed in primary metabolism pathway. Thus, the dynamic interactions between primary and secondary metabolism are very important when considering the biosynthesis and accumulation of plant natural products. This shows that essential oil content increases with delaying harvesting time of lemongrass. Older crops with highly matured leaves were found to yield higher oil. In Contrary to this finding, Tajidin et al. (2012) reported essential oil content decreased with the increase in maturity stages at harvest. In their study the plants were harvested at 5.5, 6.5, and 7.5 months after planting. The optimum percentage of essential oil was obtained when lemongrass was harvested at 5.5 months after planting. According to Lommis and Croteau (1980), essential oil metabolism was controlled by the balance between photosynthesis and the utilization of photosynthates (sucrose) or the growth differentiation balance. In the growing lemongrass leaf, the sucrose or photosynthates was obtained from older leaves, breakdown of stored starch also from photosynthetic activity (Singh and Luthra, 1987). Increased photosynthetic activity resulted in excess of carbon export compared to import in the growing leaves (Giaguinta, 1978). Excess sucrose from metabolic processes was converted to monoterpenes and finally to accumulation of essential oil. Sucrose mobilization is most rapid during the period of essential oil and citral accumulation.

The interaction effects between varieties, harvesting age showed significant difference on essential oil content of lemongrass. The highest mean essential oil content 0.74% was in the interaction between variety lomisar-I, harvested at 105 days after transplanting. Followed by essential oil content (0.69%) in the interaction between variety lomisar-I, harvested at 165 days after transplanting. The lowest mean of essential oil content(0.40%) was found in the interaction between varieties WG-lomisar-UA harvested at 105 days after transplanting (Table 2).

Percent Concentration of Citral (α-Citra and β-Citral)

Eighteen compounds were identified in the essential oils of lemongrass variety Lomisar-I and fifteen compounds in variety WG-Lomisar-AU tested at different harvesting age and drying periods (Appendix table III). Extracts from the leaves yielded aromatic oil containing 70%-90% citral as compared to other chemical component of the lemongrasses. The quality of lemongrass is generally determined by its citral content (Chisowa et al. 1998). Citral consists of the α -Citra and β -Citral. The most abundant chemical compound found in the oil samples of the lemongrass varieties used in the study was citral (a-Citra and β -Citral). However the varieties showed differences in the percent of α -Citra and β -Citral was found different among varieties (Figure 2). The highest α-Citra (42.46%) and β -Citral (36.54%) concentration was obtained for variety WG-Lomisar-AU and the lowest was recorded for variety Lomisar-I α -Citra (42.06%) and β -Citral (34.70%) (Figure 2).

The respective number of essential oil compositions components identified from lemongrass varieties harvested at 105 DAP, 135DAP, 165 DAP and 195 DAP were nine, ten, seven and nine. The major components of essential oils the α -Citra and β -Citral found were ranged from 38.00 to 48.60 % and 29.7 to 40.24% respectively. The highest concentration of α -Citra (47.69%) and β -Citral (37.96%) of lemongrass varieties were found at 165 DAP. The lowest concentration α -Citra (39.28%) and β -Citral (34.10%) were recorded at 105 DAP (Figure 4). The citral content of essential oil varied when lemongrass was harvested at different harvesting age. The citral content increased with increasing harvesting age of lemongrass which correlates with essential oil content and yield. The finding was consistent with the Tajidin et al. (2012) who reported, when lemongrass plants harvested at 5.5, 6.5, and 7.5 months after planting showed variation in the percentage of citral content and the highest content was extracted when lemongrass was harvested at 7.5 months after planting.



Figure 2. Effect of the lemongrass varieties and harvesting age on percentage of citral content both α -Citra and β -citral

CONCULUSION AND RECOMMENDATION

The tested varieties of lemongrass demonstrated a significant variation between themselves, over the testing harvesting ageing herbage yield and essential oil content. Among these two varieties the highest herbage yield per hectare can be obtained from varieties WG-Lomisar-UA and195 days after transplanting give the maximum herbage yield. There was significant influence of lemongrass varieties harvested at different age on essential oil content and essential oil yield per hectare. Variety Lomisar-I is better and can be selected for the production of essential oil than WG-Lomisar UA. The best age of harvesting for essential oil production is 195 days after transplanting. The time at which harvesting to be started for lemongrass varieties would be determined; however determination of interval of subsequent harvest on the effect of oil yield and citral accumulation could be further studied. Further studies could be mounted to observe the effect of different location on essential oil and citral content of lemongrass varieties harvesting at different age.

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