RESEARCH ARTICLE

Optimizing Variation in Agronomic Traits of *Cymbopogon flexuosus* and *Cymbopogon winterianus* in Different Fetilizer, AuxinTreatments and Altitude of Uttarakhand, India

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Abstract

Cymbopogon flexuosus and *Cymbopogon winterianus* are aromatic grasses, valued for essential oils. This study aims to develop propagation protocol for *C. flexuosus* and *C. winterianus* also evaluating transplanted seedling performance at altitudes ranging from 400 to 1900 m in Uttarakhand, India. Stumps were sterilized using a solution of 1% tween 20 and 1% bavistin for 20 minutes. Twenty-five stumps in triplicates (10 cm in length) were used for each treatment. Organic fertilizers used were manure, organic compost, vermicompost, organic compost + vermicompost. Inorganic fertilizers used were urea (46 mg N kg^{-1}), NPK (12-32–16 mg kg^{-1}), DAP (18–46-0 mg kg^{-1}), NPK+urea and DAP+urea. Auxins used were IAA, IBA and NAA at concentrations of 250, 500 and 1000 mg L^{-1}. The result indicates, that sprouting was 100% in *C. flexuosus* and *C. winterianus*. Plant growth of one year treated with DAP+urea exhibited maximum plant height (121.00, 115.33 cm), tiller/clump (107, 84), leaves/tiller (6.67, 7.33), and herbage weight/plant (1027.96, 990.67 g/plant) in *C. flexuosus* and *C. winterianus* respectively. Altitude and treatment showed significant growth response at $p \leq 0.05$ in growth of *C. flexuosus* and *C. winterianus*. Maximum plant height (125.13, 125.00 cm), tillers/clump (115.33, 95.00), leaf area (4358.67, 5721 mm^2), herbage weight/plant (1200.93, 1227.85 g/plant) and seed number/plant (377.67, 311.33) was observed in DAP+urea at an altitude of 400 m. Root length (73.70; 70.60 cm) was maximum in NAA 250 mg L^{-1} at an altitude of 400 m in *C. flexuosus* and *C. winterianus* respectively. This study recommends use of DAP+urea for mass cultivation of each species at various altitudes and agro-climatic conditions, thereby ensuring efficient protocol for mass cultivation and genetic heterogeneity.

Keywords: Altitude, Auxins, *Cymbopogon*, Fertilizers, Medicinal and aromatic, Phenotypic plasticity

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Introduction

India spans a diverse geographical expanse with wide variations in latitude, altitude, soil and microclimatic conditions. The Indian Himalayan region harbors approximately 8,000 vascular plants (Singh and Hajra, 1996), including 1,740 medicinal and aromatic plants (MAPs). Uttarakhand comprises about 701 of these MAPs species that have been identified (Samant *et al.,* 1998). This establishes India as a traditional producer and exporter of MAPs species. Aromatic plants are source of essential oils that are useful in different industries. Challenges such as the unavailability of aromatic plants in wild habitats, global scale rising demand, illegal trading, lack of information on crop productivity, and performance in different altitudes have increased the need for intensive agricultural studies (Roosta *et al.,* 2017). This would lead to genetic improvement, and improved organized cultivation in diverse altitudes or regions of India and globally.

Phenotypic plasticity refers to a genotype's ability to express different phenotypes in heterogeneous environments (Mu *et al.,* 2022). Plants exposed to various nutrient availability, altitude, light intensity, microclimate, and soil type can exhibit phenotypic plasticity (Wahl *et al.,* 2001; Li *et al.*, 2020; Ren *et al.,* 2020). Studies have been performed on many aromatic plants in different altitudes (Steiner *et al.,* 2012; Bakhtiari *et al.*, 2019; Ren *et al.*, 2020; Ramos *et al.,* 2021; Mwithiga *et al.*, 2022). Seed production in *Poa alpina* is highest at low altitudes (Steiner *et al.,* 2012). Plant growth and biomass production of *Cardamine pratensis* and *Plantago major* was higher in low altitude (Bakhtiari *et al.,* 2019). Shoot length and biomass increase at high altitudes in *Phragmites austalis* (Ren *et al.,* 2020). Secondary metabolites and essential oil content of *Piper gaudichaudianum and Cymbopogon flexuosus* show variation in response to spatiotemporal differences (Ramos *et al.,* 2021; Mwithiga *et al*., 2022).

Cymbopogon (Family: Poaceae) is a genus of aromatic grasses, essential in the pharmaceutical, perfumery, cosmetic, and food industries. *Cymbopogon* can be found in the altitudional range of 1200 to 2800 m (Negi *et al.,* 2018). The genus comprises 144 species, distributed in tropical and subtropical regions of Asia, Africa and America (Khanuja *et al.,* 2005). Some of the economically important species are *C. flexuosus* (East Indian lemongrass), *C. winterianus* (Citronella grass), *C. citratus* (West Indian lemongrass), and *C. martini* (Palmarosa) (Otify *et al.*, 2022). *Cymbopogon* is primarily propagated through vegetative mode by tillers for cultivation, due to asynchronous flowering, fruiting, limited seed production and dispersal (Kumar *et al.,* 2012). These

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grasses are demanded by several thousand tonnes globally, as they are rich sources of bioactive compounds and essential oils, such as lemongrass oil and citronella oil (Abdelsalam *et al.,* 2023; Dangol *et al.,* 2023). The overcollection, commercial utilization, and increased global scale demand cause *Cymbopogon* to a threat of extinction in wild (Abdelsalam *et al.,* 2021).

Protocol development by micropropagation of *Cymbopogon* is developed by somaclonal variation (Cornescu *et al.,* 2023), suspension culture (Abdelsalam *et al.,* 2023), rhizome culture (Bhattacharya *et al.,* 2010) and somatic embryogenesis and organogenesis (Abdelsalam *et al.*, 2022). But micropropagation is difficult to be performed in fields by growers. Despite the economic significance of the genus *Cymbopogon*, research is mainly concentrated on the essential oil content (Verma *et al.,* 2020; Aungtikun *et al.*, 2021). The application of Farm yard manure and NPK has increased the growth parameters of *C. flexuosus* (Momin and Gajbhiye, 2016). But previous studies have not assessed the extensive propagation protocol (Abdelsalam *et al.*, 2021) and performance of raised seedlings on different altitudes of Uttarakhand, India (Negi *et al.,* 2018). The study aims: (i) to develop a reliable propagation protocol for *C. flexuous* and *C. winterianus* (ii) to assess the phenotypic adaptability of transplanted seedlings in altitudes of varying climatic conditions.

Material And Methods

The research was carried out at the Himalayan Botanical Garden in Khurpatal, Uttarakhand, India. The study site is located at (29°23′09′′ N) longitude and (79°26′02′′ E) latitude. The climate of Khurpatal is characterized by short summers, long cold and snowy winter with average max-min temperature ranging between (20.33–15.66°C). The study focused on the propagation for one year in which harvesting was done in March and September 2021, while the influence of climatic conditions and altitude on transplanted plants of *C. flexuosus* and *C. winterianus* were observed for the second year 2022. Raised seedlings were transplanted across three different altitudes (Fig. 1), Haldwani (400–524 m) representing the tropical zone, Bhujiyaghat (850–1000 m) representing the subtropical zone, and Khurpatal (1600–1900 m) representing the temperate zone, covering a range of climatic conditions (temperature, humidity and rainfall) (Table 1) (Fig. 1).

Potting mixture was prepared with air-dried soil that underwent fumigation using potassium permanganate and formalin. Subsequently, the soil was treated with a solution of insecticide (imidacloprid) and systemic fungicide (bavistin) in a 1:10 ratio with water. The sterilized soil was then subjected to two distinct treatments: incorporation of various fertilizers as per experimental demands, and a control group that remained

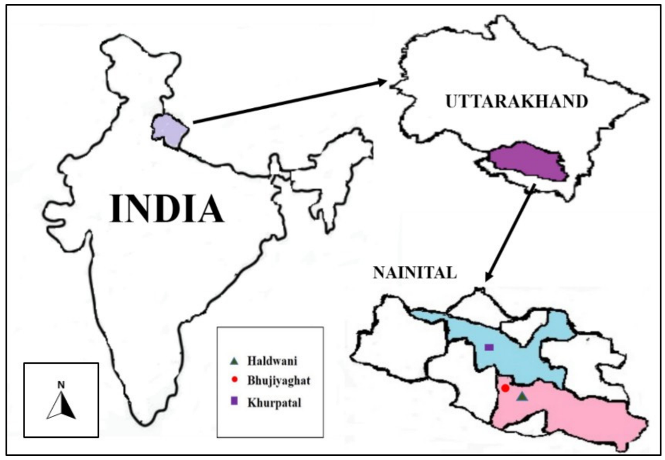

Fig. 1: Map of the study area

untreated. Planting material (stumps) was collected from the Forestry Training Institute (FTI) located in Haldwani during the months of June in 2020. Subsequently, the collected material was transported to the laboratory for further processing. The tillers were detached from the parent plant manually and leaves were removed using pruning scissors. To ensure uniformity, the stumps of both plant species were standardized to a height of 10 cm.

To ensure aseptic conditions, the stumps underwent a sterilization process involving distinct agents: a nonionic detergent (1% tween 20) for a duration of 10 minutes and a fungicide solution (1% bavistin) for 20 minutes (Lodhiyal *et al.,* 2023). Subsequent to sterilization, the stumps underwent rigorous washing with autoclaved double distilled water, and these prepared stumps were deemed suitable for use in the subsequent experimental procedures.

The experimentation was conducted within a controlled greenhouse environment, utilizing both polybags and earthen pots containing sterilized soil. The study was devised in accordance with a randomized block design (RBD), with each treatment group (organic fertilizers, inorganic fertilizers and auxins) consisting of twenty-five stumps in triplicates. These stumps were subsequently transplanted into pots, containing a soil mixture composed of soil, sand, and organic fertilizers in a ratio of 2:1:1. The organic fertilizers employed in the study encompassed manure, organic compost, vermicompost, and a combination of organic compost and vermicompost. Inorganic fertilizer regimen comprised urea (46% mg N kg^{-1}), nitrogen-phosphorus-potassium (NPK 12-32-16 mg N kg^{-1}), di-ammonium phosphate (DAP 18-46-0 mg N kg^{-1}), as well as combinations of NPK with urea and DAP with urea. The solution of inorganic fertilizers (IF) was prepared using autoclaved double distilled water. In addition, the 2 g L^{-1} solution of inorganic fertilizers

Table 1: Meteorological data of study sites

S.No	*Sites*	*Annual Max temp (°C)*	*Avg Min temp (°C)*	*Avg RH*	*Avg Rainfall (mm)*
1.	Haldwani	26.97	21.27	72.11	1.45
2.	Bhujiyaghat	24.24	18.22	65.77	9.77
3.	Khurpatal	20.33	15.76	22.45	30.78

*Avg.= Average, Max= Maximum, Min= Minimum, Temp= Temperature, Rh= Relative humidity. The meteorological data was collected from the forest training institute, Haldwani and GIC, Nainital

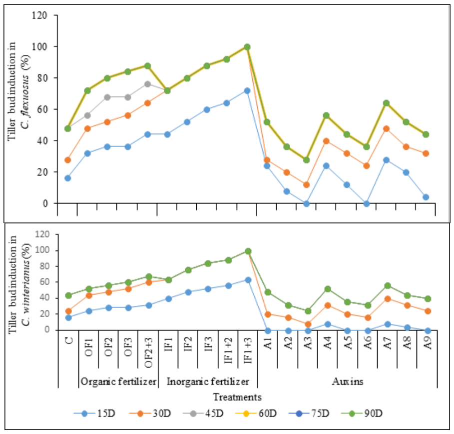

Fig. 2: Tiller bud induction in *Cymbopogon flexuosus* and *Cymbopogon winterianus* from 15-90 days (D= Days, C= Control, OF1= Manure, OF2= Organic compost, OF3= Vermicompost, OF2+3= Organic compost + vermicompost, IF1= Urea, IF2= NPK, IF3= DAP, IF1+2= Urea+NPK, IF1+3= Urea+DAP, A1= IAA 250 mg L^{-1}, A2= IAA 500 mg L^{-1}, A3= IAA 1000 mg L^{-1}, A4= IBA 250 mg L^{-1}, A5= IBA 500 mg L^{-1}, A6= IBA 1000 mg L^{-1}, A7= NAA 250 mg L^{-1}, A8= NAA 500 mg L^{-1}, A9= NAA 1000 mg L^{-1})

were sprayed to the soil (Tanwar *et al.,* 2014). Solutions of auxins were cautiously prepared within the laboratory environment. These solutions encompassed three distinct concentrations (250, 500, and 1000 mg L^{-1}) of three auxins: indole-3-acetic acid (IAA), indole-3-butyric acid (IBA), and naphthalene acetic acid (NAA). The auxins were solubilized with 1.5% ethanol, and a resultant solution volume of 100 ml was attained through the addition of autoclaved double distilled water (Chauhan *et al.,* 2020). The prepared stumps were immersed in their respective auxin solutions for a period of 24 hours and maintained in green house (23–25°C).

Plant growth assessment encompassed a two-year monitoring period, conducted across both greenhouse and field conditions. Triplicate sets of seven randomly selected plants were diligently observed. These observations were conducted to quantify the growth dynamics of both *C. flexuosus* and *C. winterianus* across varied treatment conditions. Growth parameters such as the proportion of tiller bud induction/vegetative growth were computed following (Lodhiyal *et al.,* 2023). Plant length (from soil surface to apical tip) and root length were recorded using a meter tape, while the enumeration of the

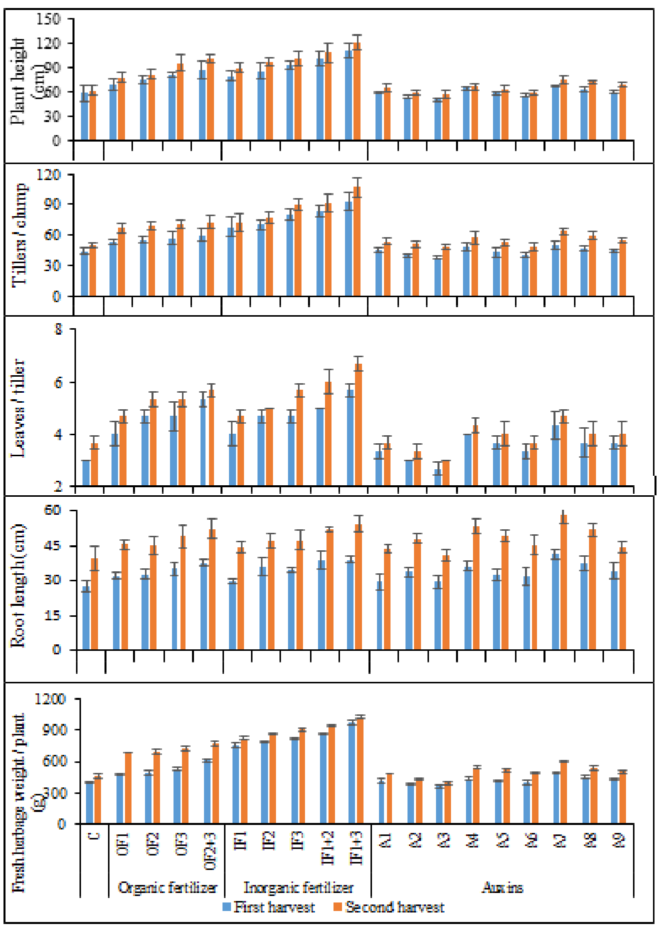

Fig. 3: Growth parameters of first and second harvest of *Cymbopogon flexuosus* (C= Control, OF1= Manure, OF2= Organic compost, OF3= Vermicompost, OF2+3= Organic compost + vermicompost, IF1= Urea, IF2= NPK, IF3= DAP, IF1+2= Urea+NPK, IF1+3= Urea+DAP, A1= IAA 250 mg L^{-1}, A2= IAA 500 mg L^{-1}, A3= IAA 1000 mg L^{-1}, A4= IBA 250 mg L^{-1}, A5= IBA 500 mg L^{-1}, A6= IBA 1000 mg L^{-1}, A7= NAA 250 mg L^{-1}, A8= NAA 500 mg L^{-1}, A9= NAA 1000 mg L^{-1})

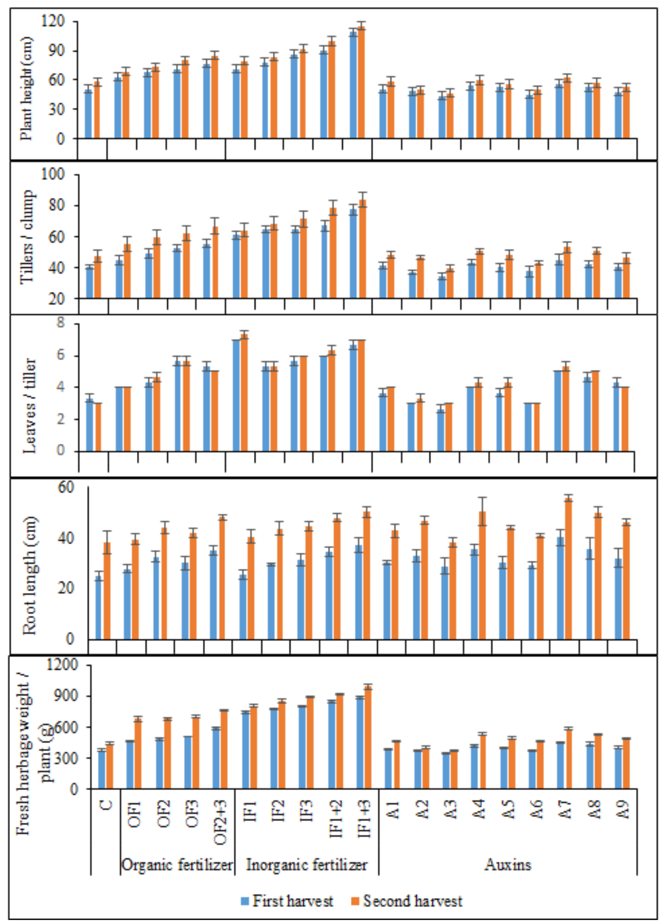

Fig. 4: Growth parameters of first and second harvest of *Cymbopogon winterianus* (C= Control, OF1= Manure, OF2= Organic compost, OF3= Vermicompost, OF2+3= Organic compost + vermicompost, IF1= Urea, IF2= NPK, IF3= DAP, IF1+2= Urea+NPK, IF1+3= Urea+DAP, A1= IAA 250 mg L^{-1}, A2= IAA 500 mg L^{-1}, A3= IAA 1000 mg L^{-1}, A4= IBA 250 mg L^{-1}, A5= IBA 500 mg L^{-1}, A6= IBA 1000 mg L^{-1}, A7= NAA 250 mg L^{-1}, A8= NAA 500 mg L^{-1}, A9= NAA 1000 mg L^{-1})

number of tillers/clumps, number of leaves/tiller and number of seeds/plant were performed by direct counting. Herbage weight/plant was precisely determined by using an electronic weighing balance.

Statistical analysis

Data of plant species was represented in the graphical representation form which was computed using Microsoft excel. Triplicates of twenty-five stumps/replicate were used for analysis of growth parameters of *C. flexuosus* and *C. winterianus*. Statistical significance among the parameters were assessed by using one-way ANOVA (Analysis of variance) at ($p < 0.05$) through SPSS software version 23.0 (SPSS Inc., Chicago, USA).

Results

Results demonstrated promising outcomes, as all treatments induced tiller buds in *Cymbopogon*. The significant success rate of 100% tiller bud induction observed in *C. flexuosus* and *C. winterianus* after thirty days using inorganic fertilizer (IF) combination DAP (diammonium phosphate) and urea (IF1+3). However, using a combination of vermicompost and organic compost (OF2+3) resulted in a more gradual release, inducing 64 and 60% of tiller buds in both species, respectively (Fig. 2). Auxins contribute to a lower degree of growth stimulation in the induction of tiller buds when compared to OF and IF. This further highlights the efficacy of IF combination IF1+3. Moreover, it was observed that *C. flexuosus* displayed early tiller bud induction compared to *C. winterianus*. In 15 days, tiller bud induction reached 88% in *C. flexuosus*, while it was slightly lower at 64% in *C. winterianus* when treated with IF1+3. Specifically, in a span of 15 days, the control group exhibited a meager 16% tiller bud induction in *C. flexuosus* and only 48% of tiller buds were induced in 90 days. However, in *C. winterianus* in a span of 15 days, the control group exhibited 16% tiller bud induction and only 44% of tiller buds were induced in 90 days. Treatments had a significant and greater effect on tiller bud induction ($p \leq 0.05$) than the control (Table S1).

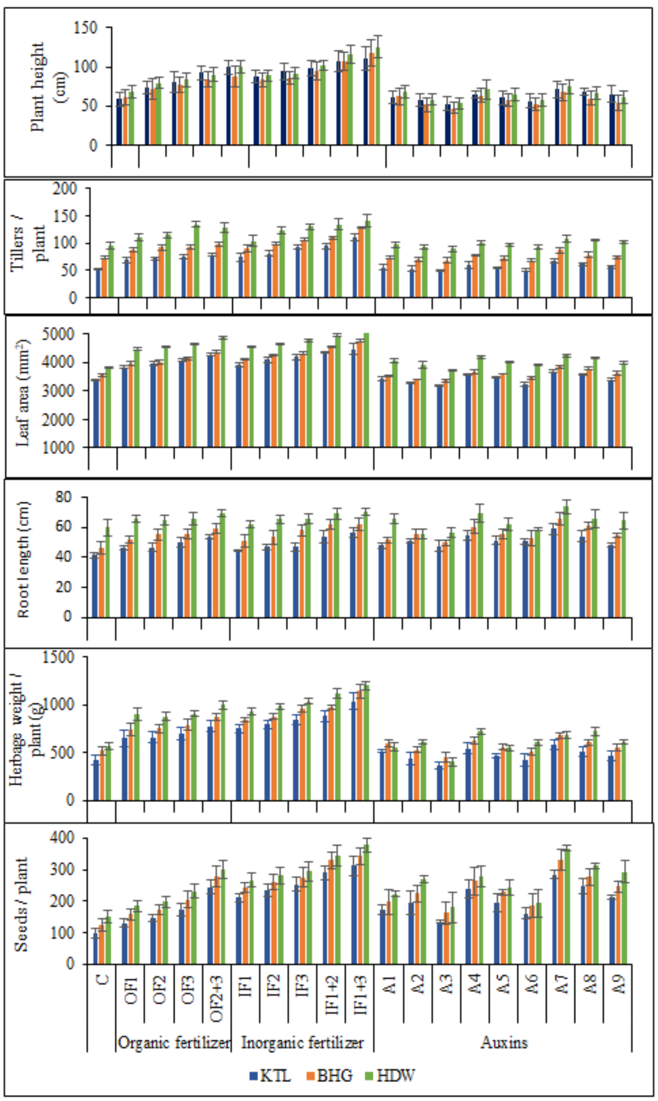

Fig. 5: Growth parameters of transplanted seedlings of *Cymbopogon flexuosus* in three sites (KTL= Khurpatal, 1900 m; BHG= Bhujiyaghat, 900 m; HDW= Haldwani, 400 m)

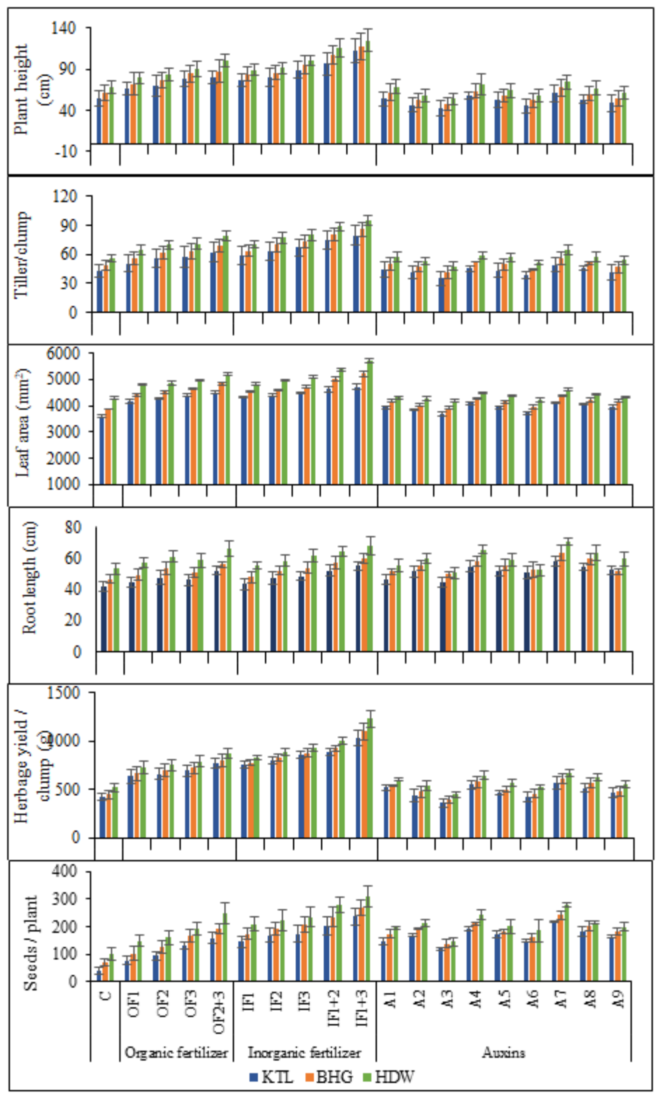

Fig. 6: Growth parameters of transplanted seedlings of *Cymbopogon winterianus* in three sites (KTL= Khurpatal, 1900 m; BHG= Bhujiyaghat, 900 m; HDW= Haldwani, 400 m)

The study included two harvests (March and September), within a greenhouse environment for one year. Application of IF, particularly IF1+3, resulted in significant improvements in growth of both species of *Cymbopogon*. Plant length (PL) (121.00, 115.33 cm), number of tillers per clump (T/C) (107.00, 84), average leaves per tiller (L/T) (6.67, 7.33), and fresh herbage weight (FHW) (1027.96, 990.67 g/clump) were observed maximum in *C. flexuosus* and *C. winterianus* treated with IF1+3, surpassing the performance of other OF, IF, and auxins (Figs 3, 4). The OF2+3 demonstrated positive effects on herbage growth, highlighting their viability as an alternative to IF for stimulating grass growth. However, root length of *C. flexuous* (55.53 cm) and *C. winterianus* (51.00 cm) was maximum in stumps treated with NAA 250 mg L^{-1} (A7), respectively. Treatments had a significant and greater effect on growth parameters except root length ($p \leq 0.05$) than the control (Table S2, S3). These findings indicate the efficacy of IF1+3 in promoting herbage growth and A7 in root growth. This highlights the importance of providing appropriate fertilization and hormonal treatments to optimize the growth and commercialization opportunities of lemongrass and citronella grass.

Seedlings raised up to the second harvest were transplanted at three different altitudes 400 m (Haldwani), 900 m (Bhujiyaghat), and 1900 m (Khurpatal). *C. flexuosus* and *C. winterianus* exhibited best growth response at an altitude of 400 m with respect to growth parameters (Figs 5, 6). Minimum growth response was observed at a high altitude of 1900 m. Altitudes and treatments had a significant and greater effect on the growth of transplanted seedling of both species ($p \leq 0.05$) (Table S4, S5). PL and T/C shows variation from low to high altitude, induced maximum at 400 m when treated with IF1+3 and minimum response was observed in control condition at 1900 m. Control group exhibited the minimum herbage weight at three altitudes. RL (73.70, 70.63 cm) were formed maximum in plants at low altitudes when treated with NAA 250 mg L^{-1} in both species respectively. S/P were formed maximum in plants at low altitudes when treated with IF1+3 in both species respectively. These findings highlight the significance of proper fertilizer management in the cultivation of *C. flexuosus* and *C. winterianus* at various altitudes and agro-climatic conditions.

Discussion

Environmental conditions and growth regulators have an impact on the growth and chemical composition of a plant (Li *et al.,* 2020; Ren *et al.,* 2020). The critical gap in existing literature, which includes propagation protocols for various altitudinal cultivation and adaptation, emphasizes the importance of this study. Bridging these gaps is critical for improving cultivation techniques and ecological understanding, thereby increasing the potential of *Cymbopogon* species at various altitudes.

Effect of OF, IF and auxins on tiller bud induction

Present finding indicated that inorganic fertilizers had the best effectiveness at promoting the induction of tiller buds among all the treatments tested. Slow release of nutrients might be responsible for less tiller bud induction in *C. flexuosus* and *C. winterianus* by organic fertilizers as compared to inorganic fertilizers. On the other hand, Auxins contribute to a lower degree of stimulation in the induction of tiller buds as compared to organic fertilizer and inorganic fertilizer. Abbas and Saeid, (2012) and Khan *et al.,* (2015) reported auxins (IBA and IAA) have no favorable effect on the tiller bud growth of *C. flexuosus* and *C. martinii*. Costa *et al.,* (2020) stated urea and monobasic phosphate increase the tiller growth in *C. winterianus*. Ghatas and Mohamed, (2018) suggested the foliar spray of micronutrients like iron, manganese and zinc for the growth of tillers in *C. citratus*. Among the IF used in the experiment, DAP and urea mixture (IF1+3), was highly effective in promoting tiller buds. This can be due to the presence of nitrogen and phosphorus in DAP+Urea. Nutrient availability, fast release, and accumulation of nutrients in the apical tissues may have induced tiller buds and expedited plant growth by inorganic fertilizers (Wang *et al.,* 2020).

Effect of OF, IF and auxins on plant growth

Growth parameters such as; plant length, tiller/clump, leaves/tiller and herbage weight/clump were induced maximum by foliar spray of 2% of DAP+Urea followed by response of other IF, OF and auxins in *C. flexuosus* and *C. winterianus*. These results are aligned with findings reported by (Mahrouk *et al.,* 2018; Mensah *et al.,* 2021; Mahmoud *et al.,* 2023). A combined dose of NPK and compost tea maximally induced plant height, tillers/clump, fresh and dry biomass and essential oil yeild (Mahrouk *et al.,* 2018). Application of 5 tonnes/hectare of compost induced plant height, tiller/clump and dry herbage weight in *C. schoenanthus,* (Mensah *et al.,* 2021). Deficiency of NPK and water level causes decline in shoot length, tillers growth, chlorosis (leaves discoloration), essential oil content and increase in proline content in *C. winterianus* (Mahmoud *et al.,* 2023). Among the three varieties (Manjusha, Mandakini and Bio-13) of *C. winterianus*, Bio-13 induced the maximum herbage growth, essential oil content, and dry matter yield when treated with leaf compost (Prakash and Adholeya, 2004).

According to Mahrouk *et al.,* (2018) and Costa *et al.,* (2020), higher plant growth by combinatory mixture of nitrogen and phosphorus can be because of their physiological roles. Nitrogen is essential as a component of amino acids, enzymes, and critical energy transmissions molecules such as chlorophyll, ADP, and ATP. Phosphorus is also required for several fundamental metabolic processes in plants, such as cell division, glucose metabolism, photosynthesis, and biological oxidation. Root growth was maximally induced by the application of NAA 250 mg L^{-1} in both species of *Cymbopogon*. This may be attributed to the effective utilization of carbohydrates and nitrogen by the use of auxins (Cline, 1996). Similar results were reported in other aromatic plants; *Rosa damascena* (Amal *et al.,* 2022), *Nepeta cataria* (Allen *et al.,* 2023) and *Nerium odorum* (Kumar *et al.,* 2020). Auxins in concentration concentration-dependent manner are also effective in the seed development of both species of *Cymbopogon*.

Effect of treatments and altitude on phenotypic plasticity of transplanted plants

The present study found that treatment and altitude with different climatic conditions strongly influenced the growth and reproduction of transplanted plants. Plants at low altitudes (400

m) showed maximum growth parameters of *C. flexuosus* and *C. winterianus* such as plant length, tillers per clump, and herbage weight per clump. Whereas comparatively less growth was observed in mid-altitude (900 m) and minimum in high altitude (1900 m). Similar findings in different geographical places were reported by (Kassahun *et al.,* 2011 and Mwithiga *et al.,* 2022; and Rathia *et al.,* 2022). *C. citratus* grown at low altitudes in different regions of Ethiopia showed higher plant length, number of leaves per clump and number of tillers per clump (Kassahun *et al.,* 2011). *C. flexuous* grown at mid and low altitudes in Kenya showed higher plant length, number of leaves per clump and number of tillers per clump whereas growth was lower at high altitudes (Mwithiga *et al.,* 2022).

Leaf area is an important parameter for determining the level of adaptation and acclimatization in a new habitat. Leaf area was found higher in low and mid-altitude. Effects of environment on leaf area were reported by previous researchers. Kassahun *et al.,* (2011) observed highest number of leaves per plant of *C. flexuosus* at low altitudes in Ethiopia. Rathia *et al.,* (2022) observed high variation in leaf length, leaf width, and leaf area of *C. flexuosus* collected from different districts of Chhattisgarh. Reproductive growth i.e. formation of a large number of flowers and seeds show a pivotal role of plant adaptability and survival in different altitude. Growth of seeds in low, mid and high altitude (400 m, 900 m, 1900 m) are the successful adaptive strategy of plants to survive in a new environment. However, Flowering and seed formation were not reported in *C. schoenanthus* by use of compost (Mensah *et al.,* 2021). No study is been reported on the effects of altitude on seed formation of *C. flexuosus* and *C. winterianus*.

The findings could be attributed to the applied nutrients, transplantation at an altitudinal gradient, and microclimatic conditions like temperature, humidity and rainfall (Table 1). Soil condition of study sites could also be a possible reason for variation in morphological and reproductive traits. High altitude area (Khurpatal) is a rocky, mountainous region with poor soil conditions whereas low altitude area (Haldwani) comes under Terai region with comparatively better soil conditions (Tripathi *et al.,* 2009). According to Abdusalam and Li, (2018) and Shaukat *et al.,* (2018) reduced growth of plants at high altitudes may be an adaptation to environmental stress, short growing season and change in rate of tissue nutrient accumulation. Plant growth are associated with supplemented plant nutrients and environmental factors. Production of essential oil and secondary metabolites of *Cymbopogon* are also influenced by nutrients provided, altitude and microclimatic conditions (Mwithiga *et al.,* 2022). The present study did not provide information about the production of essential oil and secondary metabolites, therefore further study in this regard is suggested.

Conclusion

This study examined the impact of organic, and inorganic fertilizers, and auxins, on tiller bud induction and growth of *C. flexuosus* and *C. winterianus*. The effect of treatments and altitude, ranging from 400 to 1900 meters, was also factored into the evaluation of agronomic traits of both species. The results unequivocally demonstrate that the combination of diammonium phosphate (DAP) and urea outperformed other fertilizers and auxins. Furthermore, lower altitudes consistently yielded superior morphological and reproductive traits, while mid and high altitudes showed comparatively subdued growth. These findings show the importance of precise nutrient levels, soil conditions, microclimatic factors (temperature, humidity, rainfall), and altitude in fostering the growth of *C. flexuosus* and *C. winterianus*. This study provides invaluable insights into the potential for mass cultivation, extensive phenotypic adaptability, and the potential for agricultural diversification of these aromatic plants across diverse altitudes in Uttarakhand, India.

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Author Contribution

RG: Collection of plants, experiment performed, data collection, data analysis, graph making, data compilation, and writing of first original draft of manuscript, reviewing and editing; **NL:** Manuscript corrections and finalization; **ST:** Manuscript correction and finalization; **NM:** Preparation of figures, reviewing and plagiarism check.

Conflict Of Interest

The author declares that there is no conflict of interest

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.Supplementary Tables

Table S1: Level of significance of stumps sprouting by using one-way ANOVA

Days SS		Cymbopogon flexuosus					Cymbopogon winterianus				
		df	Mean square	f	Sig.	SS	df	Mean square	f	Sig.	
15	BG	1887.47	18	104.86	21.89	.00	1621.26	18	90.07	47.53	.00
	WG	182.00	38	4.78			72.00	38	1.89		
	Total	2069.47	56				1693.26	56			
30	BG	2300.21	18	127.78	27.28	.00	2359.57	18	141.08	43.94	.00
	WG	178.00	38	4.68			122.00	38	3.21		
	Total	2478.21	56				2661.57	56			
45	BG	1373.05	18	76.28	14.94	.00	1243.57	18	69.08	16.61	.00
	WG	194.00	38	5.10			158.00	38	4.15		
	Total	1567.05	56				1401.57	56			
60	BG	1373.05	18	76.28	14.94	.00	1243.57	18	69.08	16.61	.00
	WG	194.00	38	5.10			158.00	38	4.15		
	Total	1567.05	56				1401.57	56			
75	BG	1373.05	18	76.28	14.94	.00	1243.57	18	69.08	16.61	.00
	WG	194.00	38	5.10			158.00	38	4.15		
	Total	1567.05	56				1401.57	56			
90	BG	1373.05	18	76.28	14.94	.00	1243.57	18	69.08	16.61	.00
	WG	194.00	38	5.10			158.00	38	4.15		
	Total	1567.05	56				1401.57	56			

* signifies that the treatments were significant or non-significant for growth parameter at level of Significance $p \leq 0.05$ where SS= Sum of squares, BG= Between groups, WG= Within groups

Table S2: Level of significance of *Cymbopogon. flexuosus* (6, 12 month) by using one-way ANOVA

Growth parameters SS		*First harvest*					*Second harvest*				
		df	*Mean square*	*f*	*Sig.*	*SS*	*df*	*Mean square*	*f*	*Sig.*	
PL	BG	15986.55	18	888.14	5.95	.00	19832.77	18	1101.82	6.39	.00
	WG	5671.61	38	149.25			6544.48	38	172.22		
	Total	21658.17	56				26377.26	56			
T/C	BG	14099.71	18	783.31	7.66	.00	14358.84	18	797.71	6.34	.00
	WG	3884.00	38	102.21			4778.66	38	125.75		
	Total	17983.71	56				19137.50	56			
L/T	BG	37.05	18	2.05	4.19	.00	53.05	18	2.94	6.72	.00
	WG	18.66	38	0.49			16.66	38	0.43		
	Total	55.71	56				69.71	56			
RL	BG	769.08	18	42.72	1.40	.18	1241.91	18	68.99	1.41	.18
	WG	1157.41	38	30.45			1857.56	38	48.88		
	Total	1926.50	56				3099.48	56			
FHW	BG	2004679.29	18	111371.07	103.17	.00	2020864.56	18	112270.25	100.17	.00
	WG	41017.39	38	1079.40			42586.72	38	1120.70		
	Total	2045696.68	56				2063451.29	56			

*This table signifies that the treatments were significant or non-significant for growth parameter at level of Significance $p \leq 0.05$ where SS= Sum of squares, BG= Between groups, WG= Within groups; PL= Plant length, T/C= Tiller per clump, L/T= Leaves per tiller, RL= Root length, FHW= Fresh herbage weight

Table S3: Level of significance of *Cymbopogon winterianus* (6, 12 month) by using one-way ANOVA

Growth parameters SS		*First harvest*					*Second harvest*				
		df	*Mean square*	*f*	*Sig.*	*SS*	*df*	*Mean square*	*f*	*Sig.*	
PL	BG	17268.36	18	935.35	19.09	.00	19386.84	18	1077.04	30.76	.00
	WG	1908.97	38	50.23			1330.30	38	35.00		
	Total	19177.33	56				20717.15	56			
T/C	BG	8014.21	18	445.23	14.76	.00	8225.57	18	456.97	6.99	.00
	WG	1146.00	38	30.15			2484.00	38	65.36		
	Total	9160.21	56				10709.57	56			
L/T	BG	85.71	18	4.76	22.62	.00	92.03	18	5.11	32.38	.00
	WG	8.00	38	0.21			6.00	38	0.15		
	Total	93.71	56				98.03	56			
RL	BG	833.74	18	46.31	1.75	.07	1175.35	18	65.29	2.38	.09
	WG	1004.47	38	26.43			1042.44	38	27.43		
	Total	1838.21	56				2217.80	56			
FHW	BG	1834864.92	18	101936.94	216.02	.00	1969920.63	18	109440.03	142.07	.00
	WG	17930.94	38	471.86			29271.02	38	770.29		
	Total	1852795.97	56				1999191.66	56			

*This table signifies that the treatments were significant or non-significant for growth parameter at level of Significance $p \leq 0.05$ where SS= Sum of squares, BG= Between groups, WG= Within groups; PL= Plant length, T/C= Tiller per clump, L/T= Leaves per tiller, RL= Root length, FHW= Fresh herbage weight

Table S4: Level of significance of growth parameters of *Cymbopogon flexuosus* by effect of replicates, treatments and altitudes using one-way ANOVA

Growth parameters		*Replicates*					*Treatments*					*Altitudes*				
SS		*df*	*Mean square*	*f*	*Sig.*	*SS*	*df*	*Mean square*	*f*	*Sig.*	*SS*	*df*	*Mean square*	*f*	*Sig.*	
PL	BG	59955.04	56	1070.62	6.61	.00	58812.87	18	3267.38	25.35	.00	4366.68	2	2183.34	4.60	0.01
	WG	18447.40	114	161.81			19589.58	152	128.87			79580.39	168	473.69		
	Total	78402.45	170				78402.45	170				83947.07	170			
T/C	BG	117568.5	56	2099.43	20.4	.00	46747.88	18	2597.10	4.78	.00	67172.97	2	33586.4	90.84	0.00
	WG	11713.01	114	102.74			82533.65	152	542.98			62108.55	168	369.69		
	Total	129281.5	170				129281.53	170				129281.5	170			
LA	BG	3.77	56	674371.7	47.5	.00	2.71	18	1505832.6	18.64	.00	1.04	2	5211398	30.23	0.00
	WG	1615330	114	14169.56			1.22	152	80757.63			2.89	168	172365.1		
	Total	3.93	170				3.93	170				3.93	170			
RL	BG	6856.95	56	122.44	1.88	.00	3415.79	18	189.76	2.65	.00	3006.54	2	1503.27	22.43	0.00
	WG	7405.95	114	64.96			10847.11	152	71.36			11256.36	168	67.00		
	Total	14262.90	170				14262.90	170				14262.90	170			
FHY	BG	6350022	56	113393.2	11.9	.00	5889223.87	18	327179.10	32.20	.00	454439.5	2	227219.7	5.47	0.00
	WG	1083363	114	9503.18			1544162.35	152	10158.96			6978946	168	41541.34		
	Total	7433386	170				7433386.22	170				7433386	170			
SP	BG	695734.1	56	12423.82	4.30	.00	603515.07	18	33528.61	12.09	.00	86196.92	2	43098.46	7.71	0.00
	WG	329254.0	114	2888.19			421473.11	152	2772.84			938791.2	168	5588.04		
	Total	1024988	170				1024988.18	170				1024988	170			

*This table signifies that the treatments, altitudes were significant or non-significant for growth parameter at level of Significance $p \leq 0.05$ where SS= Sum of squares, BG= Between groups, WG= Within groups; SL= Shoot length, RL= Root length, LA= Leaf area, BP= Berry per plant, SB= Seeds per berry.

Table S5: Level of significance of growth parameters of *Cymbopogon winterianus* by effect of replicates, treatments and altitudes using one-way ANOVA

Growth parameters		*Replicates*					*Treatments*					*Altitudes*				
SS		*df*	*Mean square*	*f*	*Sig.*	*SS*	*df*	*Mean square*	*f*	*Sig.*	*SS*	*df*	*Mean square*	*f*	*Sig.*	
PL	BG	67601.24	56	1207.16	2.90	.00	62278.31	18	3459.90	9.96	.00	5147.22	2	2573.61	3.93	.02
	WG	47451.95	114	416.24			52774.88	152	347.20			109905.97	168	654.20		
	Total	115053.1	170				115053.19	170				115053.19	170			
T/C	BG	29629.52	56	529.09	4.00	.00	26439.29	18	1468.85	12.23	.00	3136.04	2	1568.02	6.34	.00
	WG	15059.33	114	132.09			18249.55	152	120.06			41552.80	168	247.33		
	Total	44688.85	170				44688.85	170				44688.85	170			
LA	BG	3.237E7	56	578086.7	38.5	.00	2.295E7	18	1274770.7	17.39	.00	8686846.7	2	4343423.3	28.7	.00
	WG	1709768	114	14997.96			1.114E7	152	73268.10			2.540E7	168	151165.34		
	Total	3.408E7	170				3.408E7	170				3.408E7	170			
RL	BG	6866.86	56	122.62	2.17	.00	3382.37	18	187.91	2.88	.00	3124.36	2	1562.18	25.7	.00
	WG	6434.00	114	56.43			9918.49	152	65.23			10176.51	168	60.57		
	Total	13300.87	170				13300.87	170				13300.87	170			
FHY	BG	6252821	56	111657.5	10.1	.00	5945953.12	18	330330.72	32.10	.00	285595.74	2	142797.87	3.32	.03
	WG	1256885	114	11025.30			1563753.69	152	10287.85			7224111.0	168	43000.66		
	Total	7509706	170				7509706.81	170				7509706.8	170			
SP	BG	471446.4	56	8418.68	4.12	.00	376201.34	18	20900.07	9.69	.00	85827.61	2	42913.80	11.6	.00
	WG	232431.3	114	2038.87			327676.44	152	2155.76			618050.17	168	3678.87		
	Total	703877.7	170				703877.78	170				703877.78	170			

*This table signifies that the treatments, altitudes were significant or non-significant for growth parameter at level of Significance $p \leq 0.05$ where SS= Sum of squares, BG= Between groups, WG= Within groups; SL= Shoot length, RL= Root length, LA= Leaf area, BP= Berry per plant, SB= Seeds per berry.