

## Response of phenology and zinc on herbage, essential oil yield and its constituents in *Cymbopogon martinii* grown under Inceptisols of Jammu, (J&K)

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### ABSTRACT

An experiment was conducted to study the response of phenology and zinc on herbage, essential oil yield and its constituents in *Cymbopogon martinii* during kharif seasons of 2011 and 2012 at research farm of Indian Institute of Integrative Medicine Jammu. Results revealed the presence of geraniol % and geranyl acetate % as major constituents in the essential oil of *Cymbopogon martinii* wats(var. motia) through GLC technique. Leaf production was quantified as phyllochron which revealed that 786.01 degree days were required for production of mature single leaf irrespective of zinc application. The application of zinc as ZnSO<sub>4</sub> through soil application was optimized as 45 kg ha<sup>-1</sup> for obtaining maximum biomass, oil yield and geraniol content. The total oil content of *Cymbopogon martinii* reached its maximum towards the senescence of older leaves with ≥ 95% flowering. The geraniol % was highest in the leaf, though geranyl acetate was highest in the inflorescence at this stage. The geraniol% is maximum in the leaf at ≥ 95% flowering, The herbage and oil yield increased by 5.53% and 32.65% respectively at optimized zinc dose. The geraniol % was increased by 3.93% whereas geranyl acetate increased by 24.35%. Thus, the crop should be harvested at the end of blooming (95% flowering) and senescence of older leaves to get the maximum oil yield and the highest % of total geraniol in the oil with application of zinc sulphate.

**Key words:** *Cymbopogon martini*, zinc, phyllochron, geraniol%, senescence, phenology

### INTRODUCTION

Total geographical area of J&K is 101387 sq.km, out of which total wasteland area is 7375438sq. km (72.75%). Jammu district alone occupies 336.59 sq.km out of total geographical area of 3097 sq.km (10.87%). The only feasible alternative is to utilize these waste lands by growing other potential crops. Palmarosa is one among them, belongs to family Poaceae and has about 780 genera and 12,000 species (Christenhusz and Byng, 2016). It is important source of essential oils for the flavour and fragrance industries worldwide and is also noted for its medicinal potential (Prashar *et al.*, 2003). It is perennial densely tufted grass attaining height of 3 meter having 50 cm long and lanceolate leaf blade. Geraniol and geranyl acetate are the main constituents of palmarosa oil, constituting about 95% of the total oil. Yield of geraniol is about 65-90% in the wasteland including alkali soils (Akhila *et al.*, 1984 and Smitha and Rana, 2015). Efforts have been made to quantify phyllochron, and response of phenology and zinc on herbage, essential oil yield and its constituents in *Cymbopogon martinii* grown under inceptisols of Jammu, J&K.

### MATERIALS AND METHODS

#### Plant material and study site

The germplasm of *Cymbopogon martinii* was collected from wastelands of subtropical regions of Jammu and planted vegetatively through tussocks at Research Farm of Indian Institute of Integrative Medicine. The experiment was laid out in randomized block design. The soil was sandy loam in texture with neutral pH 6.9 (1:2.5 soil to water); organic carbon 7.6g kg<sup>-1</sup>; cation exchange capacity 10.3cmol (p+) kg<sup>-1</sup>; available N, P and K 208, 15.2 and 134 kg ha<sup>-1</sup> respectively. The five doses of zinc as control, 15, 30, 45 and 60 Kg ZnSO<sub>4</sub> ha<sup>-1</sup> were tested through soil application. All doses of zinc, 40 kg P<sub>2</sub>O<sub>5</sub> 30 kg K<sub>2</sub>O ha<sup>-1</sup> were applied as basal dose and remaining 40 kg N ha<sup>-1</sup> was applied after 30 days of planting. The treatments were replicated four times in randomized block design. The seedlings of palmarosa were planted in July, 2011 and 2012 at 45x 45 cm distance and irrigated with tube well water to establish them and later on treatments were given as and when needed. Ten plants per replication were harvested (treatments) at different phenophases for recording of morphoeconomic parameters

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along with determination of essential oil yield plant<sup>-1</sup> and essential oil quality as percent geraniol and geranyl acetate were optimized for various doses of zinc as ZnSO<sub>4</sub>.

**Isolation of essential oils:** The essential oils were obtained from the aerial parts of the plant by hydrodistillation method using a Clevenger – type apparatus. Triplicate distillate was performed in succession for each sample of 500g of fresh herbage at each leafing stage / growth stage. Oils obtained at each growth stage were dried over anhydrous Na<sub>2</sub>SO<sub>4</sub> and stored at 4 ° C for chemical analysis.

**Analysis of essential oils:** The chemical composition of the oil was analysed by GLC using a Perkin Elmer 3920 gas chromatograph equipped with TCD and a 2m × 1/8” S.S column packed with k-20 M. Hydrogen gas was used as carrier at a flow rate of 25 ml/ mt and the isothermal operation was performed at 145°. The peaks were compared with those produced by authentic samples.

**Degree days (°C days):** Sum of average daily ambient temp. minus the base temp. 5°C during the growth period. (Chakravarty and Sastry, 1983).

**Phenology:** Phenological stages were monitored throughout the growing season and final leaf counts based on the scale were made

from 3 leaf to 6.1 leaf on the main culm (Haun, 1973).

**Phyllochron:** It was quantified as regression coefficient (b) between the leafing and degree days to describe the requirement of degree days to describe the requirement of degree days (Pedersen *et al.*, 1991) for the production of mature leaf during the successive growth stages. Validation of the model was done by calculating the correlation coefficient (r) value (Panse and Sukhatme, 1957) between estimated and calculated value of degree days during the phenophases and its significance was tested at 5% probability level.

## RESULTS AND DISCUSSION

### Phyllochron

Leaves are the main source of essential oil production. For quantification of phyllochron (degree days/leaf) a linear regression equation was made which exhibited that 776.01 degree days were required for the production of single leaf on the tiller (Table 1.) Validation of the model was done by correlating the predicted and estimated values of degree days as correlation coefficient  $r = 0.97$  which was significant at 5% probability level. Similar observations were quantified while studying the phyllochron of *Cymbopogon citratus* in the Jammu region of India (Shahi *et al.*, 2005).

Table 1: Linear descriptive statistics between leafing and degree days for quantification of Phyllochron\* in *Cymbopogon martini*

Regression Coefficient(b)	Regression constant(a)	Regression equation	Validation of predicted and estimated values
7.01	-925.656	YDD= -925.66+ L	0.97**

\*Based on mean replicates of two years

\*\*Significant at 5% probability level, YDD =Degree days, L = Leafing (in numbers)

**Response essential oil yield and essential oil constituents:** Essential oil yield was found maximum at senescence of older leaves with  $\cong$  95% flowering to get the maximum oil yield. This fact can be explained by the process of essential oil distribution from roots through the plant during the crop growth stage. Similar observations were made while studying *Eryngium glaciale* Boiss (Pala-Paul *et al.*, 2005) These observations are also attributed to

vegetation phase of plant and location of the plant species (Hashemabadi and Kaviani, 2010). The remarkable increase in geraniol and constant drop of geranyl acetate content was observed in the stem, leaves and flowers from the initiation of flowering towards the senescence of older leaves with  $\cong$  95% flowering. Similar observations were made while studying *Cymbopogon martini* (Akhila *et al.*, 1984 and Rao *et al.*, 2010). The monoterpenes are

unevenly distributed in stems, leaves and flowers. The essential oil glands are present only on the surface of the leaves and flowers. The very low percentage of oil was present in the stem at all phenophases. Leaves possessed

more geraniol than flowers at any stage of plant growth (Table 2). It may be due to the glycosylation of geraniol which take place in flowers.

Table 2: Effect of phenophases on herbage, essential oil yield and major essential oil constituents in *Cymbopogon martini*

*Phenophase(s)	Plant Parts	Fresh herbage g/plant	Essential oil yield/plant (w.w%) fr.wt	Geraniol%	Geranyl acetate%
Initiation of flowering	Whole Plant	393	0.42	52.65	34.28
	Stem	170	0.13	42.92	40.92
	Leaf	223	0.68	55.62	30.98
Before Anthesis	Whole Plant	550	0.57	56.82	34.12
	Stem	310	0.18	64.23	27.13
	Leaf	205	0.75	64.98	23.92
At full bloom	Inflorescence	35	0.50	45.68	45.54
	Whole Plant	515	0.88	74.21	18.48
	Stem	325	0.15	62.63	25.43
	Leaf	92	0.85	80.02	17.18
Senescence of older leaves ( $\geq 95\%$ flowering)	Inflorescence	98	1.60	65.62	25.03
	Whole Plant	524	0.98	78.82	7.02
	Stem	350	0.15	74.03	10.52
	Leaf	82	1.80	81.52	17.12
	Inflorescence	92	1.60	70.10	13.02

\*Based on mean replicates

**Optimization of zinc dose for maximum herbage, oil yield and oil constituents:** The biomass and oil yield of the plant was maximum at the treatment where zinc was applied @ 45 kg ha<sup>-1</sup>. Plant biomass and oil yield of the whole plant showed a remarkable increasing trend with different zinc doses (Table 3) indicating zinc response to Palmarosa crop. The herbage and oil yield increased by 5.53 and 32.65%

respectively at optimized zinc dose. The geraniol % was increased by 3.93% whereas geranyl acetate increased by 24.35%. Nutrient available in the nutritional environment of plants are capable of changing yield and essential oil content in aromatic and medicinal plants. Similar results have been reported with foliar nutrition application on lemon verbena (Ibrahim *et al.*, 2015).

Table 3: Response of Zinc on herbage, essential oil yield and major essential oil constituents in *Cymbopogon martinii*

Zinc doses (ZnSO <sub>4</sub> kg ha <sup>-1</sup> )	Freshherbage g/plant	Essential oil yield/plant (w.w%) fr.wt.	Geraniol %	Geranyl acetate%
0	524	0.98	78.82	7.02
15	535	1.12	79.06	8.02
30	546	1.18	80.06	8.46
45	553	1.30	81.92	8.73
60	542	1.13	78.90	8.00
C.D (0.05)	3.23	0.15	0.86	0.98

**Response of optimized zinc on herbage, essential oil yield and major essential oil constituents in *Cymbopogon martinii* at crop phenophases**

Herbage and essential oil yield was found maximum at senescence of older leaves with  $\cong$

95% flowering at optimized zinc dose. Similar observations were made by other workers (Muller-Riebau *et al.*, 1997; Thiem *et al.*, 2011). The remarkable increase in geraniol and constant drop of geranyl acetate content was observed in the stem, leaves and flowers from the initiation of flowering towards the

senescence of older leaves with  $\cong$  95% flowering at optimized dose of zinc application (Table 4). The effect of different zinc doses on essential oil and its constituents may be due to its effect on the enzyme activity and metabolism of essential oil production Further the zinc

application has slightly improved the geraniol content (%) contributing towards total alcohol responsible for medicinal potential in the essential oil of *Cymbopogon martinii* at all crop phenophases as compared to control as also observed by (Ibrahim *et al.*, 2015).

Table 4: Response of optimized zinc dose on herbage, essential oil yield and major essential oil constituents in *Cymbopogon martinii* at crop phenophases

*Phenophase(s)	Plant Parts	Fresh herbage g/plant	Essential oil yield/plant (w.w%)fr.wt	Geraniol%	Geranyl acetate%
Initiation of flowering	Whole Plant	399	0.45	55.65	36.36
	Stem	172	0.13	43.93	41.98
	Leaf	227	0.70	56.92	32.06
Before Anthesis	Whole Plant	578	0.62	57.97	35.12
	Stem	315	0.22	64.27	27.42
	Leaf	226	0.77	65.42	24.26
At full bloom	Inflorescence	37	0.54	46.35	46.62
	Whole Plant	540	1.12	74.34	19.23
	Stem	332	0.16	63.42	27.42
Senescence of older leaves( $\cong$ 95% flowering)	Leaf	106	0.89	81.98	18.02
	Inflorescence	102	1.62	66.85	25.04
	Whole Plant	553	1.30	81.92	8.73
	Stem	372	0.16	76.02	11.02
	Leaf	85	1.83	83.52	18.00
	Inflorescence	96	1.62	73.48	13.68

\*Based on mean replicates

Thus, the crop should be harvested at the senescence of older leaves with  $\cong$  95% flowering to get the maximum oil yield and highest % of geraniol in the oil in the plantation

raised on Inceptisols. The biomass of the plant was also maximum at this stage with optimized dose of 45 kg ZnSO<sub>4</sub> ha<sup>-1</sup>.

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