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Shoot and leaf biomass yields and their phytochemical composition as affected by diverse fertilizer applications and the maturity stages in lemongrass (*Cymbopogon citratus* [DC.] Stapf.) and citronella grass (*C. nardus* [L.] Rendle) cultivated in Southern of Benin Republic

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Abstract

Cymbopogon citratus (lemongrass) and *C. nardus* (citronella grass) are perennial aromatic herbs belonging to the plant family Poaceae. They are prevalent in the semi-temperate and tropical regions of Asia, America and Africa. A strong lemon fragrance, a predominant feature of these grasses, is due to the high citral, linalool and limonene contents in their oil. The redolence of the oil enables its use in soaps, detergents and perfumes. Moreover, lemongrass is predominately used as food. These grasses also find an application in the pharmaceutical industry. A vast array of ethnopharmacological applications of lemongrass and citronella grass exists today. Apart from nutrients such as fats, proteins, fibers and minerals, they also contain various bioactive compounds which may be grouped into alkaloids, terpenoids, flavonoids, phenols, saponins and tannins. The health restorative capacity of them may be ascribed to the diverse secondary metabolites they produce. Despite their importance for centuries in the daily life of people in Benin, little research has been addressed on the agronomical requirements of the crops, particularly those studies on the fertilizer applications tending to the preservation of the environmental balances. Therefore, we have studied in a preliminary survey the effects of diverse fertilizer applications on the crop productivity and phytochemical composition of their essential oil. Early or delayed harvesting of lemongrass and citronella grass affects essential oil and citral content. The objectives of the study were to determine the effects of biological or chemical fertilizer applications and two maturity stages at harvest of lemongrass and citronella grass on dry matter biomass production, essential oil, chemical composition, citral, linalool, citronellol and limonene contents. The crop clones were planted using a randomized complete block design with four replications at two locations (Centre Songhai in Porto-Novo, and Allada, Benin) during two growing years. Shoots and leaves were harvested at 4.5 and 6.5 months after planting. After harvest, the essential oil and chemical composition were studied using the gas chromatography-mass spectrometry (GC-MS) analysis following the hydrodistillation. ANOVA, Kruskal-Wallis non-parametric test and Ward's minimum variance cluster analysis were used to distinguish the treatment, genotypic and maturity stage effects on the quality traits of concern. There were significant effects of fertilizer type and maturity stages on biomass production, essential oil and chemical composition. Biological fertilizer enhanced biomass production and citral content in essential oil in *C. citratus* than in *C. nardus*. Chemical fertilizer improved rather more the biomass yields in *C. nardus*. Lemongrass and *C. nardus* harvested at 6.5 months after planting had significantly higher oil, citral, linalool, citronellol and limonene contents than those harvested at 4.5 months. A total of 35 compounds were detected from all the probes analyzed. In all cases, citral, linalool, citronellol and limonene contents were around 40, 1, 1, and 15% respectively, and independently of the control, biological, chemical fertilizer and maturity stages. The results presented here suggest further prospects or research activities on the crops as well as the measures to boost the actual production of these aromatic species in Benin and West Africa.

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Introduction

Diversity is a condition of a balanced biosphere and its capacities towards evolution. Evolution during the last decades has been characterized by a tendency for genetic uniformity of plant resources (Essou et al., 2017a; Aoudjit-Hayet, 2006). Biodiversity conservation contributes among others to master the techniques of reproduction and propagation of the species. Their development hinges indeed upon the generative and vegetative propagation techniques (Essou et al., 2017a). Biological diversity is the set of all shapes of a living organism and is usually subdivided into three levels: genetic, specific and ecosystemic (Aoudjit-Hayet, 2006). Benin biodiversity is labelled by great species richness, but also a big variability of landscapes, ecosystems and habitats (Sinsin and Owolabi, 2001). Desertification, deforestation, genetic erosion and climate change are the most important factors responsible for the loss of biodiversity as well as the deterioration of soils in the savannah regions (Essou et al, 2017a). The resulting negative effects can be minimized thanks to reforestation programs, even if any success of these measures was tightly subjected to the severe effects of climate change.

Phytonutrients are considered unique substances that are present naturally in plants and have been found to carry disease-preventing possibilities that are not only specific but also powerful. Both kinds of essential and nonessential phytonutrients impart some specific physiological functions. They are described as bioactive food mechanisms, with descriptions of their metabolic and physical processes, related goals, and biomarkers (Zanklan and Baba-Moussa, 2025; Kiani et al., 2022; Shah et al., 2011). However, the future need for nutraceuticals will rely on consumer awareness and the association between diet and disorder. Although a useful functional diet and nutraceuticals have a noteworthy role in the improvement and care of human health, biotechnologists, regulatory toxicologists, diseases and health professionals, nutritionists, and nutraceutical industrialists should deliberately work in an organized way to create suitable rules to deliver therapeutic methods which can improve the health and welfare of human beings with efficacy, purity, and safety (Kiani et al., 2022). Innovative dietary methodologies have improved the developing theory of nutraceutical and designer foods by merging various functional ingredients to attain health assistance (Kiani et al., 2022). Dietary enrichment through nitrification provides significant health benefits due to many plant-based non-nutritive components, i.e., phytoconstituents. Plant-based therapies have, so far, appeared to be an effective strategy to address metabolic and immune dysfunction (Vélez et al., 2022). They have been used as therapeutic agents since the earliest times, in both systematized and disorganized forms (Zanklan and Baba-Moussa, 2025; Sharma et al., 2020). Plants have been well-known to be strong therapeutic agents due to the presence of some nutritional minerals and vitamins, as well as the presence of non-nutritional components including fibers and active phytochemicals, such as the terpenoids, flavonoids, lignins, sulfides, plant sterols, polyphenolics, coumarins, carotenoids, and saponins (Ali et al., 2022; Kiani et al., 2022; Kiloni et al., 2022). Plant foods are a vital source of two kinds of mixtures: polyphenolic and phenolic (Kiani et al., 2022). These phytochemicals, namely flavonoids and polyphenols, are present in the seeds of fruits and skin, but leaves frequently supply an abundant source of phenolics (Essou et al., 2017b). Aromatic plants are considered to be a potentially beneficial antioxidant source when it is processed under optimal conditions. Additionally, their leaves are a rich source of several bioactive substances, including flavonoids, terpenoids, saponins, phenols and tannins, which provide lemongrass leaves with their distinctive flavour (Sharma et al., 2020).

Lemongrass (*Cymbopogon citratus*) and citronella grass (*C. nardus*) are aromatic perennial tall grass with rhizomes and densely tufted fibrous roots from the

Poaceae plant family. They have short underground stems, ringed segments and thick, slightly leathery green leaves in dense clusters (Sharma et al., 2020; Silva et al., 2011). They are native to Southern Asia and grown in other tropical and subtropical countries like Africa (Spriha and Kumar, 2021; Silva et al. 2011). Lemongrass and citronella grass oils are extracted through the process of steam distillation from the dried leaves of the plant. They have a thin consistency and a pale or bright yellow colour. It has a strong, fresh, lemony, and earthy scent (Spriha and Kumar, 2021; Silva et al., 2011). The plant tops are upright and stout, up to 1.8 m long. Blades are long, grass green, flat, up and around the edges; ligule is very short; sheaths are terete, wider and clasp closely at the base; sheaths of the barren shoots are thick and separating. They are large flowering plants for short days. The inflorescence is about a meter-long spike. Flowers are borne on decompound spathulate; 30 to more than 60 cm long panicles (Spriha and Kumar, 2021; Haque et al., 2018; Silva et al., 2011). The genus *Cymbopogon* contains more than 50 native grass species from tropical Asia and Southern India (Spriha and Kumar, 2021). The best-known cultivated species are ornamental lemongrass (*Cymbopogon citratus*) and citronella grass (*C. nardus*), which are widely used in Asia and Africa, even though it is on a very small scale. These lemon-scented plants add flavour to soups, curries, teas, and other beverages (Majewska et al., 2019). The dominant feature of these grasses is the high citral (geranial + neral) content in their oil, which gives it a heavy lemon scent. The oil's redolence allows it to be used in soaps, detergents, and other products. The raw material used to make ionone, which is used to make vitamin A is also used. Lemongrass and citronella essential oils have more than eighty components, of which the most important ones are citronellal, geranial, neral and limonene. They are present at high concentrations in the oil and are responsible for the repellent properties of the oils. The composition of the essential oils is complex, although there were several main components in the oils and some waxes are presented in the extracted oils (Silva et al., 2011). Lemongrass and citronella are very important in the life habits of people in the tropical and subtropical regions of Africa. These species have been used for ages by humans for diverse purposes. Thus, lemongrass shoots and leaves are indicated in the treatment of a large range of pathologies, while citronella is a producer of insecticides locally. Although their importance to humans, those crops are maintained on the family small-scale cultivation. There is a real risk to genetic drift, and little is known about the use of diverse fertilizers (biological and chemical) for the cultivation of both species in Benin and West Africa.

The main objectives of the work reported here were therefore to: (i) grow local varieties of lemongrass and citronella under diverse fertilizer applications; (ii) estimate how the fertilizers affect the shoot and leaf yield; (iii) assess the effects of the fertilizers on the oil content and the essential compound composition of the biomass produced; and (iv) study the impact of different shoot and leaf harvest date upon the biomasses produced, as well as their oil content and essential compound composition. To sum up, the goal of the surveys was to study the effects of the: genotype (accession), different treatments (application of biological or chemical fertilizer, and without any fertilizer), harvest date, as well as their possible interactions on the biomass yield and its oil content and essential compound composition to identify the most suitable method that will increase the genetic parameters and quality traits evolved. This could help farmers in the production practices of both crops and could ensure sustainable agriculture as well as the promotion of such crops which could contribute to maintaining biodiversity.

Materials and methods

Study area

The study was carried out in the Benin Republic covering 114763 km2. It is located between the latitudes 6°15'N and 12° 25'N, the longitudes 0°40'E and 3°45'E, and is constituted of three main climatic zones: the Guinean, Guineo-Sudanese and Sudanese zones (MEPN, 2008). The climate is of type subequatorial in the South, tropical humid of transition in the Middle part and tropical dry in the North of the country. The present study took place in the Guinean zone, especially in the “Centre Songhai” in Porto-Novo (6°29’50’ latitude N, 2°36’15’’ longitude E) and in Allada (6°39’55’’ latitude N, 2°09’08’’ longitude E).

Plant materials

The plant materials consisted of two lemongrass and two citronella grass clones collected from farmers in the Southern of Benin Republic in West Africa. They were named CC1, CC2, CN1 and CN2, respectively. They represent the most promising accessions in terms of productivity and are frequently used by populations for their nutraceutical as well as pharmacological attributes.

Methodology and experimental protocol

The experimental study was laid out as a randomized complete block design (RCBD) with four replications at each location. Two locations were considered. Three treatments were equally applied to each genotype at both locations. They represented the application of biological, and chemical fertilizers and the control. The biological fertilizer was set up from poultry droppings and pig manure. Poultry droppings contained labile substances and had a basic pH (7.92) with a correct nitrogen content (l.9%). These labile substances were applied at the rate of 0.167% of soil. Pig manures were more lignified than droppings and had a pH of 7.7. Its nitrogen content was good (1.9%) and it was brought at the rate of 0.333% of soil. The droppings plus manure intake totalized 0.500% of the soil weight. The chemical fertilizer applied is the NPK at the general formula of 15-15-15. It is suitable to emphasize its acidic pH (3.7), which represents a disadvantage for its agricultural use. It was brought at the rate of 0.013% of the soil weight. The biological and chemical fertilizers were applied as suggested by Mallouhi and Bris (1995) for the same genotypes or clones. The original soil (Control) was ferritic, sandy loam, and well-stirred before the planting. The fertilizers were introduced into the soil 8 days before the planting date. The soil was well-stirred and -watered before the planting date of the clones. Each plot consisted of four lines, and on each line, ten clones were planted. Lines were 0.50m separated and the between-plants on each line was 0.25m. Irrigation was applied whenever it was not rained at both locations. Weeds were removed manually and regularly during the growth and development stages of the crop. The experimentation lasted six and a half months at both locations. Two harvest dates of shoots and leaves were done, namely by four and a half and by the end of the growth and development time, respectively. After every harvest, the fresh and dry biomass yields were determined. The fresh samples were dried in the oven at 50^{o}C until the weight no longer varied. Each probe of 500g dry biomass was taken to extract the oil contained in harvested shoots and leaves per hydro-distillation. The phytochemical composition of the oil extracted was assessed per Gas Chromatography–Mass Spectrometry (GC - MS). For each plot, the shoot and leaf biomasses were harvested from the central rows to avoid the border effects. The oil extraction was accomplished two times for the plot, and the results were averaged on a plot basis. It was the same procedure for the phytochemical composition examination of the essential oil. The trials were conducted over two growing years.

The recorded traits from the genotypes and the procedure of measurements are listed in Table 1.

Table 1. Morpho-agronomic and quality traits recorded, codes, measurement units and measurement procedures.

Character	Code	Measurement unit and measurement/sampling procedure
Shoot and leaf fresh biomass yield	SLFBY	(kg/ha) – Weighted immediately after harvest
Shoot and leaf dry matter biomass yield	DBY	(kg/ha) – Weighted after drying in the oven at 50^{o}C for many days
Oil harvest index	OHI	(%) – oil content reported to dry matter biomass used for its extraction
α- pinene	AP	(%) – measured as the percentage of essential oil
β- myrcene	BM	(%) – measured as the percentage of essential oil
6-methyl-5 hepten-2-one	MHO	(%) – measured as the percentage of essential oil
Sabinene	SAB	(%) – measured as the percentage of essential oil
β- pinene	BP	(%) – measured as the percentage of essential oil
β- ocimene	BO	(%) – measured as the percentage of essential oil
1-octyn-3-ol	OC	(%) – measured as the percentage of essential oil
Octanal	OCT	(%) – measured as the percentage of essential oil
α- phellandrene	APE	(%) – measured as the percentage of essential oil
α- terpinene	AT	(%) – measured as the percentage of essential oil
p- cymene	PC	(%) – measured as the percentage of essential oil
1,8-cineole	CIN	(%) – measured as the percentage of essential oil
Limonene	LIM	(%) – measured as the percentage of essential oil
δ- terpinene	DT	(%) – measured as the percentage of essential oil
Linalool	LIN	(%) – measured as the percentage of essential oil

Character	Code	Measurement unit and measurement/sampling procedure
Myrcenol	MYR	(%) – measured as the percentage of essential oil
Citronellal	CIT	(%) – measured as the percentage of essential oil
1-pentanol, 5-cyclo-propylidene	PCPE	(%) – measured as the percentage of essential oil
3-undecyne	UDY	(%) – measured as the percentage of essential oil
Cis-β-terpineol	CBT	(%) – measured as the percentage of essential oil
Decanal	DEC	(%) – measured as the percentage of essential oil
Citronellol	CIO	(%) – measured as the percentage of essential oil
Nerol	NO	(%) – measured as the percentage of essential oil
Neral	NA	(%) – measured as the percentage of essential oil
Geranial	GA	(%) – measured as the percentage of essential oil
Citral	CAL	(%) – measured as the percentage of essential oil
Undecanal	UDA	(%) – measured as the percentage of essential oil
(Z)-γ-farnesene	ZYFE	(%) – measured as the percentage of essential oil
α- humulene	AHE	(%) – measured as the percentage of essential oil
α- farnesene	AFE	(%) – measured as the percentage of essential oil
Isocaryophyllene	ICE	(%) – measured as the percentage of essential oil
γ- cardinene	YCE	(%) – measured as the percentage of essential oil
T-cardinol	TCO	(%) – measured as the percentage of essential oil
α- cardinol	ACO	(%) – measured as the percentage of essential oil
Other compounds	OTO	(%) – measured as the percentage of essential oil

Statistical analysis

Statistical analysis of the data was achieved by the software Plabstat (Utz, 2011), JMP version 7 (SAS Institute NC, 2007) and MINITAB 19 (2019). Shoot and leaf yields and the phytochemical composition of their oil were subjected to one- and two-way analyses of variance at 5%. The Fisher-Snedecor test was applied to analyze the relative importance of the factors Year, Location, Treatment, Genotype and Harvest date as well as of their interactions in the observed variability.

Parameter means were compared between Years, Locations, Treatments, Genotypes and Harvest dates by the test of Tukey-Kramer implemented in either JMP or MINITAB 19. The interactions between the various provenances (genotypes or accessions), treatments and harvest dates were completed using the software MINITAB version 19. The Kruskal-Wallis test implemented in MINITAB 19 was used for the comparison of the treatments, accessions and the two biomass harvest dates studied. This test permitted to identify the best treatment as well as the best accession and harvest date driving to higher rates of biomass production and better phytochemical composition (citral, linalool, citronellol and limonene contents in the essential oil) in the neglected species of concern in the current survey.

Following El-Hendawy (2004), all the data were converted to treatment (fertilizer) or maturity stage effect indices before cluster analysis (with JMP 7) to allow comparisons among genotypes for the effects of treatment or cutting date (maturity stage – harvest date) on the shoot and leaf biomass dry matter yield and oil phytochemical composition (citral, linalool, citronellol and limonene contents) by using multiple agronomic parameters. An effect of treatment or harvest date index was defined as the observation divided by the average of the controls. Cluster group ranking numbers can be assigned to cluster groups based on cluster means and was used to score genotypes. Cluster analysis followed the methods described by Jollife et al. (1989). Cluster group rankings were obtained based on Ward's minimum variance cluster analysis of the averages of the treatment or harvest date indices for six parameters at vegetative stage (i.e., shoot and leaf biomass dry matter yield, essential oil harvest index, citral, linalool, citronellol and limonene contents). Cluster group rankings were obtained based on Single-Link cluster analysis of the means. All procedures are described fully in the JMP User's Guide (SAS Institute, 2007). The cluster group rankings were obtained from the average of means of the multiple parameters in each cluster group. If a genotype scored the minimum by the hierarchical cluster analysis for a given treatment or maturity stage, it was considered as well influenced by the given treatment or harvest date rather than others.

Results

Study of the phenotypic and genotypic variabilities using descriptive statistics, one-way and two-way ANOVA

The results of the descriptive statistics are shown in Table 2. All 38 traits measured showed a high range (minimum and maximum) and coefficient of variation It was around 40% for all the characters under investigation. The coefficient of variation ranged from 19.23 (for OHI: essential oil content in the shoot and leaf dry matter harvest index) to 67.76 (for OTO: other compounds in the essential oil). The variability was then very high, even though the trials took place in two locations, two years and were laid out in a randomized block design with four replications at each location.

Table 2. Descriptive statistics (range, mean, standard deviation [SD] and coefficient of variation, CV) for 38 quantitative morpho-agronomic and essential oil quality traits in *Cymbopogon citratus* and *C. nardus* evaluated over two years and two locations in each year.

Trait	Range		Mean	SD	CV (%)
	Minimum	Maximum			
SLFBY	311.900	1994.800	920.800	451.600	49.05
DBY	64.730	454.270	199.800	98.010	49.05
OHI	1.040	4.200	2.595	0.498	19.23
AP	0.020	2.330	0.823	0.446	54.180
BM	0.190	15.360	7.205	3.026	42.000
MHO	0.010	1.120	0.526	0.220	41.92
SAB	0.100	7.000	3.398	1.414	41.620
BP	0.090	6.370	3.074	1.597	41.110
BO	0.070	4.620	2.209	0.897	40.630
OC	0.010	0.400	0.190	0.077	40.540
OCT	0.040	2.720	1.334	0.554	41.530
APE	0.000	0.200	0.086	0.036	42.400
AT	0.010	0.300	0.143	0.057	40.170
PC	0.010	0.990	0.479	0.198	41.270
CIN	0.060	4.660	2.184	0.917	41.990
LIM	1.400	19.060	10.954	3.117	28.450
DT	0.050	3.530	1.701	0.687	40.430
LIN	0.010	1.890	0.869	0.395	45.42
MYR	0.010	0.200	0.094	0.038	40.600
CIT	1.540	29.830	10.788	5.724	53.050
PCPE	0.010	10.090	2.826	1.911	67.64
UDY	0.070	5.180	2.475	0.999	40.370
CBT	0.000	0.500	0,096	0.044	45.810
DEC	0.020	0.720	0.343	0.138	40.430
CIO	0,030	2.040	1.038	0.428	41.23
NO	0.030	1.920	0.936	0.385	41.210
NA	5.320	26.050	13.774	3.747	27.21
GA	8.110	42.830	25.100	5.837	23.25
CAL	16.600	68.880	38.874	9.435	24.27
UDA	0.020	0.740	0.361	0.146	40.500
ZYFE	0.000	0.160	0.079	0.033	42.930
AHE	0.000	0.610	0.299	0.124	41.550
AFE	0.000	0.130	0.062	0.026	42.980
ICE	0.030	2.020	0.945	0.393	41.640
YCE	0.000	1.980	0.947	0.378	39.950
TCO	0.000	0.160	0.075	0.030	40.280
ACO	0.000	0.390	0.189	0.076	40.500
OTO	0.030	9.950	4.385	2.971	67.76

As most proved in previous surveys in *Cymbopogon citratus* and *C. nardus*, the citral (CAL), linalool (LIN), citronellol (CIO) and limonene (LIM) contents are the important compounds of their essential oil, so the further report of interest here was limited to those compounds together with the shoot and leaf biomass dry matter yields (DBY) as well as their essential oil harvest index (OHI).

The results from the one-way and two-way ANOVA realized on the data of the present survey are shown in Tables 3 and 4, as well as in Figures 1-4. Results show that the main sources of variation concerning yield and its components such as the citral, linalool, citronellol and limonene contents are the year, location, treatment, genotype and maturity stage (Tables 3 and 4, Figs. 1-4). The variance components of those characters are very high, demonstrating the high variability existing in the materials used. These variations are followed by interactions, i.e. Genotype × Treatment, Genotype × Maturity Stage (Figs 1-4). Out of the analysis, it can be concluded that the main effects of the biological fertilizer (BioF) are to reduce the shoot and leaf biomass dry matter production, particularly in citronella grass (*Cymbopogon nardus*). The chemical fertilizer (CheF) gave contrary results by enhancing the dry matter yield apart from *Cymbopogon nardus* genotypes (Fig. 1A). In general, the main effects of genotype are enough clear (Fig. 1A). CC1 and CC2 biomass yields are lower and reduced by any treatment whenever compared to CN1 and CN2, which yields were improved by the effects of genotype (Fig. 1A). The same figures are observed according to the main effects of the maturity stages (Fig. 1B). The biomass yields were higher by the second harvest date (SecHa) than the first (FirHa) (Fig. 1B). The genotype main effects were higher by citronella grass than by lemongrass. Interactions Genotype × Treatment and Genotype × Maturity Stage showed the same trend in importance regardless of the clone considered (Tables 3 and 4, Fig. 1B). For the character "essential oil harvest index" (OHI), the effects of interaction Genotype × Treatment were high. They were high and significantly positive for CC1, CC2 (under biological fertilizer applications), CN1 and CN2 (under chemical fertilizer applications and the control, respectively) (Fig. 2). Under biological fertilizer applications, the main effects for treatment were low contrarily to the chemical fertilizer. For "OHI", the effects of genotype were significantly low for CC1 and CC2 (*C. citratus*) and high for CN1 and CN2 (*C. nardus*) (Fig. 2A). The same trends were observed for the effects of maturity stages on "OHI". Analyzing the means for citral content in biomass dry matter yield (DBY), it was noted that biological fertilizer dropped down the citral content in biomass, while no variation was shown for the control. The main effects of genotype resulted in an augmentation of citral content in DBY, notably with lemongrass (CC1 and CC2) (Fig. 3). A reduction was observed rather in citronella grass (CN1 and CN2) as shown in Fig. 3. The citral content in DBY was higher in all genotypes at the end of the developmental stage (6.5 months after planting) than by the first harvest date (4.5 months after planting). Interactions were not high (Fig. 3B). Also, with relevance to the citral content in DBY, the main effects for treatment let deduce a negative action of biological fertilizer, whereas, positive effects were generated from chemical fertilizer applications and the control.(Fig. 3). Generally, *C. citratus* shoots and leaves delivered more citral than those of *C. nardus*. The interactions Genotype × Treatment were however low (Fig. 3A). Citral contents were higher by the end of the developmental stages (6.5 months after planting) than by the middle time (4.5 months after planting). Interactions were also low for citronellol (Fig. 4B). The biological fertilizer applications gave better responses than chemical fertilizer. No strong variation was noted with the control (Fig. 4A). The first harvest (FirHa) delivered here better outputs by increasing the citronellol content in DBY than by the end of the trials (6.5 months after planting) (Fig. 4B).

Kruskal-Wallis non-parametric test to analyze the effects of genotypes, diverse treatments and harvest dates on biomass dry matter yield and its phytochemical composition

Instead of comparing mean values of the four populations or clones analyzed using a one-way or two-way ANOVA, the Kruskal-Wallis test was applied to detect the best treatment, harvest date and genotype with concerns to the most common parameters: shoot and leaf dry matter biomass yield (DBY), essential oil harvest index (OHI), citral (CAL), linalool (LIN), citronellol (CIO), limonene (LIM) contents in essential oil. According to this test, H' indicates if the median values of the given factor are equal or not, and allows so their classifying.

Effects of diverse fertilizers and maturity stages on shoot and leaf dry matter yields (DBY)

The sample medians for the three treatments (considering the trait DBY) were 134.7, 143.1 and 298.7 for the control, the biological fertilizer and the chemical fertilizer, respectively. The z value for the control is -7.96, the smallest value of z. This size indicated that the mean rank for this treatment differs least from the mean rank for all observations. Moreover, the mean rank of this treatment (128.8) was lower than the mean of the observation set (192.5), and the z value is negative (z = -7.96). The mean rank for the treatment "Chemical Fertilizer (CheF)" is higher than in the whole experiment, with a positive z value (z=15.18) (Table 5).

Table 3. One-way ANOVA (Year, Location, Treatment, Maturity Stage or Clone effects) for Shoot and Leaf Biomass Dry Matter Yield (DBY), and its Essential Oil Harvest Index (OHI) , citral (CAL), linalool (LIN), citronellol (CIO) and limonene (LIM) in *Cymbopogon* species cultivated in Benin.

Trait	Source of Variation	Degree of Freedom	Sum of Square	Mean of Square	F Ratio	Prob>F*
	Year effects	1	29499	29499	3.09	0.089
	Residuals	382	3649289	9553	-	-
	Total variance	383	3678788	-	-	-
	Location effects	1	313944	313944	35.640	0.000
	Residuals	382	3364844	8808	-	-
	Total variance	383	3678788	-	-	-
	Treatment effects	2	2385016	1192508	351.180	0.000
	Residuals	381	1293772	3396	-	-
	Total variance	383	3678788	-	-	-
DBY	Clone effects	3	590791	196930	24.230	0.000
	Residuals	380	3087997	8126	-	-
	Total variance	383	3678788	-	-	-
	MS** effects	1	6667	0.690	0.690	0.405
	Residuals	382	3672121	-	-	-
	Total variance	383	3678788	-	-	-
	Year effects	1	8.219	8.219	36.040	0.000
	Residuals	382	87.113	0.228	-	-
	Total variance	383	95.333	-	-	-
	Location effects	1	13.643	13.643	63.800	0.000
	Residuals	382	81.690	0.214	-	-
	Total variance	383	95.333	-	-	-
	Treatment effects	2	13.597	6.798	31.690	0.000
OHI	Residuals	381	81.736	0.215	-	-
	Total variance	383	95.333	-	-	-
	Clone effects	3	5.836	1.945	8.260	0.000
	Residuals	380	89.497	0.236	-	-
	Total variance	383	95.333	-	-	-
	MS** effects	1	0.028	0.028	0.110	0.739
	Residuals	382	95.305	0.249	-	-
	Total variance	383	95.333	-	-	-
CAL	Year effects	1	759,7	759,7	8,71	0,003
	Residuals	382	33334,8	87,3	-	-
	Total variance	383	34094,5	-	-	-
	Location effects	1	222.000	222.000	2.500	0.114
	Residuals	382	33872.500	88.700	-	-
	Total variance	383	34094.500	-	-	-
	Treatment effects	2	1920.700	960.300	11.37	0.000
	Residuals	381	32173.800	84.400	-	-
	Total variance	383	34094.500	-	-	-
	Clone effects	3	5180.000	1726.700	22.69	0.000
	Residuals	380	28914.400	76.100	-	-
	Total variance	383	34094.500	-	-	-
	MS** effects	1	4285.500	4285.500	54.92	0.000
	Residuals	382	29809.000	78.000	-	-
	Total variance	383	34094.500	-	-	-

Table 3. Continued

Trait	Source of Variation	Degree of Freedom	Sum of Square	Mean of Square	F Ratio	Prob>F*
	Year effects	1	3.085	3.085	20.800	0.000
	Residuals	382	56.671	0.148	-	-
	Total variance	383	59.756	-	-	-
	Location effects	1	0.447	0.447	2.880	0.091
	Residuals	382	59.309	0.155	-	-
	Total variance	383	59.756	59.756	-	-
	Treatment effects	2	3.276	1.638	11.050	0.000
	Residuals	381	56.481	0.148	-	-
	Total variance	383	59.756	-	-	-
LIN	Clone effects	3	0.580	0.193	1.24	0.294
	Residuals	380	59.176	0.156	-	-
	Total variance	383	59.756	-	-	-
	MS** effects	1	10.435	10.435	80.82	0.000
	Residuals	382	49.322	0.129	-	-
	Total variance	383	59.756	-	-	-
	Year effects	1	3.981	3.981	22.97	0.000
	Residuals	382	66.205	0.173	-	-
	Total variance	383	70.186	-	-	-
	Location effects	1	0.611	0.611	3.360	0.068
	Residuals	382	69.575	0.182	-	-
	Total variance	383	70.186	-	-	-
	Treatment effects	2	3.597	1.798	10.29	0.000
	Residuals	381	66.589	0.175	-	-
CIO	Total variance	383	70.186	-	-	-
	Clone effects	3	0.639	0.213	1.160	0.323
	Residuals	380	69.547	0.183	-	-
	Total variance	383	70.186	-	-	-
	MS** effects	1	11.964	11.964	78.50	0.000
	Residuals	382	58.222	0.152	-	-
	Total variance	383	70.186	-	-	-
	Year effects	1	129.500	129.500	13.780	0.000
	Residuals	382	3590.830	9.400	-	-
	Total variance	383	3720.330	-	-	-
	Location effects	1	32.920	32.92	3.410	0.056
	Residuals	382	3687.410	9.650	-	-
	Total variance	383	3720.330	-	-	-
	Treatment effects	2	259.930	129.970	14.310	0.000
	Residuals	381	3460.400	9.080	-	-
LIM	Total variance	383	3720.330	-	-	-
	Clone effects	3	40.220	13.410	1.380	0.247
	Residuals	380	3680.120	9.680	-	-
	Total variance	383	3720.330	-	-	-
	MS** effects	1	561.970	561.97	67.97	0.000
	Residuals	382	3158.360	8.270	-	-
	Total variance	383	3720.330	-	-	-

* (Prob<0.05): Significant factor effect according to F-test at 5% threshold; ** Maturity stage: harvest date.

Table 4. Two-way ANOVA for shoot and leaf biomass dry matter yield and its citral (CAL), linalool (LIN), citronellol (CIO) and limonene (LIM) contents of essential oil at two maturity stages in lemongrass and citronella grass cultivated in Benin.

Trait	Source of Variation	Degree of Freedom	Sum of Square	Mean of Square	F Ratio	Prob>F*
	Year	1	29499	29499	31.650	<0.0001
	Location	1	313944	313944	336.870	<0.0001
	Treatment	2	2385016	1192508	1279.580	<0.0001
	Clone	3	590791	196930	211.310	<0.0001
DBY	MS**	1	6667	6667	7.150	<0.0001
	Interactions	144	3622553.700	25156.600	106.970	<0.0001
	Residuals	239	56234.300	235.300	-	-
	Total variance	383	3678788.100	-	-	-
	Year	1	8.219	8.219	64.530	<0.0001
	Location	1	13.642	13.642	107.100	<0.0001
	Treatment	2	5.836	1.945	15.270	<0.0001
	Clone	3	0.027	0.020	0.220	0.641
OHI	MS**	1	78.276	0.543	7.617	<0.0001
	Interactions	144	17.056	0.071	-	-
	Residuals	239	47.385	0.127	-	-
	Total variance	383	95.332	-	-	-
	Year	1	759.700	759.700	13.130	<0.0001
	Location	1	222.000	222.000	3.840	0.051
	Treatment	2	1920.700	960.300	16.590	<0.0001
	Clone	3	51.800	1726.700	29.840	<0.0001
CAL	MS**	1	4285.500	4285.500	29.840	<0.0001
	Interactions	144	25979.154	180.411	74.050	<0.0001
	Residuals	239	8115.302	33.955	5.313	-
	Total variance	383	34004.456	-	-	-
	Year	1	3.085	3.085	27.560	<0.0001
	Location	1	0.446	0.446	3.990	0.046
	Treatment	2	3.275	1.637	14.630	<0.0001
	Clone	3	0.580	0.193	1.730	0.045
LIN	MS**	1	10.434	10.440	93.210	<0.0001
	Interactions	144	42.725	0.296	-	-
	Residuals	239	17.030	0.071	-	-
	Total variance	383	59.756	-	-	-
	Year	1	3.981	3.981	30.190	<0.0001
	Location	1	0.611	0.611	4.630	0.032
	Treatment	2	3.596	1.798	13.64	<0.0001
	Clone	3	0.639	0.213	1.620	0.185
CIO	MS**	1	11.963	11.963	0.185	<0.0001
	Interactions	144	50.930	50.930	0.353	-
	Residuals	239	19.255	19.255	4.390	-
	Total variance	383	5570.185	-	-	-
	Year	1	129.503	129.503	18.03	<0.0001
	Location	1	32.924	4.580	4.580	0.033
	Treatment	2	259.935	129.967	18.090	<0.0001
	Clone	3	40.216	13.405	1.870	0.135
LIM	MS**	1	561.972	561.972	78.230	<0.0001
	Interactions	144	2535.941	17.610	3.553	<0.0001
	Residuals	239	1184.391	-	-	-
	Total variance	383	3720.330	-	-	-

* (Prob<0.05): Significant factor effect according to F-test at 5% threshold; ** Maturity stage: harvest date.

Table 5. Kruskal-Wallis non-parametric test of biomass yield and phytochemical composition of essential oil comparisons in two *Cymbopogon* species grew in Benin under diverse fertilizer applications.

Trait	Factor		N†	Median	Average Rank	Z
DBY	Treatment	Control	128	134.7	128.8	-7.96
		BioF*	128	143.1	134.6	-7.23
		CheF*	128	298.7	314.1	15.18
		Overall	384	-	192.5	-
	Clone (Genotype)	CC1	96	125.2	132.5	-6.12
		CC2	96	137.1	157.5	-3.57
		CN1	96	198.9	237.9	4.62
		CN2	96	205.8	242.1	5.06
		Overall	384	-	192.5	-
	Maturity stage	FirHa**	192	175.7	186.0	-1.15
		Secha**	192	178.7	199.0	1.15
		Overall	384	-	192.5	-
OHI	Treatment	Control	128	2.700	232.0	4.93
		BioF	128	2.370	133.4	-7.37
		CheF	128	2.650	212.1	2.44
		Overall	384	-	192.5	-
	Clone (Genotype)	CC1	96	2.470	168.8	-2.41
		CC2	96	2.490	163.3	-2.98
		CN1	96	2.740	228.2	3.64
		CN2	96	2.630	209.7	1.75
		Overall	384	-	192.5	-
	Maturity stage	FirHa	192	2.580	193.5	0.17
		Secha	192	2.570	191.5	-0.17
		Overall	384	-	192.5	-
CAL	Treatment	Control	128	40.13	194.0	0.19
		BioF	128	35.10	160.2	-403
		CheF	128	41.90	223.3	3.85
		Overall	384	-	192.5	-
	Clone (Genotype)	CC1	96	45.20	239.0	4.74
		CC2	96	44.01	237.2	4.55
		CN1	96	33.70	140.4	-5.31
		CN2	96	33.88	153.4	-3.98
		Overall	384	-	192.5	-
	Maturity stage	FirHa	192	35.10	153.5	-6.89
		Secha	192	43.12	231.5	6.89
		Overall	384	-	192.5	
LIN	Treatment	Control	128	0.8300	186.7	-0.73
		BioF	128	0.9900	225.2	4.08
		CheF	128	0.7700	165.7	-3.35
		Overall	384	-	192.5	-
	Clone (Genotype)	CC1	96	0.9100	206.7	1.45
		CC2	96	0.8600	197.2	0.48
		CN1	96	0.8100	181.8	-1.09
		CN2	96	0.8600	184.3	-0.84
		Overall	384	-	192.5	-
	Maturity stage	FirHa	192	1.0200	238.1	8.06
		Secha	192	0.7100	146.9	-8.06
		Overall	384	-	192.5	-

*: BioF = Biological Fertilizer, ChemF = Chemical Fertilizer (NPK); ** FirHa = First Harvest Date, SecHa = Second Harvest Date; † = Number of observations.

Table 5. Continued

Trait	Factor		N[†]	Median	Average Rank	Z
	Treatment	Control	128	0.9400	185.8	-0.83
		BioF*	128	1.1800	224.9	4.04
		CheF*	128	0.9100	166.8	-3.21
		Overall	384	-	192.5	-
		CC1	96	1.0500	206.5	1.43
		CC2	96	1.0400	196.1	0.37
CIO	Clone (Genotype)	CN1	96	0.9500	182.2	-1.05
		CN2	96	1.0400	185.2	-0.74
		Overall	384	-	192.5	-
		FirHa**	192	1.2100	237.6	7.97
	Maturity stage	Secha**	192	0.8400	147.4	-7.97
		Overall	384	-	192.5	-
		Control	128	10.600	193.5	0.12
	Treatment	BioF	128	9.815	156.7	-4.47
		CheF	128	12.230	227.4	4.35
		Overall	384	-	192.5	-
		CC1	96	11.695	207.8	1.56
		CC2	96	11.505	201.2	0.89
LIM	Clone (Genotype)	CN1	96	9.865	175.4	-1.75
		CN2	96	9.930	185.6	-0.71
		Overall	384	-	192.5	-
		FirHa	192	9.635	150.5	-7.42
	Maturity stage	Secha	192	12.180	234.5	7.42
		Overall	384	-	192.5	-

*: BioF = Biological Fertilizer, ChemF = Chemical Fertilizer (NPK); ** FirHa = First Harvest Date, SecHa = Second Harvest Date.

Table 6. Rankings of the three treatments (Control, BioF, CheF) and two maturity stages (First and Second Harvest) in terms of shoot and leaf dry matter biomass yield (DBY), citral (CAL), linalool (LIN), citronellol (CIO) and limonene (LIM) contents in essential oil extracted in a cluster analysis (Ward's minimum variance analysis) from the *Cymbopogon* genotypes for their relative responses to those factors.

Trait	Genotype	Treatment			Maturity stage	
	(Clones)	Control	BioF*	CheF*	FirH**	SecH**
	CC1	28 (2)[†]	15 (1)	30 (2)	39 (2)[††]	15 (1)
DBY	CC2	26 (2)	14 (1)	27 (2)	43 (2)	10 (1)
	CN1	24 (2)	15 (1)	31 (3)	41 (2)	15 (1)
	CN2	28 (2)	13 (1)	28 (2)	42 (2)	5 (1)
	CC1	19 (2)	6 (1)	4 (1)	26 (1)	31 (2)
OHI	CC2	16 (3)	8 (2)	3 (1)	26 (2)	19 (1)
	CN1	13 (2)	26 (3)	3 (1)	24 (2)	18 (1)
	CN2	15 (2)	24 (3)	6 (1)	28 (2)	21 (1)
	CC1	13 (2)	15 (2)	4 (1)	24 (1)	28 (1)
CAL	CC2	18 (3)	10 (1)	15 (2)	37 (1)	23 (1)
	CN1	17 (2)	14 (1)	13 (1)	27 (2)	23 (1)
	CN2	15 (2)	12 (1)	21 (3)	30 (1)	29 (1)
	CC1	20 (3)	9 (1)	8 (1)	28 (1)	28 (1)
LIN	CC2	18 (3)	14 (2)	2 (1)	24 (1)	23 (1)
	CN1	16 (3)	9 (2)	2 (1)	22 (1)	33 (2)
	CN2	14 (3)	7 (1)	10 (2)	23 (1)	33 (2)
	CC1	18 (3)	8 (1)	7 (1)	26 (1)	33 (2)
CIO	CC2	21 (3)	14 (2)	2 (1)	25 (1)	30 (2)
	CN1	19 (3)	6 (1)	6 (1)	23 (1)	30 (2)
	CN2	18 (3)	6 (1)	11 (2)	20 (1)	30 (2)
	CC1	14 (3)	9 (2)	9 (2)	29 (1)	28 (1)
LIM	CC2	15 (1)	19 (3)	16 (1)	30 (2)	22 (1)
	CN1	19 (3)	12 (1)	14 (1)	28 (1)	27 (1)
	CN2	19 (2)	7 (1)	17 (2)	31 (1)	32 (1)

*: BioF = Biological Fertilizer, ChemF = Chemical Fertilizer (NPK); ** FirHa = First Harvest Date, SecHa = Second Harvest Date; [†] : The value as indices indicated in parenthesis shows on the same line the effects of a given treatment on the performance of a particular genotype (clone). Lower the value is (compared to others), and strengthened is the effect of a treatment (compared to another) on the performance of the genotype in relevance to the expression of a specific trait; [††] : The value as indices in parenthesis indicates how a special harvest date influences the performance of a given genotype. Lower it is, compared to another on the same line, and more powerful is the special harvest date on the genotypic expression concerning a specific character.

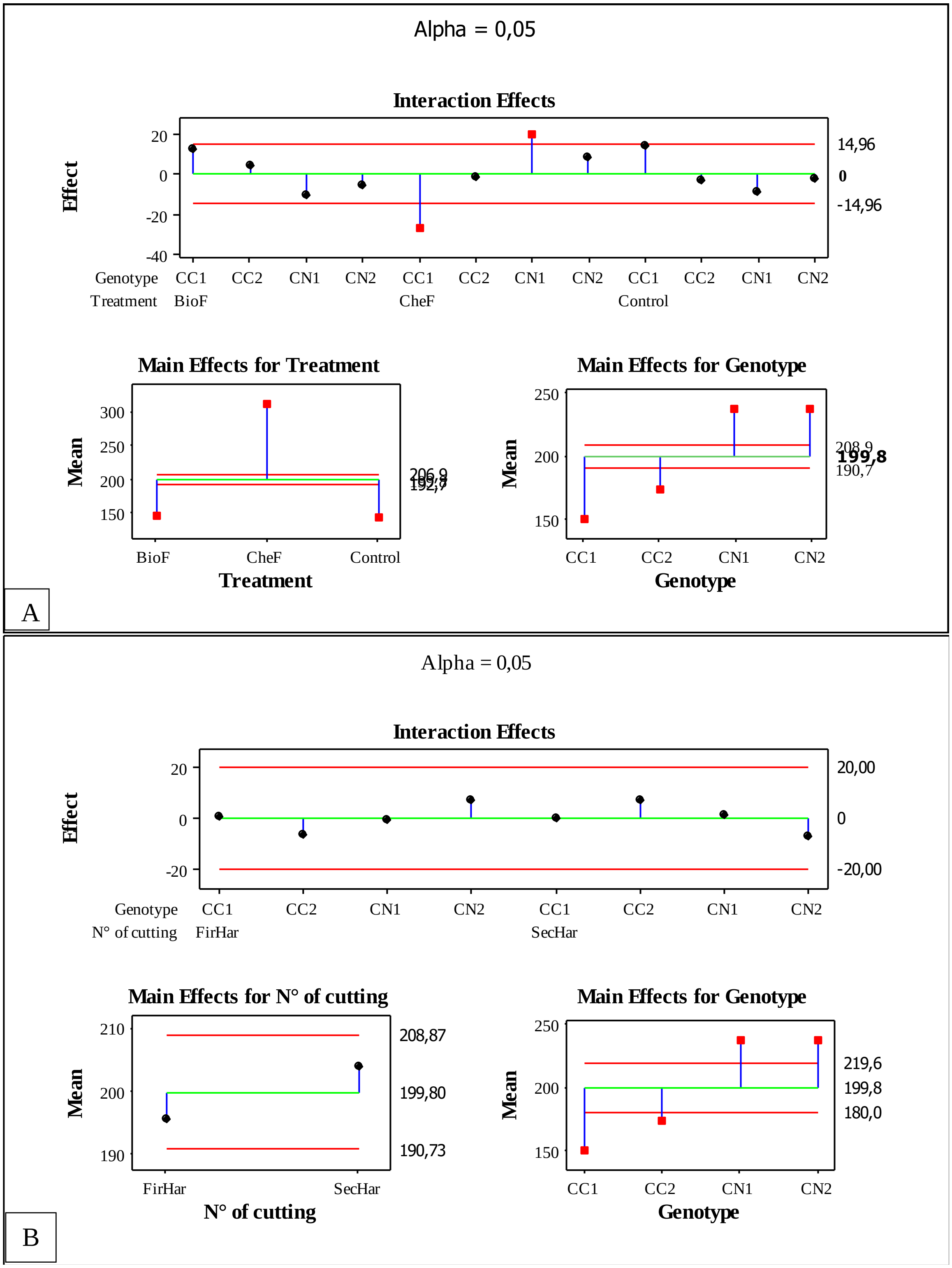

Fig. 1: Analysis of means for the biomass dry matter yield and interactions Genotype × Environment in two *Cymbopogon* species cultivated in Benin. A. under three treatments (fertilizer applications); B. for two harvest dates (4.5 and 6.5 months, respectively) after planting during the development stages.

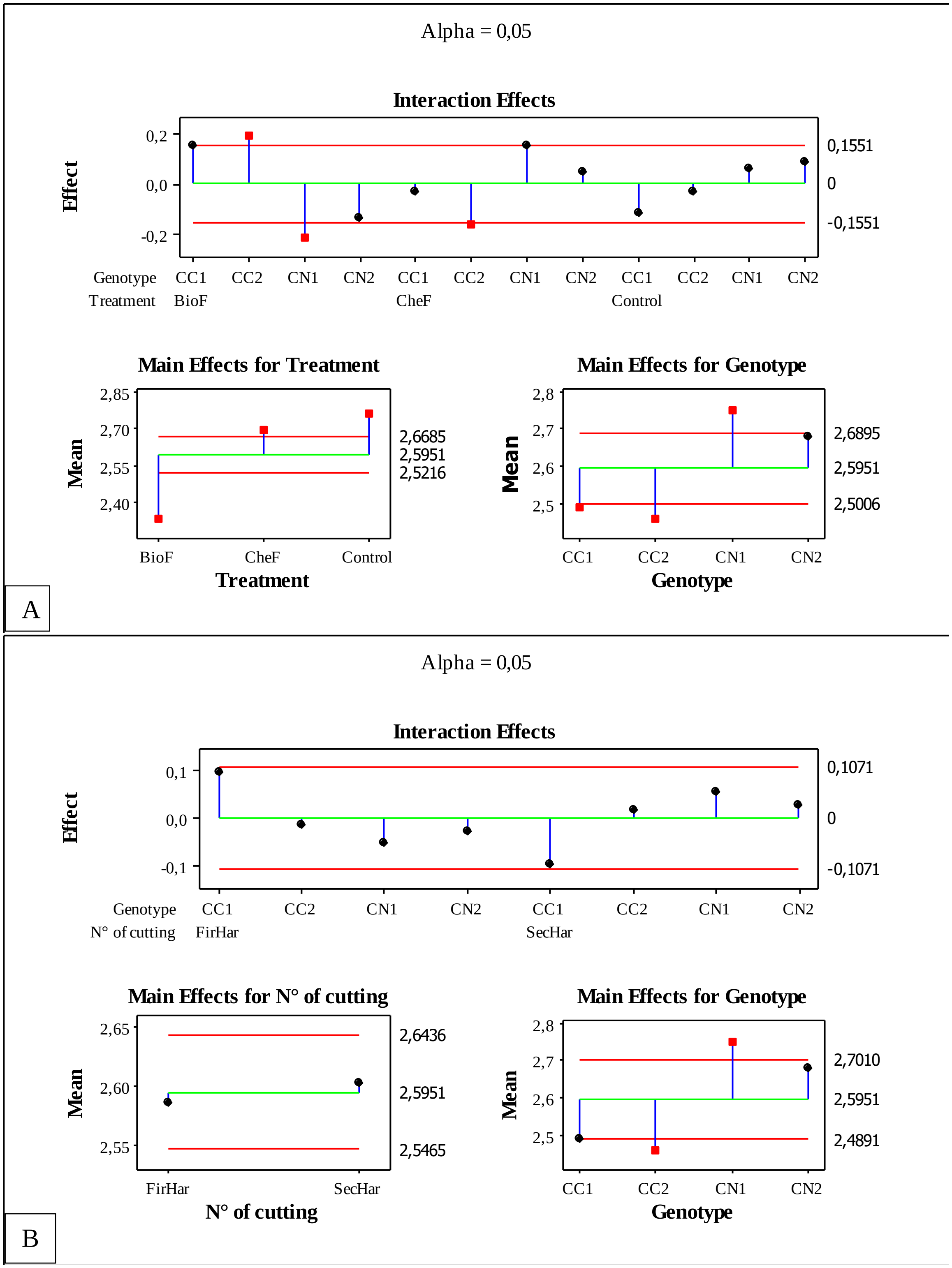

Fig. 2: Analysis of means for the essential oil harvest index (OHI) and interactions Genotype × Environment in two *Cymbopogon* species cultivated in Benin. A. under three treatments (fertilizer applications); B. for two harvest dates (4.5 and 6.5 months, respectively) after planting during the development stages.

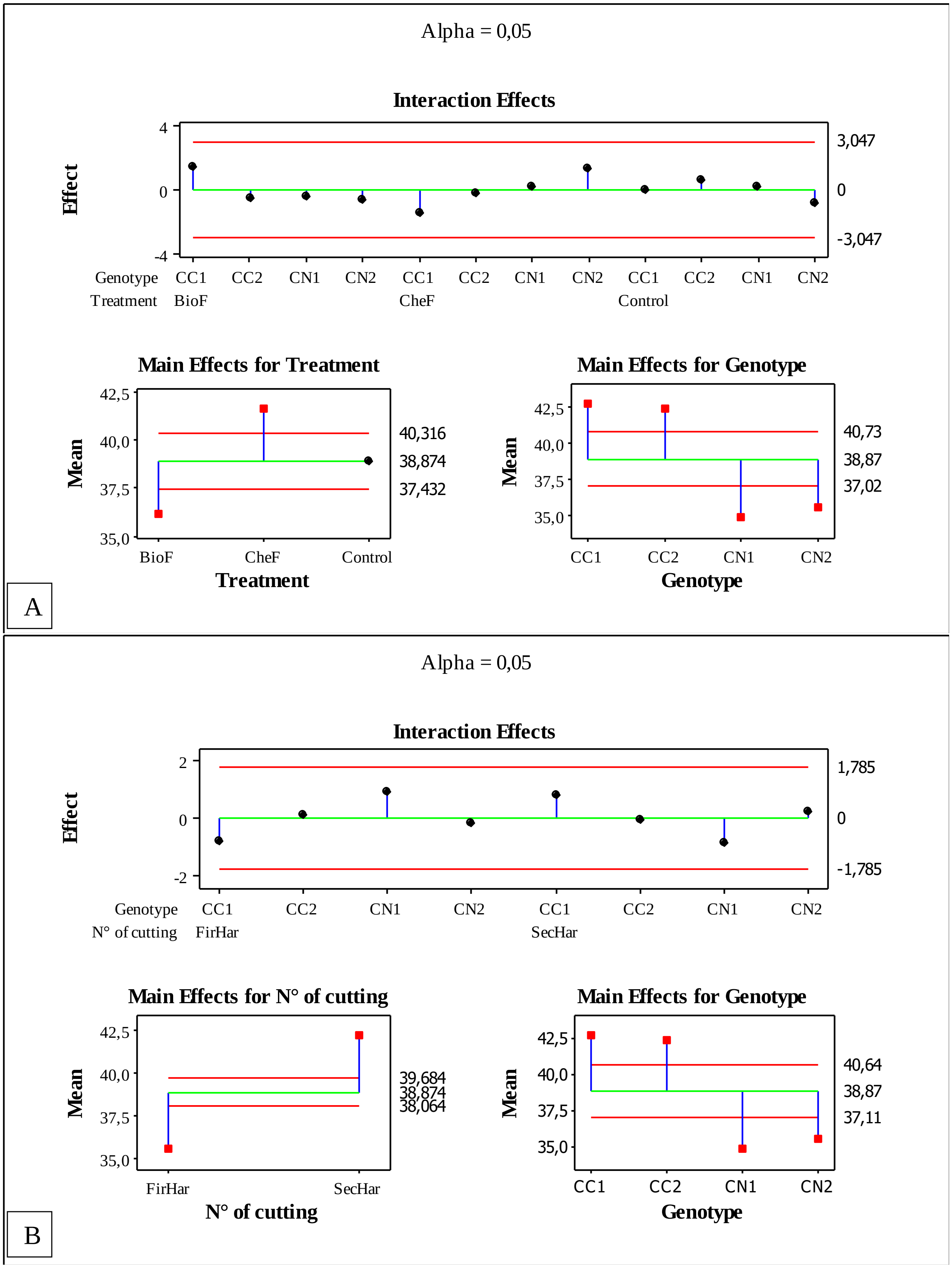

Fig. 3: Analysis of means for the citral content in essential oil and interactions Genotype × Environment in two *Cymbopogon* species cultivated in Benin. A. under three treatments (fertilizer applications); B. for two harvest dates (4.5 and 6.5 months, respectively) after planting during the development stages.

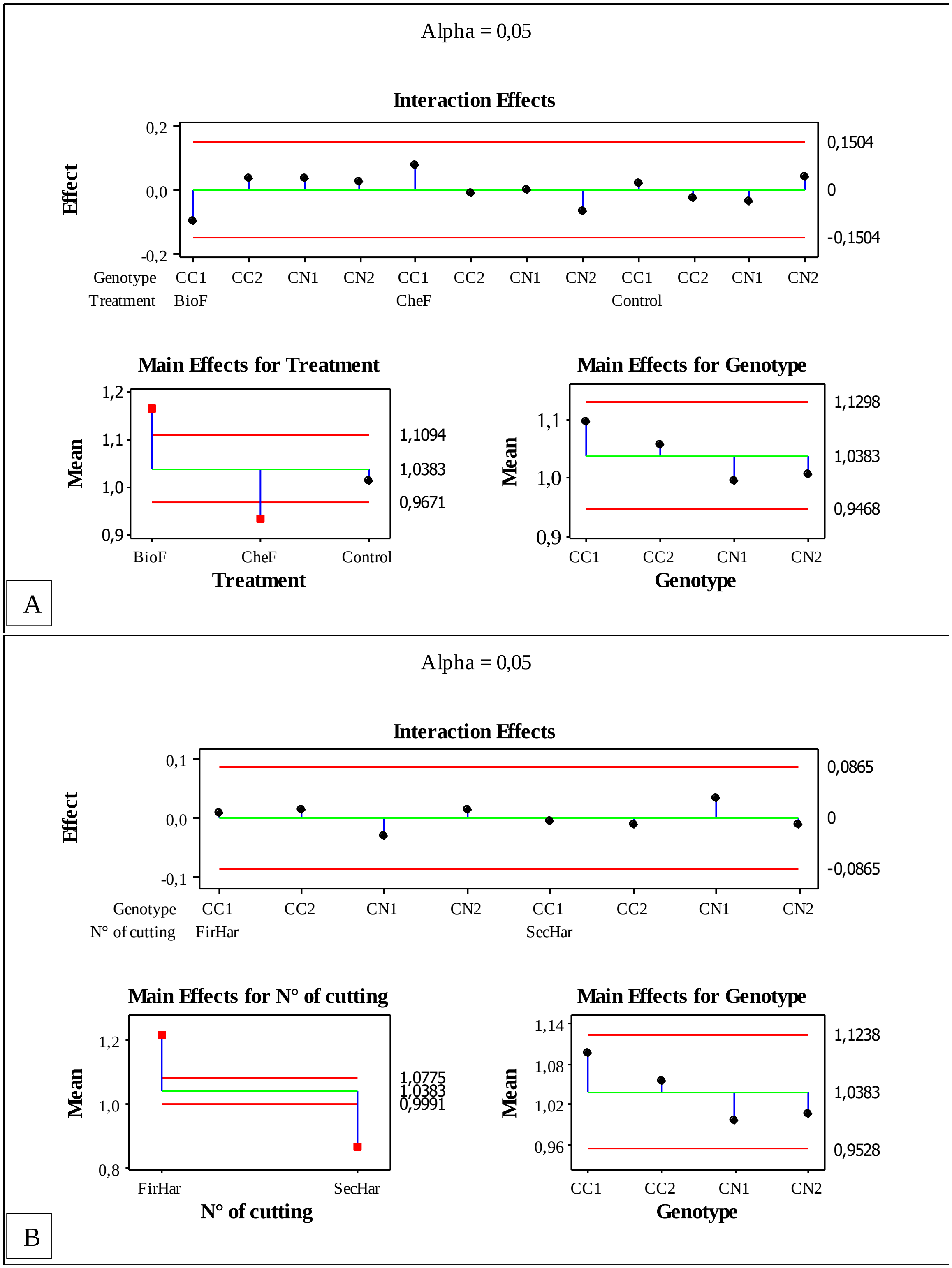

Fig. 4: Analysis of means for the citronellol content in essential oil and interactions Genotype × Environment in two *Cymbopogon* species cultivated in Benin. A. under three treatments (fertilizer applications); B. for two harvest dates (4.5 and 6.5 months, respectively) after planting during the development stages.

The test statistics (H = 230.73) had a *p*-value of 0.000 when it is adjusted for ties, or not, indicating that the null hypothesis can be rejected at levels higher than 0.000 in favour of the alternative hypothesis of the existence of at least one difference among the treatment groups. The treatment CheF appeared the most significant to a relatively good production of biomass in *Cymbopogon* spp collected and cultivated in different agroecological zones of Benin.

The sample medians for the four clones under investigation and for the trait DBY were 125.2, 137.5, 198.9 and 205.8 for CC1, CC2, CN1 and CN2, respectively as delivered by the Kruskal-Wallis test. The z value for CC2 is -3.57, the smallest absolute value of z. This size shows that the mean rank for this clone is the one that differs least from the mean rank across all observations. Furthermore, the mean rank of this provenance (157.5) was lower compared to the average over the whole study (192.5), and the z value is negative (z = -3.57). CN2 mean rank is higher, with a positive z value (z = 5.06) (Table 5). The test statistics (H = 72.12) had a *p*-value of 0.000 when it is adjusted or not, showing that the null hypothesis can be rejected at levels superior to 0.000 in favour of the alternative hypothesis of the existence of at least a difference among the groups of accessions or clones. The biomass dry matter yield in CN2 proved to be significantly the best whatever the treatment in *Cymbopogon*.

Finally, for DBY, the sample medians for the two harvest dates were 175.7 and 178.7 for FirHa and SecHa, respectively. The z value for FirHa is -1.15, the smallest absolute value of z. This size shows that the mean rank for this maturity stage is the one that differs least from the mean rank across all observations. Moreover, the mean rank of this harvest date (186.0) was lower compared to the average over the whole study (192.5), and the z value is negative (z = -1.15). The second maturity stage mean rank is higher, with a positive z value (z = 1.15) (Table 5).

The test statistics (H = 1.33) had a *p*-value of 0.250 when it is adjusted or not, showing that the null hypothesis can be rejected at levels superior to 0.250 in favour of the alternative hypothesis of the existence of at least a difference among both maturity stages. The biomass dry matter yield at the second harvest date (SecHa: 6.5 months after planting) proved to be significantly the best period for harvesting whatever the treatment, and the genotype in *Cymbopogon*.

Effects of diverse fertilizers and maturity stages on shoot and leaf oil harvest index (OHI)

For the trait "essential oil harvest index (OHI), the sample median values recorded for the three treatments were 2.700, 2.370 and 2.650% for the control, BioF and CheF, respectively. The z value for treatment BioF is -7.37, the smallest absolute value of z. This dimension shows that the mean rank for this treatment differs least from the overall mean. The mean rank of this treatment (133.4) was revealed lower than the overall mean rank (192.5), and the value of z is negative (z = -7.37). The mean rank for the control (232.0) was here higher, with a positive z value (z = 4.93) (Table 5). The test statistics (H = 56.43 and H = 56.45 when adjusted for ties) had a *p*-value of 0.000, indicating that the null hypothesis can be rejected at levels superior to 0.000 in favour of the alternative hypothesis of the existence of at least a difference among the groups of treatments. The probability being inferior to 0.05, the differences between the three treatments are therefore significant. CheF appeared to be however more efficient in obtaining better results in *Cymbopogon* from different agroecological zones of Benin. The calculation of the sample medians for the four accessions delivered 2.470, 2.490, 2.740 and 2.630% of essential oil in DBY, respectively. The z value for CC2 is -2.98, the smallest absolute value of z. This size shows that the mean rank for this accession is the one that differs least from the overall mean rank. CC2 mean rank (163.3) was lower compared to the general average (192.5), and the z value is rather negative (z = -2.98). CN1's mean rank (228.2) was higher, with a positive z value (z = 3.64) (Table 5). The Kruskal-Wallis test statistics (H = 23.26) had a *p*-value of 0.000 when she/it is adjusted, H = 23.27, showing that the null hypothesis can be rejected at levels superior to 0.000 in favour of the alternative hypothesis of the existence of at least a difference among the groups of accessions. The probability is distinctly lower than 0.05. Thus, the differences between the four clones are indeed very significant. CN1 essential oil content seemed better whatever the treatment.

The sample median values recorded for the two maturity stages were 2.580 and 2.570%, respectively. The z value for SecHa is -0.17, the smallest absolute value of z. This dimension demonstrates that the mean rank for this harvest date differs least from the overall mean. The mean rank of this maturity stage (191.5) was revealed lower but similar to the overall mean rank

(192.5), and the value of z is negative (z = -0.17). The mean rank for the FirHa (193.5) was here higher, with a positive z value (z = 0.17) (Table 5). The test statistics (H = 0.03) had a *p*-value of 0.866, indicating that the null hypothesis can be rejected at levels superior to 0.866 in favour of the alternative hypothesis of the existence of at least a difference among the groups of treatments. The probability being superior to 0.05, the differences between both harvest dates surveyed are therefore non-significant.

Effects of diverse fertilizers and maturity stages on citral content (CAL) in essential oil

The sample median values recorded for the three treatments with a focus on citral contents in DBY were 40.13, 35.10 and 41.90% in the essential oil of *Cymbopogon* surveyed for the control, BioF and CheF, respectively. The z value for treatment BioF is -4.03, the smallest absolute value of z. This dimension shows that the mean rank for this treatment differs least from the overall mean. The mean rank of this treatment (160.2) was revealed lower than the overall mean rank (192.5), and the value of z is negative (z = -4.03). The mean rank for CheF was here higher, with a positive z value (z = 3.85) (Table 5). The test statistics (H = 20.74) had a *p*-value of 0.000 when it is adjusted or not, indicating that the null hypothesis can be rejected at levels superior to 0.000 in favour of the alternative hypothesis of the existence of at least a difference among the groups of treatments. The probability being inferior to 0.05, the differences between the treatments are therefore significant. CheF appeared to be however more efficient to obtain better results in the report presented here. The calculation of the sample medians for the four genotypes delivered 45.20, 44.01, 33.70 and 33.88% citral contents in oil for CC1, CC2, CN1 and CN2, respectively. The z value for CN1 is -5.31, the smallest absolute value of z. This size shows that the mean rank for this clone (genotype) is the one that differs least from the overall mean rank. CN1's mean rank (140.4) was lower compared to the general average (192.5), and the z value is rather negative (z = -5.31).

CC1 mean rank (239.0) was higher, with a positive z value (z = +4.74) (Table 5). The Kruskal-Wallis test statistics (H = 65.46) had a *p*-value of 0.000 when it is adjusted or not, showing that the null hypothesis can be rejected at levels superior to 0.000 in favour of the alternative hypothesis of the existence of at least a difference among the groups of accessions. The probability is distinctly lower than 0.05. Thus, the differences between the four accessions are indeed very significant.

The sample median values recorded for the two maturity stages were 35.10 and 43.12% citral content, respectively. The z value for FirHa is -6.89, the smallest absolute value of z. This dimension demonstrates that the mean rank for this harvest date differs least from the overall mean. The mean rank of this maturity stage (153.5) was revealed lower than the overall mean rank (192.5), and the value of z is negative (z = -6.89). The mean rank for the SecHa (231.5) was here higher, with a positive z value (z = 6.89) (Table 5). The test statistics (H = 47.45) had a *p*-value of 0.000, indicating that the null hypothesis can be rejected at levels superior to 0.000 in favour of the alternative hypothesis of the existence of at least a difference among the groups of harvest date. The probability being inferior to 0.05, the differences between both harvest dates surveyed are therefore very significant.

Effects of diverse fertilizers and maturity stages on linalool content (LIN) in essential oil

The sample medians for the three treatments were 0.8300, 0.9900 and 0.7700% of linalool for the control, the BioF and the CheF, respectively. The z value for treatment "CheF" is -3.35, the smallest value of z. This size indicated that the mean rank for this treatment differs least from the mean rank for all observations. Moreover, the mean rank of this treatment (165.7) was lower than the mean of the observation set (192.5), and the z value is negative (z = -3.35). The mean rank for the treatment BioF is higher than in the whole experiment, with a positive z value (z = +4.08) (Table 5). The test statistics (H = 18.90 or 18.91) had a *p*-value of 0.000 when it is adjusted for ties or not, indicating that the null hypothesis can be rejected at levels higher than 0.000 in favour of the alternative hypothesis of the existence of at least one difference among the treatment groups. The treatment BioF appeared the most significant to a relatively good linalool content in *Cymbopogon* cultivated in Benin.

The sample medians for the four accessions (clones or genotypes) were 0.9100, 0.8600, 0.8100 and 0.8600% of linalool for CC1, CC2, CN1 and CN2, respectively as delivered by the Kruskal-Wallis test. The z value for CN1 is -1.09, the smallest absolute value of z. This size shows that the mean rank for this accession is the one that differs least from the mean rank across all observations. Furthermore, the mean rank of this provenance (181.8) was lower compared to the average over the whole study (192.5), and the z value is negative (z = -1.09). CC1 mean rank is higher, with a positive z value (z = 1.45) (Table 5). The test statistics (H = 3.16) had a *p*-value of 0.368 when it is adjusted or not, showing that the null hypothesis can be rejected at levels superior to 0.368 in favour of the alternative hypothesis of the existence of at least a difference among the groups of accessions.

The sample medians for the two harvest dates of interest in this study were 1.0200 and 0.7100% of linalool for the first and second harvest dates, respectively. The z value for SecHa is -8.06, the smallest value of z. This size indicated that the mean rank for this treatment differs least from the mean rank for all observations. Moreover, the mean rank of this maturity stage (146.9) was lower than the mean of the observation set (192.5), and the z value is negative (z = -8.06) (Table 5). The test statistics (H = 64.92 or 64.95) had a *p*-value of 0.000 when it is adjusted for ties or not, indicating that the null hypothesis can be rejected at levels higher than 0.000 in favour of the alternative hypothesis of the existence of at least one difference among the harvest date groups.

Effects of diverse fertilizers and maturity stages on citronellol content (CIO) in essential oil

The sample medians for the treatments were 0.9400, 1.1800 and 0.9100% of citronellol for the control, BioF and CheF, respectively. The z value for CheF is -3.21, the smallest value of z. This size indicated that the mean rank for this treatment differs least from the mean rank for all observations. Moreover, the mean rank of this treatment (166.8) was lower than the mean of the observation set (192.5), and the z value is negative (z = -3.21). The mean rank for BioF is higher than in the whole experiment, with a positive z value (z = +4.04) (Table 5). The test statistics (H = 18.19 or 18.20) had a *p*-value of 0.000 when it is adjusted for ties or not, indicating that the null hypothesis can be rejected at levels higher than 0.033 in favour of the alternative hypothesis of the existence of at least one difference among the treatment groups. BioF appeared to be the most significant to a relatively good citronellol content in lemongrass as well as citronella grass examined in this survey.

The sample medians for the four accessions or clones under survey were 1.0500, 1.0400, 0.9500 and 1.0300% of citronellol content in DBY for CC1, CC2, CN1 and CN2, respectively as delivered by the Kruskal-Wallis test. The z value for CN1 is -1.05, the smallest absolute value of z. This size indicates that the mean rank for this accession is the one that differs least from the mean rank across all observations. Furthermore, the mean rank of this provenance (182.2) was lower compared to the average over the whole study (192.5), and the z value is negative (z = -1.08). CC1 mean rank is higher, with a positive z value (z = +1.43) (Table 5). The test statistics (H = 2.88) had a *p*-value of 0.411 when it is adjusted or not, showing that the null hypothesis can be rejected at levels superior to 0.411 in favour of the alternative hypothesis of the existence of at least a difference among the groups of accessions.

For the citronellol content in DBY, the sample medians for both harvest dates were 1.2100 and 0.8400% FirHa and SecHa, respectively. The z value for SecHa is -7.97, the smallest absolute value of z. This size demonstrates that the mean rank for this harvest date differs least from the mean rank across all observations. Moreover, the mean rank by this maturity stage (147.4) was lower compared to the overall average rank (192.5). The mean rank by FirHa higher (239.8) is higher, with a positive z value (z = +7.97). H = 63.52 or 63.53 and had a *p*-value of 0.000 whenever it was adjusted or not, indicating that the null hypothesis can be rejected at levels superior to 0.000 in favour of the alternative hypothesis of the existence of at least a difference among both harvest dates under study here.

Effects of diverse fertilizers and maturity stages on limonene content (LIM) in essential oil

The sample median values recorded were 10.600, 9.815 and 12.230% of limonene (LIM) content in biomass oil for the control, BioF and CheF, respectively. The z value for BioF is -4.47, the smallest absolute value of z. This dimension shows that the mean rank for this treatment differs least from the overall mean. The mean rank of this treatment (156.7) was revealed lower than the overall mean rank (192.5), and the value of z is negative (z = -4.47). The mean rank for the CheF was here higher, with a positive z value (z = 4.35) (Table 5). The test statistics (H = 25.95 or 25.96) had a *p*-value of 0.000 when it is adjusted or not, indicating that the null hypothesis can be rejected at levels superior to 0.000 in favour of the alternative hypothesis of the existence of at least a difference among the groups of treatments.

The probability being inferior to 0.05, the differences between the three treatments are therefore significant.

The calculation of the sample medians for the four accessions delivered 11.695, 11.505, 9.865 and 9.930% of limonene (LIM) in biomass dry matter essential oil for CC1, CC2, CN1 and CN2, respectively. The z value for CN1 is -1.75, the smallest absolute value of z. This size shows that the mean rank for this accession is the one that differs least from the overall mean rank. CN1's mean rank (175.4) was lower compared to the general average (192.5), and the z value is rather negative (z = -1.75). CC1's mean rank (207.8) was higher, with a positive z value (z = 1.56) (Table 5). The Kruskal-Wallis test statistics (H = 5.08) had a *p*-value of 0.166 when it is adjusted or not, showing that the null hypothesis can be rejected at levels superior to 0.166 in favour of the alternative hypothesis of the existence of at least a difference among the groups of accessions. The probability is distinctly higher than 0.05. Thus, the differences between the four accessions are indeed non-significant.

The sample median values recorded for the two harvest dates were 9.615 and 12.180% of limonene for the first and second harvest dates, respectively. The z value for FirHa is -7.42, the smallest absolute value of z. This dimension shows that the mean rank for this maturity stage differs least from the overall mean. The mean rank of this harvest date (150.5) was revealed lower than the overall mean rank (192.5), and the value of z is negative (z = -7.42). The test statistics (H = 55.04) had a *p*-value of 0.000 when it is adjusted or not, indicating that the null hypothesis can be rejected at levels superior to 0.000 in favour of the alternative hypothesis of the existence of at least a difference among the groups of treatments. The probability being inferior to 0.05, the differences between both maturity stages are therefore significant.

Rankings of the three treatments (Control, BioF, CheF), four genotypes and two maturity stages (First and Second Harvest) in terms of their effects on yield and its related traits in Cymbopogon

The results on the ranking are shown in Table 6. The statistical analysis, involving the hierarchical clustering approaches using Ward's minimum variance cluster analysis implemented in JMP 7 (SAS Institute NC 2007), showed clearly the ranking of the treatments and maturity stages about their influences on the six quantitative characters examined in the genus *Cymbopogon* cultivated in Benin.

BioF enhanced significantly shoot and leaf biomass dry matter yield (DBY) than the control and CheF (Table 6). CheF improved rather than BioF the traits such as the essential oil harvest index (OHI), linalool (LIN), citronellol (CIO) and limonene (LIM) contents in oil and then in biomass produced. BioF improved the citral (CAL) contents in shoot and leaf dry matter more than others. For most of the traits, and independently of the genotypes, better results were obtained at the end of the maturity period than by the mid-time (Table 6).

Discussion

Information on the variation available for different plant attributes has enabled germplasm collections to be effectively utilized in plant breeding. Meaningful comparison of variation in quantitative traits requires controlling for both the dimension of the varying entity and the dimension of the factor generating variation (Pélabon et al., 2020). Advanced statistical models to handle increasingly large and complex datasets are often employed at the expense of attention given to the meaning of the numbers (Pélabon et al., 2020; Tarka et al., 2015). The coefficient of variation (CV) in this research report varies strongly from 19.23 for the essential oil harvest index (OHI) to 67.76% for "other compounds" (OTO) that were not identified with GC-MS. Most of the traits presented values around 40%. This can show the phenotypic variation that could exist in the plant materials used. However, the experimentation reported here has been laid out in a randomized block design with four replications at two locations during two different years. Irrigation was also applied consequently to prevent possible lack due to bad environment. Experimentations were thus laid out with much accuracy so that the highness of CV could be attributed mainly to genotypic differences between and within the clones examined.

The demand for lemongrass (*Cymbopogon citratus*) and citronella grass (*C. nardus*) is great for their high citral content. Early or delayed harvesting of lemongrass affected essential oil and citral content (Tajidin et al., 2012).

Essential oils are natural products obtained from plants. They are formed by varied and complex volatile mixtures of chemical compounds, with a predominance of terpenes associated with aldehydes, alcohols and ketones which are deposited in various structures of the plant (Zanklan and Baba-Moussa, 2025; Pandey, 2024; Salsabila et al., 2023; Sivamaruthi et al., 2022; Tajidin et al., 2012; Linares et al., 2005). Lemongrass contains mainly citral (Pandey, 2024; Sivamaruthi et al., 2022; Sharma et al., 2020; Tajidin et al., 2012). Essential oil and citral of lemongrass were detected to gather at parenchyma tissue cells, specifically in the adaxial surface of leaf mesophyll (Pandey, 2024; Tajidin et al., 2012; Lewinsohn et al., 1998). The citral of lemongrass is a natural combination of two isomeric aldehydes, namely isomers geranial (α-citral) and neral (β-citral) (Pengelly, 2004). Other unusual active components are limonene, citronellal, ß-myrcene and geraniol (Pandey, 2024; Schaneberg and Khan, 2002). Essential oil and citral contents were influenced by factors such as temperature, light intensity, soil moisture, fertilizer, and maturity stage (Zanklan and Baba-Moussa, 2025; Sharma et al., 2020). Research reports showed that overall essential oil production is associated with the early growth stage in plants such as *Cymbopogon flexuosus* (Singh et al., 1989), *Cymbopogon martini* (Sangwan et al., 1982) and *Mentha* (Caskill and Croteau, 1995). In general, the yield of essential oil is highly correlated with the yield of biomass (Zanklan and Baba-Moussa, 2025; Sharma et al., 2020). The production of higher-quality oil with high citral content (75%) is determined by the proportion of young to older leaves when harvested at a given point. These findings did not match our research here as it is found that old shoots and leaves after 6.5 months from planting delivered better citral as well as other important compounds such as linalool, citronellol and limonene than previously at 4.5 months of harvest date. Normally, lemongrass oil is extracted in various ways such as the solvent, accelerated solvent, Soxhlet (Sargenti and Lancas., 1997), dense carbon dioxide (Carlson et al., 2001), solid-phase matrix (Pham-Tuan et al., 2001), and supercritical fluid (Schaneberg and Khan, 2002) extraction methods. However, the common procedure for extracting essential oil is the hydrodistillation method (Zanklan and Baba-Moussa, 2025; Sharma et al., 2020; Kulkarni et al., 2003). Gas chromatography-mass spectrometry (GC-MS) has been the most applied analytical technique for essential oil analysis (Zanklan and Baba-Moussa, 2025; Masada, 1976) followed by supercritical fluid extraction-gas chromatography (Liu et al., 1993). Due to the complexity of essential oil compositions, sophisticated instruments such as high-performance liquid chromatography in combination with gas chromatography (HPLC-GC) (Tajidin et al., 2012; Mondello et al., 1996) are the preferred analysis. HPLC is effective for a broad class separation of a sample, which can be introduced into a GC for further high-resolution separation. In the research reported here, the hydrodistillation method was applied, followed by GC-MS.

Lemongrass and citronella grass were widely used for their therapeutical properties due to the phytochemical compounds such as citral, linalool, citronellol, limonene, citronellal, geraniol and nerol that they contain (Pandey, 2024; Salsabila et al., 2023; Sharma et al., 2020; Singh and Kumar, 2017; Ghosh, 2013). As Singh and Kumar (2017) earlier found, those compounds were found with the same range in *Cymbopogon* cultivated in Benin. That is why, the authors of the present report chose to concentrate their further prospects on the citral, linalool, citronellol and limonene contents in leaves and shoots of *Cymbopogon*. Moreover, strong antifungal activity against several species of *Aspergillus*, *Penicillium* and *Eurotium* due to the presence of citronellal and linalool components of citronella oil, has also been reported (Pandey, 2024; Salsabila et al., 2023; Kiani et al., 2022; Arpiwi et al., 2020; Singh and Kumar, 2017). Essential oils of citronella and lemongrass play an important role in improving human health due to their potential of bioactive components that can act as anti-inflammatory, antioxidant, anticancer, anticonvulsant agents and central nervous system (CNS) disorders (Pandey, 2024; Salsabila et al., 2023; Kiani et al., 2022; Mukarram et al., 2021; Sharma et al., 2020; Mishra et al., 2019; Singh and Kumar, 2017). The essential oils of Java Citronella are most commonly blended monoterpenes, monoterpenoids, and phenylpropanoids containing chiefly limonene, linalool, geraniol, elemol, geranyl acetate, α-bisafalol, citronellol, Citronellal (Salsabila et al., 2023; Kiani et al., 2022; Mukarram et al., 2021; Arpiwi et al., 2020; Mishra et al., 2019; Singh and Kumar, 2017; Cassel and Vargas, 2006). Citronella herb is found in many parts of India and Africa. Those studies found out in most cases that citronella and lemongrass contain in their leaves and shoots more than 40% citral, and between 1-10% of the other compounds. Their findings are in accordance with the survey reported here. But, little is known about the effects that the soils, where the clones are planted, could have upon the outcome of such amounts of chemical compounds which are found in essential oils extracted from the plant organs. The study reported here focused on the importance that the biological or chemical fertilizer should have on the production of biomass, and its essential oil harvest index as well as the phytochemical composition.

Essential oils are volatile secondary metabolites of plants that can be extracted from several parts of plants, such as flowers, seeds, fruits, twigs, leaves, etc. They are the major constituent of perfumes, fragrances, cosmetics and pharmaceutics (Sivamaruthi et al., 2022) and are used to treat oral problems (Kiani et al., 2022; Sivamaruthi et al., 2022). The composition of the oil is responsible for its bioactivity. The need for surveying agronomic conditions and practices necessary for the good growth and development of citronella and lemongrass is crucial, as their importance for human beings and their animals as well as other plant species has been stressed for centuries. Tajidin et al. (2012) in a study on the effects of maturity stages and the oil harvest and composition in lemongrass concluded that those parameters were higher expressed at 5.5 and 6.5 months after the planting than at the 7.5 months stages. In our study, the first harvest happened in 4.5 and the second in 6.5 months after planting. It resulted in better biomass yields at 6.5 months than at 4.5 after planting. It could not be concluded whether their findings were in accordance or not with the results exposed here. It might be necessary to run a kinetics of harvest date and its effects on biomass production, oil harvest and chemical composition, in relation to soil type and fertilizer applications. Amirthavarshini and Anuja (2024) studied how diverse mixtures of chemical (NPK) and biological fertilizers could act from the Recommended Dose of Fertilizer (RDF). They found that the application of 75% Recommended Dose of Fertilizer with vermicompost 5 t/ha and 2% foliar spray of seaweed extract has been identified as the best treatment for maximizing the growth characteristics of citronella. This report here did not handle the same approaches as these authors. The mixture of chemical and biological fertilizers was not done to examine the best agricultural practices in the crops under surveys. The report presented here could be completed by their findings to obtain the best way for farmers in Benin and West Africa. However, it shall be detected the best practices for sustainable agriculture, and the protection of environmental balances. In the same trend, Karde et al. (2024) reported the influence of sulphur-enriched compost and growth regulators on the growth of lemongrass and highlighted the importance of a well-balanced combination of sulphur-enriched compost and plant growth regulators in optimizing lemongrass cultivation, offering insights that can contribute to improved agricultural practices. Herwita et al. (2024) studied also with the same tendency the effect of fertilizer doses and harvest interval on the intensity of leaf spot diseases, production and quality of citronella grass (*Cymbopogon nardus* L.) essential oils in ultisols soil. Their conclusions were similar to the earlier analyses.

From the current report, some statistical tools (ANOVA, Analysis of means, Kruskal-Wallis non-parametric test, Ward's minimum variance cluster analysis) were used to compare genotypic variances, biomass production, its essential oil content and phytochemical composition of citronella and lemongrass cultivated in Benin. It is the first time that such research has been undertaken in this country. Biological fertilizer applications favoured an improved growth rate rather than chemical fertilizer, independently of the species, but especially in *Cymbopogon citratus*. Biological fertilizer affected also positively the citral contents of the shoots and leaves, rather than other compounds present in the essential oils that were screened with GC-MS.

Chemical fertilizers enhanced the percentage of other compounds such as citronellol, linalool and limonene in oils from *C. nardus*. Further prospects are needed to confirm or infirm these preliminary findings. The number of clones to be surveyed should be increased. The diversification of components in fertilizers should be also of interest. Under those conditions, it should be possible, for each compound present in essential oils from citronella and lemongrass, to study the inheritance and the phenotypic and genotypic variation existing in the crops locally. Moreover, the genetic advance, path coefficient analysis and inter-character relationships will be examined. These steps are necessary to better understand the pathway of biosynthesis concerning each metabolite present in the essential oil. Such investigations will let controlling better the amount of a given plant product in the oil in question. It might be possible to have some compounds in the oils that derived from one another during the growth and development of these fascinating crops. Furthermore, the kinetics of biomass production as well as the essential oil harvest index in relation to the phytochemical composition shall be better surveyed at diverse maturity stages and under different fertilizer (biological, chemical) application regimes.

Conclusions

The study presented here permits an easy understanding of the agricultural status of *Cymbopogon* genus in Benin and West Africa. Biomass yields along with phytochemical characterization of lemongrass and citronella grass under different regimes of fertilizer applications allow to conclude that each genotype of *C. citratus* and *C. nardus* remains heterogenous and the diversity is very high. The statistical analyses performed here were useful to identify a great variation in the responses of the local clones studied in terms of their adaptation to biological versus chemical fertilization and to bring onto farmers clones with preferred phytochemical quality of interest. The crops reacted diversely to the treatments they have been subjected to. No clear difference between and within the clones or between the two species investigated was observed. Nonetheless, citronella grass was partly better adapted to chemical fertilizer than lemongrass, which rather delivered the best response to biological fertilizer applications as it showed better composition in citral content (above 40%) and dry matter biomass yields. This work provided an important contribution to further analysis of phytochemical composition under a large pan of diverse doses of biological and chemical fertilizers as well as their mixtures corresponding to a panel of them in different doses. The number of clones to be used in such investigations should be augmented in other to better analyze the genetic parameters involved. This might be a prerequisite to limit fully the excessive utilization of chemical *vs* inorganic fertilizers which unfortunately show the worst consequences to maintaining the environmental balances. Moreover, studies on the accurate harvest date have to be deepened to find out the right period to plant and harvest these crops to get better yields, essential oil amounts and good phytochemical compounds inside without any special difficulty. In conclusion, there is a need to fulfill the collection, evaluation and selection of genotypes, which can help breeders to enhance and promote the crops regarding their potential.

Conflict of interest statement

Authors declare that they have no conflict of interest.

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Author Contributions

Ahissou Séraphin Zanklan: Investigation, methodology, formal analysis, writing original draft. Farid Abdel Kader Baba-Moussa: Validation, writing-reviewing.

Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

References

Ali, A., Zahid, H.F., Cottrell, J.J. and Dunshea, F.R., 2022. A Comparative Study for Nutritional and Phytochemical Profiling of *Coffea arabica* (*C. arabica*) from Different Origins and Their Antioxidant Potential and Molecular Docking. Molecules 2022, 27, 5126.

Amirthavarshini, E.S and Anuja, S., 2024. Effect of Integrated Nutrient Management on Growth Parameters of Citronella (*Cymbopogon winterianus* Jowitt.). International Journal of Plant & Soil Science 36(10): 504-509.

Aoudjit-Hayet, M., 2006. Etude de la germination des grains du *Pistacia atlantica* Desf. (pistachier de l'Atlas) et essai de multiplication de *Pistacia vera* L. (pistachier vrai) en pépinière selon deux types de greffes Ecusson et « Chip budding ». Mémoire de Magistère en Sciences Agronomiques, Institut National Agronomique El Harrach, 93p.

Arpiwi, N.L., Muksin, I.K. and Kartini, N.L., 2020. Essential oil from *Cymbopogon nardus* and repellant activity against *Aedes aegypti*. Biodiversitas 21(8) : 3873-3878. DOI: 10.13057/biodiv/d210857.

Carlson, L.H.C., Machado, R.A.F., Spricigo, C.B., Pereira, L.K. and Bolzan, A., 2001. Extraction of lemongrass essential oil with dense carbon dioxide. J. Supercritical Fluids. 21: 33-39.

Caskill, D. and Croteau, R., 1995. Monoterpene and sesquiterpene biosynthesis in glandular trichomes of peppermint (*Mentha piperita*) rely exclusively on plastid derived isopentenyl diphosphate. Planta. 197: 49-56.

Cassel, E. and Vargas, R.M.F., 2006. Experiments and modeling of the *Cymbopogon winterianus* essential oil extraction by Steam Distillation. J Mex Chem Soc 50(3): 126–129.

El-Hendawy, S.E.-S., 2004. Salinity Tolerance in Egyptian Spring Wheat Genotypes. PhD Thesis, Technische Universität Muenchen, Germany, 116p.

Essou, J.I.L., Zanklan, A.S., Adomou, A.C. and Dassou, G.H., 2017a. Effects of Diverse Pretreatments on Seed Germination in *Cleome gynandra* L. (Capparidaceae) - A Threatened Species, Collected from Different Agro-Ecological Zones in Benin. Int. J. Curr. Res. Biosci. Plant Biol. 4(7): 47-59.

Essou, J.I.L., Zanklan, A.S., Adomou, A.C. and Assogba, F., 2017b. Studies on Phytochemistry and Antioxidant Potential of *Cleome gynandra* L. (Capparidaceae) Collected from Contrasted Agro-Ecological Zones in Benin. European Journal of Scientific Research 147 (3): 251-274.

Ghosh, K., 2013. Anticancer effect of lemongrass oil and citral on cervical cancer cell lines. Pharmacognosy Communications 3(4) : 1-8.

Haque, A.N.M.A., Remadevi, R. and Naebe, M., 2018. Lemongrass (*Cymbopogon*): a review of its structure, properties, applications, and recent developments. Cellulose: *25*(10): 5455-5477.

Herwita, I., Nurmansyah, Wiratno, Mayura, E., Riska, Budiyanti, T., Gustia, H. and Ramadhan, A.I, 2024. Effect of doses fertilizer and harvest interval on the intensity of leaf spot diseases, production and quality of citronella grass (*Cymbopogon nardus* L.) essential oils in ultisols soil. Heliyon 10(5), e26822.

Jollife, I.T., Allen, O.B. and Christie, B.R., 1989. Comparison of variety means using cluster analysis and dendrograms. Exp. Agric. 25: 259-269.

Karde, R.Y., Parlawar, N.D., Patke, N.K., Paslawar, A.N. and Jadhao, S.D., 2024. Influence of sulphur enriched compost and plant growth regulators on growth of lemon grass. International Journal of Advanced Biochemistry Research 2024; 8(9): 421-424.

Kiani, H.S., Ali, A., Zahra, S., Hassan, Z.U., Kubra, K.T., Azam, M. and Zahid, H.F., 2022. Phytochemical Composition and Pharmacological Potential of Lemongrass (Cymbopogon) and Impact on Gut Microbiota. Applied Chem 2: 229–246. *https://doi.org/10.3390/appliedchem2040016*.

Kiloni, S.M., Akhtar, A., Cáceres-Vélez, P.R., Dunshea, F. and Jusuf, P., 2022. P06-05 Zebrafish embryo acute toxicity and antioxidant characterization of native Australian plants: Towards safe and effective glaucoma treatments. Toxicol. Lett. 2022, 368, S115.

Kulkarni, R.N., Baskaran, K. and Ramesh, S., 2003. Five cycles of recurrent selection for increased essential oil content in East Indian Lemongrass: response to selection, and effects on heritabilities of traits and inter trait correlations. Plant Breeding. 122: 131-135.

Lewinsohn, E., Dudai, N., Tadmor, Y., Katzir, I., Ravid, U., Putievsky, E. and Joel, D.M., 1998. Histochemical localization of citral accumulation in lemongrass leaves (Cymbopogon *citratus* (DC.) Stapf., Poaceae). Ann. Bot. 81: 35-39.

Linares S., Gonzalez, N., Gómez, E., Usubillaga, A. and Darghan, E., 2005. Effect of the fertilization, plant density and time of cutting on yield and quality of the essential oil of *Cymbopogon citratus* Stapf. Revist de la Facultad de Agron Ća LUZ. 22: 247-260.

Liu, Z., Ostrovsky, I., Farnsworth, P.B. and Lee, M.L., 1993. Instrumentation for comprehensive two-dimensional Supercritical Fluid-Gas Chromatography. Chromatographia, 35: 567-573.

Majewska, E., Kozlowska, M., Gruszczynska-Sekowska, E., Kowalska, D. and Tarnowska, K., 2019. Lemongrass (*Cymbopogon citratus*) essential oil: extraction, composition, bioactivity and uses for food preservation-a review. Polish Journal of Food and Nutrition Sciences: *69*(4).

Mallouhi, N. and Bris, B., 1995. Impact des Engrais sur la Production du Gingembre et de la Citronnelle Cultives sur Sols Ferralitique et Ferrugineux. Bulletin de la Recherche Agronomique 11 : 15-22.

Masada, Y., 1976. Analysis of essential oils by GC and MS. John Willey and Sons, Chichester.

MEPN (Ministère de l’Environnement et de la Protection de la Nature), 2008. Programme d’action national d’adaptation aux changements climatiques du Bénin. Convention-cadre des Nation Unies sur les changements climatiques, Cotonou, Bénin, 81p.

MINITAB, 2019. Release 19.1 Minitab Inc., USA. ISBN 0-925636-48-7.

Mishra, C., Khalid, M.A., Fatima, N., Singh, B., Tripathi, D., Waseem, M. and Mahdi, A.A., 2019. Effects of citral on oxidative stress and hepatic key enzymes of glucose metabolism in STZ/HFD induced diabetic dyslipidemic rats. Iran J Basic Med Sci 2019; 22:49-57.

Mondello, L., Dugo, G. and Bartle, K.D., 1996. On-line

HPLC-HRGC-MS for the analysis of natural complex mixtures. J. Chromatogr. Sci. 34:174-181.

Mukarram, M., Choudhary, S., Khan, M.A., Poltronieri, P., Khan, M.M.A., Ali, J., Kurjak, D. and Shahid, M., 2021. Lemongrass Essential Oil Components with Antimicrobial and Anticancer Activities. Antioxidants 2022, 11, 20.

Pandey, S., 2024. An Exploration of *Cymbopogon nardus* or *Cymbopogon winterianus* and its Oil. International Journal of Novel Research and Development 9(8): b859-b874. ISSN: 2456-4184.

Pélabon, C., Hilde, C.H., Einum, S. and Gamelon, M., 2020. On the use of the coefficient of variation to quantify and compare trait variation. Evolution Letters 4-3: 180–188.

Pengelly, A., 2004. The Constituents of medicinal plants (Eds): An Introduction to the chemistry and therapeutics of herbal medicine. United Kingdom: CABI Publishing. pp. 85-103.

Pham-Tuan, H., Vercammen, J. and Sandra, P., 2001. A versatile robotic arm for static headspace sampling with SPME. LC–GC Europe, 14: 215-228.

Salsabila, D.U., Wardani, R.K., Hasanah, N.U., Tafrihani, A.S., Zulfin, U.M., Ikawati, M. and Meiyanto, E., 2023. Cytoprotective Properties of Citronella Oil (*Cymbopogon nardus* (L.) Rendl.) and Lemongrass Oil (*Cymbopogon citratus* (DC.) Stapf) through Attenuation of Senescent-Induced Chemotherapeutic Agent Doxorubicin on Vero and NIH-3T3 Cells. Asian Pac J Cancer Prev, 24 (5), 1667-1675.

Sangwan, N.K, Dhindsa, K.S., Malik, O.P., Sharma, G.D. and Paroda, R.S., 1982. Quantitative changes in levels of essential oil in *C. martinii* var *motia* during different growth stages and on ageing the harvested crop in the field and laboratory. In: Proc. Nat. Sem. Med. Arom. Plants., India.

Sargenti, S.R. and Lancas, F.M., 1997. Supercritical fluid extraction of *Cymbopogon citratus* (DC.) Chromatographia, 46: 285-290.

SAS Institute, 2007. JMP User Guide, Release 7. SAS Institute Inc., Cary, NC, USA. ISBN 978-1-59994-408-1.

Schaneberg, B.T. and Khan, I.A., 2002. Comparison of extraction methods for marker compounds in the essential oil of lemongrass by GC. J. Agric. Food Chem. 50: 1345-1349.

Shah, G.; Shri, R.; Panchal, V.; Sharma, N.; Singh, B. and Mann, S., 2011. Scientific basis for the therapeutic use of *Cymbopogon citratus* Stapf (Lemon grass). J. Adv. Pharm. Technol. Res. 2011, 2, 3–8.

Sharma, S., Sharma, S. and Soni, K., 2020. Lemongrass: its uses, medicinal and industrial properties. Just Agriculture 1(3): 1-10.

Silva, C.F., Moura, F.C., Mendes, M.F., Pessoa, F.L.P., 2011. Extraction of Citronella (*Cymbopogon nardus*) Essential oil using Supercritical Carbon Dioxide: Experimental Data and Mathematical Modelling. Brazillian Journal of Chemical Engineering 28(2), 343-350.

Singh, A. and Kumar, A., 2017. Cultivation of Citronella (*Cymbopogon winterianus*) and evaluation of its essential oil, yield and chemical composition in Kannauj region. International Journal of Biotechnology and Biochemistry 13(2): 139-146.

Singh, N., Luthra, R., Sangwan, R.S. and Thakur, R.S., 1989. Metabolism of monoterpenoids in aromatic plants. Current Res. Med. Aroma. Plants. 11: 174-197.

Sinsin, B. and Owolabi, L., 2001. Monographie Nationale de la Diversité Biologique. Rapport de Synthèse. MEHU/PNUD, Cotonou, Bénin.

Sivamaruthi, B.S., Kesika, P. and Chaiyasut, C., 2022. The composition, pharmacological and economic importance of essential oil of *Litsea cubeba* (Lour.) Pers. Food Sci. Technol, Campinas, v42, e35720, 2022 1.

Spriha, R.S. and Kumar, A., 2021. Lemongrass a Wonder Herb of Poaceae Family: An Overview. Biological Forum – An International Journal 13(2): 298-308.

Tajidin, N.E., Ahmad, S. H, Rosenani, A.B., Azimah, H. and Munirah, M., 2012. Chemical composition and citral content in lemongrass (*Cymbopogon citratus*) essential oil at three maturity stages. African Journal of Biotechnology 11(11) : 2685-2693.

Tarka, M., Bolstad, G.H.S., Wacker, K.R., Hansen, T.F. and Pélabon. C., 2015. Did natural selection make the Dutch taller? A cautionary note on the importance of quantification in understanding evolution. Evolution 69:3204–3206.

Utz, H.F., 2011. PLABSTAT: Ein Computerprogramm zur statistischen Analyse von pflanzenzüchterischen Experimenten. Version 3A, Institut für Pflanzenzüchtung, Saatgutforschung und Populationsgenetik, Universität Hohenheim, Germany.

Vélez, P.R.C., Ali, A., Fournier-Level, A., Dunshea, F. and Jusuf, P.R., 2022. P06-04 Antioxidant activity and embryotoxicity of *Citrus australasica*, *Tasmannia lanceolata* and *Diploglottis australis* extracts in zebrafish. Toxicol. Lett. 2022, 368, S114.

Zanklan, A.S. and Baba-Moussa, F.A.K., 2025. Effects of Diverse Fertilizations on Rhizome Yields, their Essential Oil Content and its Phytochemical Composition in Ginger (*Zingiber officinale* Roscoe) Cultivated in the Southern of Benin Republic. Int.J.Curr.Microbiol.App.Sci. 14(01): 124-147.

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