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ORIGINAL RESEARCH ARTICLE

Bioactivity of essential oils of *Cymbopogon citratus* (DC) Stapf and *Cymbopogon nardus* (L.) W. Watson from Benin against *Dinoderus porcellus* Lesne (Coleoptera: Bostrichidae) infesting yam chips

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Abstract

Dinoderus porcellus L. (Coleoptera: Bostrichidae) is a most abundant and damageable pest of stored yam chips in West Africa. In view of the negative effects related to the use of synthetic chemical insecticide, it is important to develop alternative eco-friendly methods of control, such as the use of plant essential oils. The chemical composition and biological effects (repellent, antifeedant, contact and fumigant toxicity) of essential oils from *Cymbopogon citratus* (DC) Stapf and *Cymbopogon nardus* (L.) W. Watson were evaluated for the first time against *D. porcellus*. The chemical composition of *C. citratus* essential oil revealed that neral (24.64%), geranial (23.46%), and beta-pinene (21.90%) were the predominant compounds whereas citronellal (37.87%), nerol (19.88%) and citronellol (9.11%) were identified in *C. nardus* essential oil. Data showed that both essential oils were more effective as repellents and fumigant than the commercial insecticide Actellic 50 EC. The results also revealed the low contact toxicity of *C. citratus* and *C. nardus* essential oils against *D. porcellus*. However, similarly to Actellic 50 EC, *C. citratus* essential oil presented good antifeedant activity against *D. porcellus* and reduced yam chips weight loss. The results obtained suggest the good potential of C. *citratus* as both antifeedant and fumigant toxic agent against *D. porcellus*. While, *C. nardus* essential oil could be recommended as repellent. However, further studies are required to evaluate the insecticidal activity of both Cymbopogon essential oils under farmers' storage conditions and to develop a good formulation as biopesticide.

Keywords Insecticidal activity · Botanical extract · Citronella grass · Lemongrass · Yam chips

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Introduction

Yam (*Dioscorea* spp) is an important food crop largely produce in West Africa (91% of worldwide production) and specifically in Benin where it contribute to food security and alleviate poverty (FAO 2019). This crop, which produce edible starchy storage tubers, is a good source of carbohydrates, proteins, and both macro minerals (Ca, P, K and Mg) and micro minerals (Fe, Cu, Zn, Se and Mn) (Otegbayo et al. 2017, Olatoye and Arueya 2019). Yam tubers is consumed in different forms, mainly boiled, fried, or often dried and milled into flour for various products (Adepoju et al. 2018). Yam is a cash crop, which offers employment and income to millions of value chain actors and plays an important role in the livelihood people in West Africa (Agba et al. 2019). In addition, yam is anchored in the

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sociocultural life of the populations as evidenced by the many festivals organized for the release of new yams (Osunde and Orhevba 2009).

Unfortunately, yam tubers are high perishable and post-harvest loss can rise up to 25% of the raw weight (Ferraro et al. 2016). To overcome this constraint yam tubers are traditionally processing in dehydrated chips through peeling, slicing, blanching, steeping, and sun-drying steps (Omohimi et al. 2017). However, stored yam chips are very susceptible to insect attacks which causes an important quantitative (reducing them to powdery waste) and qualitative (unsuitable for human consumption) losses in few months (Loko et al. 2019a, b). In West Africa, and particularly in Benin, *Dinoderus porcellus* Lesne (Coleoptera: Bostrichidae) remains by far the most abundant and most damaging storage pest to yam chips (Osuji 1980, Loko et al. 2013). The use of synthetic chemical pesticides is the main method used by farmers and traders to protect yam chips (Loko et al. 2013). However, very often farmers use unadapt chemicals, which have remarkably toxic properties and acute poisoning tendency such as DDT (Dichlorodiphenyltrichloroethane (Sosan and Oyekunle 2017, Pii et al. 2019). It is therefore urgent to develop alternative pest management measures respecting human and the environmental health.

Plant essential oils, which can have several negative effects on insect pests (repellence, antifeedant, insecticidal) appears as a suitable alternative to the use of chemical insecticides for yam chips protection. In fact, their easy biodegradation in the environment and their non-toxicity towards mammals make plant essential oils into good biopesticides (Caballero-Gallardo et al. 2012). Despite these advantages, very few studies, if any, have evaluated the susceptibility of *D. porcellus* to plant essential oils. Among potential aromatic plants, lemon grass (*Cymbopogon citratus* (DC) Stapf) and citronella grass (*Cymbopogon nardus* (L.) W. Watson) appears as the good candidate because widely distributed in Benin. Although some studies revealed the repellent and/or toxic effects of *C. citratus* and / or *C. nardus* essential oils on several stored product insects such as *Tribolium castaneum* Herbst (Olivero-Verbel et al. 2010, Bossou et al. 2015, Ahmad et al. 2018), *Sitophilus oryzae* L. (Franz et al. 2011), *Sitophilus zeamais* Motschulsky (Kabera et al. 2011, Doumbia et al. 2014), *Tenebrio molitor* L. (Wang et al. 2015), *Callosobruchus maculatus* F. (Alves et al. 2019), *Rhyzopertha dominica* F. (Doumbia et al. 2014), their use in the management of *D. porcellus* in stored yam chips is hitherto unexplored. Therefore, this study was carried out to reveal the chemical composition and evaluate the repellent, fumigant, contact toxicity, and antifeedant activity of *C. citratus* and *C. nardus* essential oils against *D. porcellus*.

Material and methods

Plant materials

The plant material consists of the leaves of *C. citratus* collected at Cotonou (6° 21' 55" N, 2° 25' 0.5" E) and, *C. nardus* collected at Natitingou (10° 18′ 14″ N, 1° 22′ 46″ E) in March 2018. The plant specimens were identified by botanist at the National Herbarium of the University of Abomey-Calavi, Benin. The authenticated voucher specimens were deposited in the Benin national herbarium under the numbers AA6635/HND and AA6637/HND for *C. citratus* and *C. nardus* respectively.

Essential oil extraction

The collected lemongrass and citronella grass leaves were washed with distilled water and dried at 16 °C in the laboratory in the shade of the sunlight during two weeks. The dried leaves (100 g) of each species were subjected to hydrodistillation (in 500 mL of distilled water) using a Clevenger type apparatus for 3 h. Essential oils were separated from the water by decantation and, dried with magnesium sulfate ($MgSO_4$). Essential oils were stored in tightly closed dark vial at 4 °C until further tests. Essential oil extraction from each plant was repeated three times and the average yield was determined. The yields were calculated according to the volume of essential oil obtained relative to the weight of the plant material before distillation.

Gas chromatography–mass spectrometry analysis

The chemical profile of the essential oils of *C. citratus* and *C. nardus* obtained was analysed by gas chromatography coupled with mass spectrometry (GC/MS). GC/MS were performed on a GC 2000 TRACE series (Thermo Quest, Rodano, Italy), equipped with an AS2000 autosampler (Thermo Quest). The GC system was coupled to a Trace MS (Thermo Quest) type mass spectrometer operating in the electronic impact mode. The GC/MS was equipped with a capillary column DB-WAX 122–7032 (Agilent) of dimensions 30 × 0.25 mm with 0.25 μm internal diameter. The samples were injected in splitless mode (injected volume: 1 μl, inlet temperature: 230 °C). Helium was used as the carrier gas at a constant rate of 1.3 mL/min. The coupling temperature of the GC was 260 °C. The ionization mode electron impact was 70 eV and the transfer line temperature was 250 °C. The mass spectra data was recorded and analysed with Xcalibur 1.1 software (Thermo Quest). The compounds were identified by comparison with those of reference compounds of the National Institute of Standards and Technology mass spectrometry library (NIST / EPA / NIH 98) (library similarity and reverse match > 800).

Rearing of *D. porcellus*

The beetle *D. porcellus* collected from infested yam chips purchased at the market of Dassa-Zoumé was reared on healthy yam chips as described by Loko et al. (2019a, b). The rearing material used is made of plastic containers (10.5 cm height, 16.8 cm diameter) which were covered by a fine mesh cloth for ventilation. Dried yam chips (500 g) were infested in the plastic boxes with 50 unsexed adults of *D. porcellus*. The plastic containers were kept in the laboratory under temperature conditions of 25 ± 2 °C, relative humidity of 70 ± 5%, and photoperiodicity of 12 L/12D (Oni and Omoniyi 2012). After two weeks, adult beetles have been removed from the rearing boxes in order to obtain a F1 progeny that will be used for all experiments (Loko et al. 2019a, b).

Repellent activity

The repellent effect of the essential oils of *C. citratus* and *C. nardus* against *D. porcellus* was evaluated using the area preference method on Whatman filter paper (diameter 9 cm) described by Alcala-Orozco et al. (2019). Four doses (5, 10, 15 and 20 µl) of each essential oil were prepared by dilution in 0.5 ml of acetone. While, the commercial insecticide Actellic 50 EC (Pirimiphos-Méthyl) intended for the protection of stored products against pest attack was used as positive control with a recommend concentration of 5 mL/m^2. Each filter paper was divided into two equal part, one-half (31.8 cm^2) was treated with each essential oil diluted solution and the other half was treated with acetone only (negative control). Both pieces of filter paper were dried for 10 min at room temperature to allow evaporation of the solvent. Treated and untreated halves were fixed using adhesive tape and placed in a Petri dish (diameter 9 cm). Twenty mixed-sex adult of *D. porcellus* were released at the center of each Petri dish. The dishes were then covered and placed in continual darkness. Four replications were used for each concentration of essential oils. The number of *D. porcellus* present on the treated and untreated part of filter paper were recorded after 30 min, 2 h, 4 h and, 8 h of exposure (Lale and Alaga 2001).

$$\text{Percentage of repellence} = [(\text{Nc}-\text{Nt})/(\text{Nc}+\text{Nt})] \text{ x } 100$$

where Nc was the number of insects on the untreated area after the exposure interval and Nt was the number of insects on the treated area after the exposure interval. The average repulsion value of each essential oil was assigned to repulsion classes from 0 to V: class 0 (PR ≤ 0.1%), class I (PR = 0.1–20%), class II (PR = 20.1–40%) Class III (40.1–60%), Class IV (60.1–80%) and Class V (80.1–100%). The repellency index (RI) was calculated with the formula:

$$\text{RI} = 2\,\text{g}/\text{G} + \text{P}$$

where G = percentage of insects attracted to the treatment and P = percentage attracted to the control. The RI values range between zero and two (Gusmao et al. 2013), and RI = 1 indicates similar repellency between the treatment and the control (neutral treatment), RI > 1 indicates lower repellency of the treatment compared to the control (attractive treatment) and RI < 1 corresponds to a greater repellency of the treatment compared to the control (repellent treatment) (Padin et al. 2013).

Contact toxicity

The insecticidal activity of lemongrass and citronella grass essential oil against *D. porcellus* adults was evaluated by the topical application method (Hu et al. 2019). Thus, four solutions of each essential oil (1, 2, 4 and 8 µl / mL of acetone) were prepared by diluting known amounts of oil in acetone. A volume of 0.5 µl of each dilution was topically applied onto the thorax of individual adults (3–5 days old) using a micropipette applicator. Insects treated with only acetone served as a negative control and those treated with the contact insecticide Actellic 50 EC as positive control at the recommended dose (100 mL of Actellic in 10 L of sterilized water). Controls and each dilutions of lemongrass and citronella grass essential oils were replicate six times with ten unsexed adults in each replication. After treatment the insects were transferred into glass dishes (9 cm diameter) containing healthy yam chips. The dishes were then be placed in the dark at 26 ± 2 °C and 75 ± 5% relative humidity. The number of dead insects was counted 24, 48 and 72 h after treatment and the corrected mortality percentage was calculated using Abbott's formula (2) to eliminate natural mortalities.

$$\text{Percent mortality} = \frac{\textit{Number of dead D.porcellus}}{\textit{Number of introduced D.porcellus}} \times 100 \quad (1)$$

Corrected mortaliy

$$= \frac{(\%\textit{of death in treadted} - \%\textit{death in control})}{(100 - \%\textit{death in control})} \times 100 \quad (2)$$

Antifeedant activity

The antifeedant activity of lemongrass and citronella grass essential oils was evaluated according to the methodology described by Descamps et al. (2011) with slight modifications. Four concentrations of each essential oil (2, 4, 8 and 16 µl/ml of acetone) were tested. Dried yam chips (10 g) were treated with each essential oils concentration and air dried to allow the solvent to evaporate for 15 to 20 min. All dishes were infested with twenty (2–3 day old) adults of *D. porcellus* starved for

3 h (Taghizadeh et al. 2014). Insects treated with only acetone served as a negative control and those treated with the antifeedant insecticide Actellic 50 EC as positive control at the recommended dose (0.01% in sterilized water). Each treatment was replicated six times. The counts of dead insects were made at 1, 3, 7, 14 and 21 days after exposure (Othira et al. 2009). The percentage of insect mortality was calculated according to Eqs. (1) and (2). The weight loss percentage was calculated according to the following equation:

$$\text{Weight loss}\ (\%) = \frac{\text{Initial weight} - \text{Final weight}}{\text{Initial weight}} \quad (3)$$

The feeding deterrence index (FDI) was calculated following Isman et al. (1990) using the formula:

$$FDI(\%) = \frac{consumption\ of\ untreated\ yam\ chips - consumption\ of\ treated\ yam\ chips}{consumption\ of\ untreated\ yam\ chips} \times 100$$

Positive values expressed a feeding deterrent effect and negative values expressed a feeding stimulant effect (Stefanazzi et al. 2011).

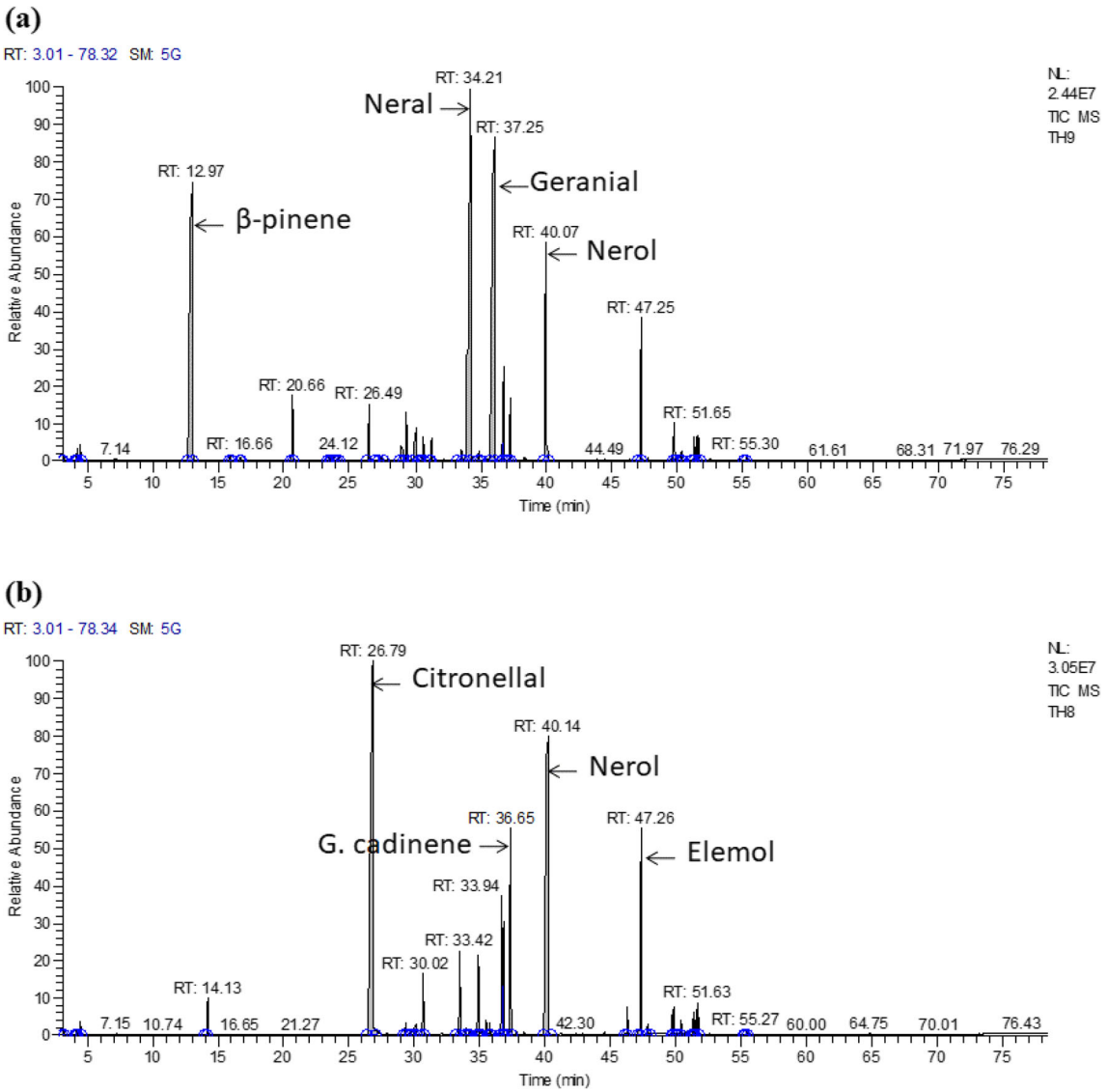

Fig. 1 GC-MS chromatograms of (**a**) *Cymbopogon citratus* essential oil, (**b**) *Cymbopogon nardus* essential oil

Table 1 Main compounds in *Cymbopogon citratus* and *Cymbopogon nardus* essential oils. Compounds that are more abundant are in bold font

Compounds	RT/GC-MS	RSI	Molecular weight	Percentage
Essential oil of *C. nardus*				
Limonene	14.13	952	136	1.21
Citronellal	26.79	934	154	**37.87**
Beta-elemene	30.63	929	204	1.86
cis-2,6-Dimethyl-2,6-octadiene	33.42	922	138	2.71
Germacrene D	34.88	939	204	2.86
Alpha-Cadinene	35.44	944	204	0.49
Gamma-cadinene	36.65	902	204	4.65
Geranyl Acetate	36.80	941	196	4.41
Citronellol	37.30	952	156	**9.11**
Nerol	40.14	880	154	**19.88**
Germacrene D-4-ol	46.18	862	222	0.75
Elemol	47.26	953	222	**7.4**
Eugenol	49.66	929	164	0.57
Gayol Acetate	48.82	889	222	1.03
Alpha-Eudesmol	51.26	924	222	0.64
Beta-Eudesmol	51.50	942	222	0.68
Alpha-cadinol	51.63	942	222	0.89
			Total	**97.01**
Essential oil of *C. citratus*				
Beta-pinene	12.97	911	136	**21.90**
5-Hepten-2-one, 6-methyl	20.66	947	126	1 0.70
Citronellal	26.49	916	154	1.52
Linalool	29.31	957	154	1.19
Verbenol	30.04	858	152	1.11
Neral (Citral B)	34.21	900	152	**24.64**
Geranial (Citral A)	36.05	896	152	**23.46**
Geranyl Acetate	36.81	958	196	2.61
Citronellol	37.25	957	156	1.49
Nerol	40.07	887	154	**8.54**
Elemol	47.25	954	222	3.84
			Total	**92.0**

RT: Retention Time; RSI value: Reverse match (matching coefficient (> 800 compared to the NIST database))

Fumigant toxicity

Fumigation bioassays were carried out as reported by Kim et al. (2013) with slight modifications. Ten pairs of *D. porcellus* adults were placed in 250 mL glass vial containing 10 g of yam chips and fitted with a screw cap. An aliquot of 5, 10, 20 and 40 µl of each essential oil dissolved in acetone corresponding to doses of 20, 40, 80 and 160 µl / L of air respectively were uniformly applied to a disk of Whatman filter paper (2 cm in diameter) (Nattudurai et al. 2015). The impregnated filter paper was allowed to dry for 2 min, before being suspended on the underside of the screwcap of the glass vial with adhesive tape. The caps of the glass vials were screwed tightly and placed in the dark. Filter paper disk treated with only acetone served as negative control and the commercial insecticide Actellic 50 EC with a recommend concentration of 5 mL/m^2 was used as positive control. Each treatment was replicated six times. Insect mortality was observed daily for one week. The percent mortality of the insects was calculated according to Eqs. (1 and 2).

Statistical analysis

Data on corrected percent mortality and repellence were first homogenized using arcsine-transformation to correct for heterogeneity of treatment variance before being subjected to ANOVA using IBM SPSS Statistics version 25 software package, and means were separated using Student Newman Keuls test. Data obtained from various concentration-response bioassays were further log-

Table 2 Percent repellence (mean ± SEM) of *Dinoderus porcellus* adults and the repellent class of essential oils of *Cymbopogon citratus* and *Cymbopogon nardus* leaves at different concentration to varying exposure time

Essential oils	Concentration (µL/cm^2)	Percentage of repellence of treatments after				Mean repellence	Repellence class	Repellence index	Classification
		30 min	2 h	4 h	8 h				
C. citratus	5	60.0 ± 4.8ab	65.0 ± 5.0a	70.0 ± 5.8c	74.9 ± 6.4b	67.5 ± 2.8c	IV	0.3 ± 0.1	Repellent
	10	57.5 ± 15.5ab	50.0 ± 10.8a	35.0 ± 18.9bc	52.5 ± 14.9ab	48.8 ± 7.2bc	III	0.5 ± 0.3	Repellent
	15	47.5 ± 13.8ab	50.0 ± 17.3a	60.0 ± 16.3bc	52.5 ± 12.5ab	52.5 ± 6.9bc	III	0.5 ± 0.2	Repellent
	20	25.0 ± 9.6a	57.5 ± 16.5a	36.4 ± 3.8bc	52.5 ± 4.8ab	42.8 ± 5.6b	III	0.6 ± 0.2	Repellent
C. nardus	5	17.5 ± 4.8a	32.5 ± 4.8a	5.0 ± 2.9a	30.0 ± 4.1a	21.3 ± 13.6a	II	0.8 ± 0.1	Repellent
	10	43.9 ± 4.7ab	45.0 ± 8.7a	20.0 ± 4.1ab	40.0 ± 7.1ab	37.2 ± 15.5b	II	0.6 ± 0.2	Repellent
	15	57.5 ± 2.5ab	52.5 ± 4.8a	42.5 ± 2.5bc	37.5 ± 6.3ab	47.5 ± 11.3bc	III	0.5 ± 0.1	Repellent
	20	72.5 ± 4.7b	62.5 ± 4.8a	42.5 ± 4.8bc	47.5 ± 2.5ab	56.3 ± 4.5bc	III	0.4 ± 0.1	Repellent
Actellic 50 EC	0.5	44.7 ± 8.3ab	43.4 ± 2.6a	30.7 ± 3.3bc	62.8 ± 8.3ab	45.4 ± 16.3b	III	0.5 ± 0.1	Repellent
F		3.281	0.820	4.782	2.471	7.199			
p-value		0.010	0.592	0.001	0.038	0.000			

Means within the same rows followed by the same letter are not significantly different ($p < 0.05$)

transformed before being subjected to probit regression analysis using XLSTAT version 2019.3.2 software (Addinsoft 2019). LD_{50} (lethal dose that killing 50% of exposed insects), LC_{50} (lethal concentration that kills 50% of exposed insects) or RD_{50} (dose that repels 50% of exposed insects) values and corresponding 95% fiducial limits were obtained from derived regression equations. The LC_{50}, LD_{50} and RD_{50} values in a column were considered significantly different when 95% fiducial limits did not overlap.

Results

Chemical composition

The essential oil yields of *C. citratus* and *C. nardus* were 1.1% and 1.3% (v/w) respectively. GC-MS analyses of essential oil of *C. citratus* oil showed the presence of 11 compounds (Fig. 1a), which formed 92% of the total oil composition. While, 17 chemical compounds were identified in *C. nardus* essential oil (Fig. 1b), representing 97.01% of the detected compounds (Table 1). The chemical composition of *C. citratus* essential oil revealed that neral (24.64%), geranial (23.46%), and beta-pinene (21.90%) were the predominant compounds. While, the essential oil of *C. nardus* is mainly composed of citronellal (37.87%), nerol (19.88%), citronellol (9.11%), elemol (7.40%), and gamma-cadiene (4.65%).

Repellent activity

The results showed that both essential oils of *C. citratus* and *C. nardus* act as repellent against *D. porcellus* even at low concentration (Table 2). Contrary to *C. citratus* essential oil, the repellent activity of *C. nardus* essential oil was dose-dependent (df = 3, 63; F = 37.35; P = 0.000) and significantly increase with the exposure time (df = 3, 63; F = 16.84; P = 0.000). In addition, there is significant interaction between

Table 3 Repellency of the essential oils of *Cymbopogon citratus* and *Cymbopogon nardus* leaves at various periods

Essential oil	Exposure time	RD_{50} (µL/cm^2)	95% fiducial limits	Slope ± SEM[a]	Intercept	R	P-value
C. citratus	30 min	14.63	6.5–32.7	-1.129 ± 0.18	6.315	0.72	0.551
	2 h	59.4	10.7-331.2	-0.537 ± 0.38	5.952	0.77	0.952
	4 h	26.7	8.6–83.0	-0.802 ± 0.25	6.155	0.37	0.435
	8 h	52.9	15.0-183.5	-0.749 ± 0.28	6.291	0.78	0.930
C. nardus	30 min	9.1	5.7–14.5	2.007 ± 0.10	3.075	0.98	0.965
	2 h	5.6	1.39–11.45	0.880 ± 0.23	4.470	0.96	0.998
	4 h	14.94	7.82–28.51	1.417 ± 0.14	3.337	0.94	0.850
	8 h	5.51	0.76–39.63	0.456 ± 0.43	4.662	0.79	0.952

[a] SEM: Standard error of mean

Table 4 Mean corrected mortality rates of *Dinoderus porcellus* after topical application of essential oils of *Cymbopogon citratus* and *Cymbopogon nardus* leaves and weight loss (mean ± ES) of yam chips 3 days after treatment

Essential oils	Concentration (µL/mL)	Corrected mortality rate (% ± SEM) after different periods of exposure			Mean
		24 h	48 h	72 h	
C. citratus	1	6.5 ± 4.4a	4.6 ± 4.6ab	8.7 ± 1.8a	6.6 ± 8.9a
	2	6.7 ± 3.3a	6.7 ± 2.1ab	8.5 ± 4.2a	7.3 ± 7.7a
	4	8.5 ± 1.7a	10.0 ± 3.7ab	22.6 ± 3.9a	13.7 ± 9.9abc
	8	19.1 ± 3.4a	17.3 ± 1.7b	25.9 ± 4.0a	20.8 ± 8.3c
C. nardus	1	17.2 ± 5.3a	2.3 ± 3.3a	6.7 ± 3.6a	7.3 ± 12.5a
	2	17.0 ± 4.3a	7.2 ± 3.6ab	8.5 ± 3.1a	10.9 ± 9.7ab
	4	16.9 ± 4.2a	16.3 ± 3.7b	11.9 ± 3.1a	15.0 ± 8.8bc
	8	23.9 ± 4.1a	13.6 ± 4.7ab	13.7 ± 2.0a	17.4 ± 9.9abc
Actellic 50 EC	100	64.8 ± 7.8b	71.9 ± 5.4c	84.0 ± 6.3b	73.6 ± 17.2d
F		8.955	14.635	22.216	41.388
p-value		0.000	0.000	0.000	0.000

Means within the same rows followed by the same letter are not significantly different (< 0.05)

concentration and exposure time (df = 9, 63; F = 3.00; P = 0.006) on the repellent activity of *C. nardus* essential oil against *D. porcellus*. However, *C. citratus* essential oil was significantly more repellent (df = 2, 143; F = 4.65; P = 0.011) than *C. nardus* essential oil and the commercial insecticide Actellic 50 EC. Treatment with *C. nardus* essential oil at 20 μLcm^{-2} showed the highest repellent activity (df = 8, 35; F = 3.28; P = 0.010) against *D. porcellus* adults after 30 min of exposure time. After this interval time, treatments with the essential oil of *C. citratus* at the lowest concentration showed the higher repellent activity against *D. porcellus* (Table 2). In addition, the pooled mean repellent activity was significantly higher at the lower concentration of *C. citratus* essential oil and ranged in repellence class IV. However, the RD_{50} values indicated that essential oil of *C. nardus* was significantly more repellent than that of *C. citratus* (Table 3).

Contact toxicity

Our results showed that, essential oils of *C. citratus* and *C. nardus* at various concentration had a low insecticidal effect by topical application to *D. porcellus* adults comparatively to the commercial insecticide Actellic 50EC (Table 4). No significant difference was observed between the two essential oils in terms of contact toxicity (df = 1, 143; F = 0.007; P = 0.934). The mortality of *D. porcellus* adults had significantly increased with the concentration of *C. citratus* (df = 3, 71; F = 9.05; P = 0.000) and *C. nardus* (df = 3, 71; F = 3.56; P = 0.019). Similarly, the time of exposition to essential oils of *C. citratus* (df = 2, 71; F = 4.68; P = 0.013) and *C. nardus* (df = 2, 71; F = 5.63; P = 0.006) had significant effect on mortality of *D. porcellus* adults. However, no significant interaction between concentration of essential oils of *C. citratus* (df = 6, 71; F = 0.47; P = 0.826) or *C. nardus* (df = 6, 71; F = 1.02; P = 0.416) and time exposure was observed. The highest mortality of *D. porcellus* adults was observed with *C. citratus* essential oil at the concentration of 8 µl/mL after 72 h of exposure time (Table 4). At 24 and 48 h of exposure time, *C. nardus* essential oil was more toxic against *D. porcellus* adults than *C. citratus* oil (Table 5). However, at 72 h after exposure time, the LD_{50} values shown that *C. citratus* essential oil were more toxic against *D. porcellus* adults than *C. nardus* (Table 5).

Table 5 Contact toxicity of the of essential oils of *Cymbopogon citratus* and *Cymbopogon nardus* leaves against *Dinoderus porcellus* adults

Essential oil	Exposure time (h)	LD_{50} (µL/adult)	95% fiducial limits	Slope ± SEM[1]	Intercept	R	P-value
C. citratus	24	322.1	45.6-2276.2	0.636 ± 0.43	3.391	0.75	0.612
	48	179.5	33.4-965.4	0.748 ± 0.37	3.313	0.97	0.945
	72	41.7	12.3-141.5	0.905 ± 0.27	3.535	0.85	0.601
C. nardus	24	226.9	22.2-2320.8	0.442 ± 0.52	3.944	0.60	0.593
	48	30.1	11.5–78.8	1.282 ± 0.21	3.101	0.78	0.194
	72	549.7	65.9-4584.7	0.605 ± 0.47	3.342	0.99	0.993

[1] SEM: Standard error of mean

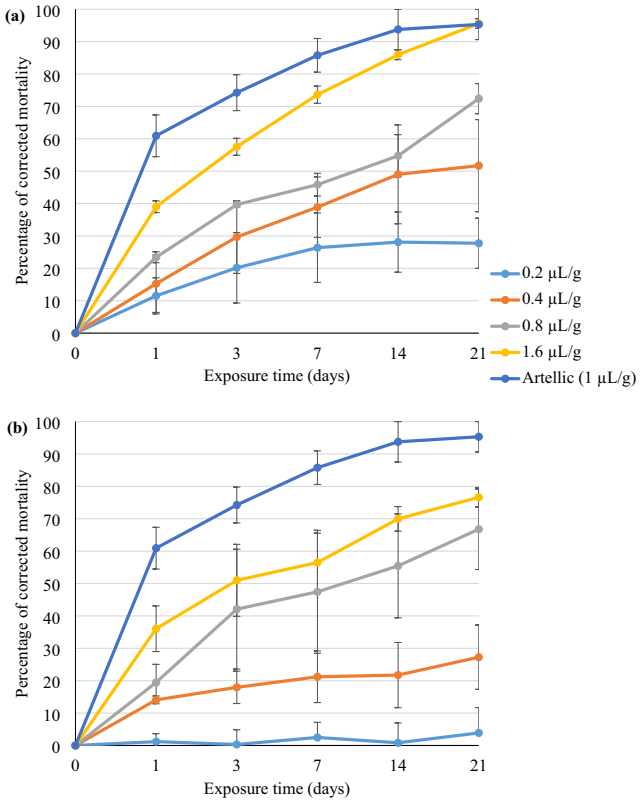

Fig. 2 Corrected mortality rate of *Dinoderus porcellus* after ingestion of yam chips treated with (**a**) *Cymbopogon citratus* essential oil, (**b**) *Cymbopogon nardus* essential oil. Vertical bars indicate standard error of mean

Ingestion toxicity

The consumption of yam chips treated with essential oil of *C. citratus* and *C. nardus* induced significant mortality of *D. porcellus* adults at the higher concentration (Fig. 2). The ingestion toxicity of *C. citratus* essential oil was higher (df = 1, 159; F = 8.58; P = 0.004) than those of *C. nardus*. The mortality of *D. porcellus* adults had significantly increased with the concentration of *C. citratus* (df = 3, 79; F = 29.75; P = 0.000) and *C. nardus* (df = 4, 79; F = 2.73; P = 0.037). Similarly, the mortality of *D. porcellus* adults induced by yam chips treated with essential oils of *C. citratus* (df = 4, 79; F = 15.37; P = 0.000) and *C. nardus* (df = 2, 71; F = 5.63; P = 0.006) significantly increased with time. However, there is no significant interaction between concentration of essential oils of *C. citratus* (df = 12, 79; F = 0.64; P = 0.800) or *C. nardus* (df = 12, 79; F = 0.58; P = 0.850) and time exposure. Except the lower concentration (0.2 µl/g) of *C. citratus* all the others tested concentrations induced a mortality of more than 50% of the population of *D. porcellus* after 21 days of exposure (Fig. 1a). The same trend was observed with the higher concentrations (0.8 µl/g and 1.6 µl/g) of *C. nardus* essential oil (Fig. 1b). The mortality caused by the commercial insecticide Actellic 50EC was significantly (df = 8, 35; F = 11.89; P = 0.000) higher than the mortality caused by the treatment of *C. citratus* and *C. nardus* essential oils at 0.2 µl/g and 0.4 µl/g, 21 days after treatment. However, this mortality was not different from those caused by the higher concentrations of *C. citratus* and *C. nardus* essential oils (0.8 µl/g and 1.6 µl/g). An estimation of LC_{50} values showed that *C. nardus* oil (LC50 = 19.4 µl/g) at 1-day interval time had the most active feeding toxicity against *D. porcellus* than *C. citratus* oil (LC_{50} = 19.7 µl/g). However, after the first day of experiment *C. citratus* essential oil had the most active feeding toxicity against *D. porcellus* than *C. nardus* (Table 6).

Figure 3 showed a significant difference (df = 9, 39; F = 44.34; P = 0.000) between the weight loss caused by *D. porcellus* to the yam chips treated with the essential oils of *C. citratus* and *C. nardus* and those of negative and positive control. Results showed that, *C. citratus* and *C. nardus* essential oils at all tested concentration decreased the consumption of yam chips by *D. porcellus* (Fig. 3). However, the feeding deterrent action of both essential against *D. porcellus* was significantly different (df = 8, 35; F = 25.31; P = 0.000) of those caused by Actellic 50 EC (Fig. 4). The maximum feed rate inhibition and the lowest weight loss were obtained for yam chips treated with essential oil of *C. citratus* at the higher

Table 6 Oral toxicity of treated yam chips by *Cymbopogon citratus* and *Cymbopogon nardus* essential oils on *Dinoderus porcellus*

Essential oil	Exposure time (days)	LC_{50} (µL/g)	95% fiducial limits	Slope ± SEM	Intercept	R	P-value
C. citratus	1	19.7	6.9–55.9	0.985 ± 0.23	3.724	0.96	0.824
	3	7.6	3.1–19.1	1.028 ± 0.20	4.092	0.98	0.949
	7	4.4	2.0-9.6	1.184 ± 0.17	4.240	0.93	0.760
	14	2.8	1.5–5.5	1.428 ± 0.14	4.354	0.93	0.746
	21	1.9	1.3–2.9	2.504 ± 0.01	4.283	0.97	0.883
C. nardus	1	19.4	7.9–47.9	1.214 ± 0.20	3.434	0.94	0.029
	3	8.2	4.2–16.3	1.478 ± 0.15	3.644	0.92	0.008
	7	6.2	3.7–10.4	2.022 ± 0.11	3.392	0.93	0.318
	14	4.2	2.5–7.1	1.857 ± 0.11	3.837	0.98	0.835
	21	3.2	2.1–4.9	2.325 ± 0.01	3.837	0.96	0.766

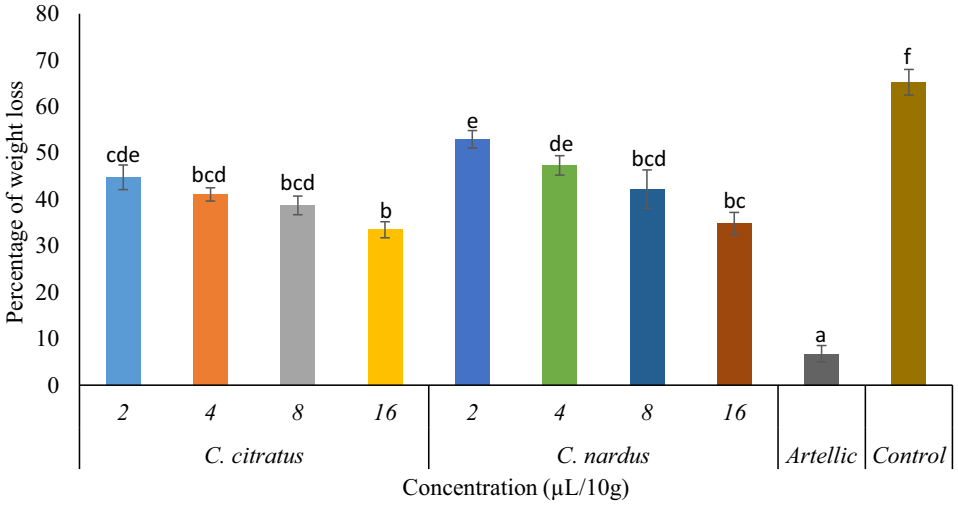

Fig. 3 Weight loss (mean ± ES) of yam chips treated with essential oils of *Cymbopogon citratus* and *Cymbopogon nardus* caused by *Dinoderus porcellus* at 21 days interval time. Different letters indicate statistical difference among weight loss ($p < 0.05$)

concentration of 1.6 µl/ml with values of 48.7% and 33.5% respectively.

Fumigant toxicity

Essential oils of *C. citratus* and *C. nardus* leaves showed high fumigant activity against *D. porcellus* adults at different concentrations and exposure times comparatively to the commercial insecticide Actellic (Fig. 5). However, the fumigation of *C. citratus* essential oil was significantly more toxic for *D. porcellus* than those of *C. nardus* oil (df = 1, 335; F = 22.87; P = 0.000). When *D. porcellus* adults were exposed to *C. citratus* essential oil, the mortality had significantly increased with concentration (df = 3, 167; F = 193.41; P = 0.000) and the exposure time (df = 6, 167; F = 51.21; P = 0.000). Similarly, mortality of *D. porcellus* adults increased significantly with the concentration (df = 3, 167; F = 118.03; P = 0.000) and exposure time (df = 6, 167; F = 48.92; P = 0.000) when exposed to *C. nardus* essential oil. There is a significant interaction between *C. nardus* essential oil concentration and exposure time (df = 18, 167; F = 2.40; P = 0.002) on *D. porcellus* mortality. The mortality values reached 80% when the *D. porcellus* adults were exposed to the high concentration of *C. citratus* and *C. nardus* oils at 7 days interval time (Fig. 5). The fumigant activity of Actellic 50 EC was low and was not significantly different from mortality caused by low concentrations both essential oils (20 and 40 µl/L air). Probit analysis showed that LC_{50} values of *C. citratus* essential oil against *D. porcellus* adults were lower than those of *C. nardus* at all exposure time (Table 7).

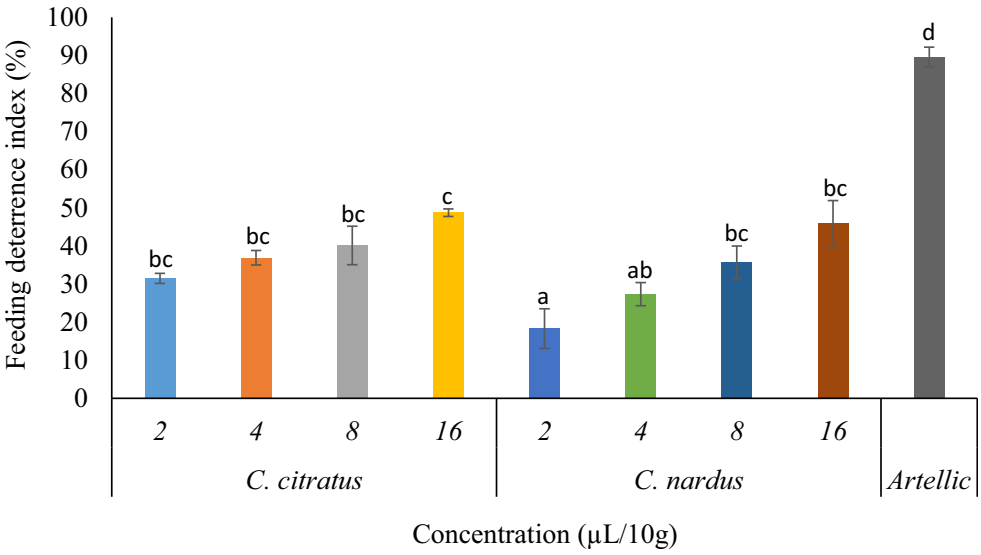

Fig. 4 Feeding deterrence index (mean ± ES) of yam chips treated with essential oils of *Cymbopogon citratus* and *Cymbopogon nardus* against *Dinoderus porcellus* at 21 days interval time. Different letters indicate statistical difference among treatment ($p < 0.05$)

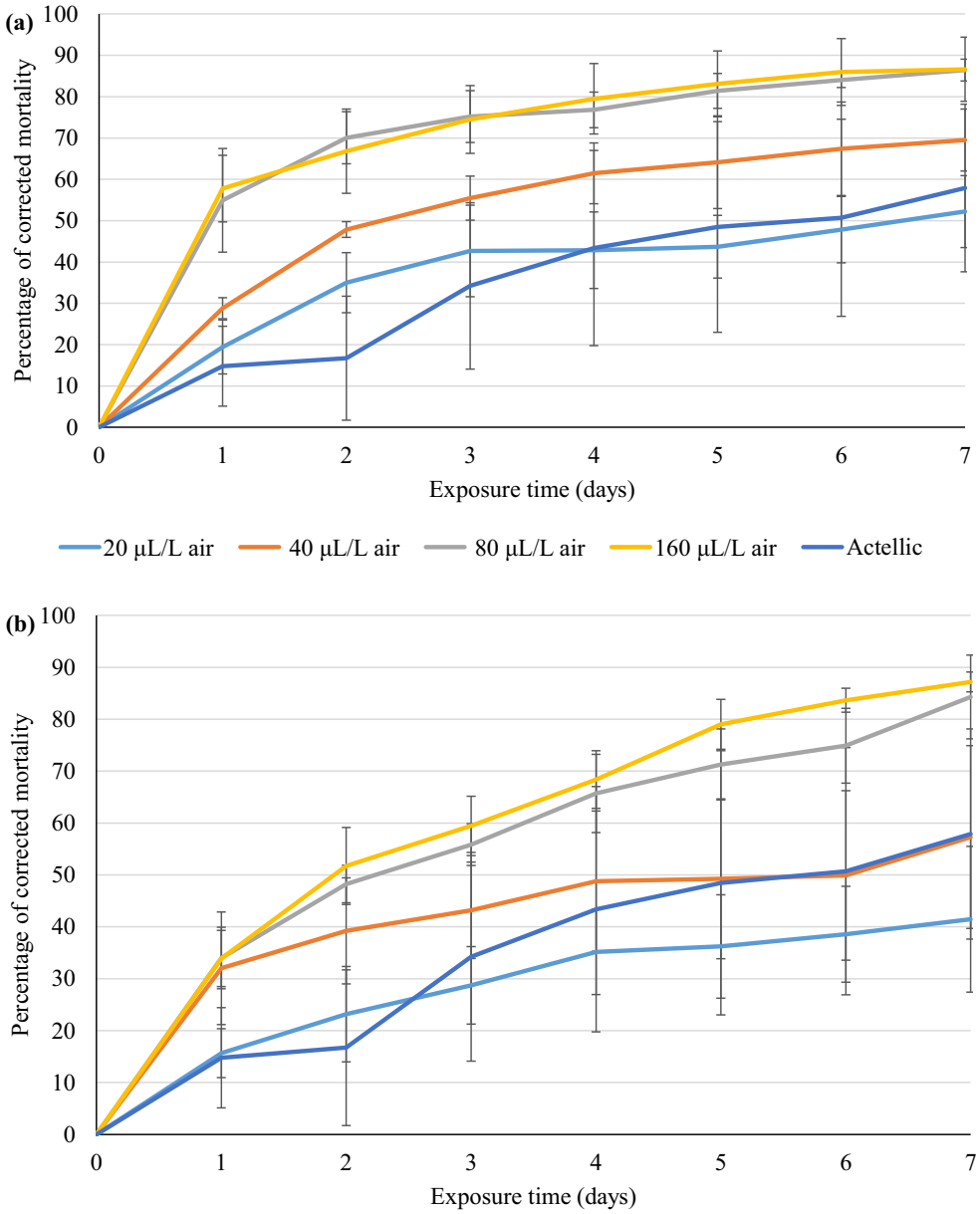

Fig. 5 Mortality (%) of *Dinoderus porcellus* adults exposed to essential oils from (**a**) *Cymbopogon citratus* and (**b**) *Cymbopogon nardus* for various periods. The mortality was corrected with formula of Abott (1925). Vertical bars indicate standard error of mean

Discussion

The *C. citratus* oil yield obtained in this study (1.1% w/w) was higher than those of leaves of this same species collected in Uganda (0.39% w/w; Ocheng et al. 2015), Benin (0.71% w/w; Kpoviessi et al. 2014) and Brazil (1.04% w/w; Costa et al. 2013), but lower than those obtained in leaves collected at Cotonou in Benin (1.7% w/w; Bossou et al. 2013). Similarly, the essential oil yield of *C. nardus* (1.3% w/w) was higher those obtained by Kpoviessi et al. (2014) with leaves collected at Abomey-calavi in Benin (1.06% w/w). The difference of oil yields could be explain by external factors such as climate, soil composition, time of collection and mode of extraction (Ocheng et al. 2015). The chemical composition of *C. citratus* essential oils rich in citral (mixture of geranial and neral) were similar to those obtained by Alves et al. (2019), Fouad et al. (2015), and Bossou et al. (2013) with slight differences in their relative abundance. Likewise, the richness of *C. nardus* essential oil in citronellal, nerol, and citronellol was similar to essential oil obtained by Setiawati et al. (2011) and Kpoviessi et al. (2014) in the same plant species collected respectively in Indonesia and Benin. However, some studies showed that geraniol is among the main component of *C. nardus* essential oil, which is not in accordance with our results (Ganjewala 2009, Nyamador et al. 2010). Environmental factors and genotypic variations could explained the difference in the chemical composition of essential oils of this plant (Pinto et al. 2015). Therefore, further

Table 7 Fumigant toxicity of essential oils from *Cymbopogon citratus* and *Cymbopogon nardus* against *Dinoderus porcellus* at various periods

Essential oil	Exposure time (days)	LC_{50} (μL/L air)	95% fiducial limits	Slope ± SEM	Intercept	R	P-value
C. citratus	1	4.8	2.3–10.2	1.261 ± 0.16	4.137	0.91	0.505
	2	2.3	0.9–5.8	0.977 ± 0.20	4.652	0.84	0.636
	3	1.5	0.6–3.8	0.978 ± 0.21	4.837	0.88	0.816
	4	1.4	0.6–3.6	1.026 ± 0.20	4.826	0.92	0.838
	5	1.1	0.5–2.4	1.282 ± 0.17	4.938	0.92	0.854
	6	0.9	0.4-2.0	1.292 ± 0.17	5.038	0.93	0.895
	7	0.7	0.3–1.7	1.226 ± 0.18	5.141	0.90	0.908
C. nardus	1	25.6	4.8-137.9	0.562 ± 0.37	4.212	0.66	0.281
	2	5.3	1.6–16.9	0.779 ± 0.26	4.439	0.91	0.695
	3	3.2	1.1–9.6	0.825 ± 0.24	4.582	0.94	0.843
	4	2.4	0.9–6.6	0.895 ± 0.22	4.660	0.94	0.861
	5	1.8	0.9–3.7	1.329 ± 0.16	4.673	0.98	0.928
	6	1.6	0.8–3.2	1.473 ± 0.14	4.696	0.97	0.857
	7	1.3	0.7–2.4	1.591 ± 0.13	4.827	0.94	0.888

studies regarding lemon grass and citronella grass essential oils quality control and standardization are required.

The both tested essential oils of Cymbopogon species showed a strong repellent against *D. porcellus*. This is not a surprising because Cymbopogon species are well known for their repellent activity against different kind of insects. The repellent activity of Cymbopogon species was also demonstrated against storage insect pests such as *Sitophilus oryzae* L. (Saljoqi et al. 2006), *T. castaneum* (Olivero-Verbel et al. 2010), *Oryzaephilus surinamensis* (L.) and *S. zeamais* (Hernandez-Lambraño et al. 2015). The repellent activity of these essential oils could be attributed to the presence of components such as geranial, neral, limonene, linalool, eugenol, citronellol and citronellal, which have been reported as a repellent (Nerio et al. 2010; Olivero-Verbel et al. 2010; Baldacchino et al. 2013). Similarly, to Hernandez-Lambraño et al. (2015) our study showed that *C. nardus* essential oil was the most active repellent against storage insect pests than *C. citratus*. However, *C. citratus* at the lower concentration showed the higher repellent activity against *D. porcellus* (class IV). This could be explained by the interaction and/or inactivation of some *C. citratus* essential oil components at high concentration. Further studies should be done to evaluate the bioactivity of each citral components including in *C. citratus* essential oil and their interactions against *D. porcellus*. Knowing that repellency play a role directly in the reduction of egg-laying and hence the emergence of adults (França et al. 2012), essential oils of the both Cymbopogon species can be used as repellents against *D. porcellus* for stored yam chips protection.

Our study revealed the low activity of *C. citratus* and *C. nardus* essential oils as contact insecticide against *D. porcellus*. This result is in line with those of Stefanazzi et al. (2011) which reported no contact toxicity of *C. citratus* essential oil on *T. castaneum* larvae and adults. Similarly, *C. nardus* essential oil showed a low contact toxicity against *Musca domestica* (Samarasekera et al. 2006). However, some studies revealed the high contact toxicity of *C. citratus* and/or *C. nardus* essential oils against several storage insect pests such as *S. oryzae* (Franz et al. 2011), *S. zeamais* (Kabera et al. 2011), *Palorus subdepressus* Wollaston, *Rhyzopertha dominica* Fabricius, and *Cryptolestes* sp (Doumbia et al. 2014). This low sensitive of *D. porcellus* to the both Cymbopogon species essential oils could be due to the cuticle of this insect, which acts as a barrier. This difference in action could also be related to the variability of chemical constituents of the *C. citratus* essential oils used. Our results showed that the contact toxicity of *C. citratus* and *C. nardus* essential oils increase with concentration. This insecticidal activity of *C. nardus* essential oil was probably due to the compounds such as citronellal and geranyl acetate while that of *C. citratus* essential oil could be due mainly to the presence of citral (Samarasekera et al. 2006).

Our study showed that both Cymbopogon species essential oils exhibited strong antifeedant activity against *D. porcellus*. This antifeedant activity could be attributed to the presence of monoterpenes such as citral or citronellal in these essential oils, which can act by stimulating a deterrent receptor of gustatory sensillum in an insect causing an unpalatable taste or by penetrating the body through the digestive system (Arasu et al. 2013; Arivoli and Tennyson 2013). Our results showed that *C. citratus* have more antifeedant activity and was more toxic than *C. nardus* at the higher concentration. In fact, some studies indicated the antifeedant activity of *C. citratus* essential oil against *T. castaneum* larvae (Stefanazzi et al. 2011). Moreover, *C. citratus* essential oil presented insecticidal activity close to the synthetic insecticide Actellic 50 EC.

Therefore, *C. citratus* essential oil could be a source of natural antifeedant, which can be used for yam chips protection. Knowing that antifeedants never kill the target insects directly and allow them to be available to their natural enemies (Arivoli and Tennyson, 2013), the combination of *C. citratus* essential oil with the predator *Alloeocranum biannulipes* Montrouzier and Signoret (Loko et al. 2019a, b) for integrated pest management of *D. porcellus* must be evaluated.

A high fumigant toxicity of *C. citratus* and *C. nardus* essential oils on *D. porcellus*, comparatively to the synthetic insecticide Actellic 50 EC was observed. However, *C. citratus* essential oil was significantly more toxic for *D. porcellus* than those of *C. nardus*. This is not surprising because several studies have shown the strong fumigation effects of *C. citratus* essential oil comparatively to *C. nardus* essential oil against *T. castaneum* (Bossou et al. 2015), *S. oryzae* (Paranagama et al. 2004), *O. surinamensis* and *S. zeamais* (Hernandez-Lambraño et al. 2015). Similarly, Stefanazzi et al. (2011) observed a high fumigant toxicity of *C. citratus* against *T. castaneum*. The low fumigant toxicity of *C. nardus* essential oil could be due to the no fumigant insecticidal activity of citronellol (Bossou et al. 2015). Before supplementary trials, which will be conducted under farmers storage conditions, we can recommended the use of *C. citratus* essential oil in the framework of an integrated pest management program against *D. porcellus*.

Conclusions

Our results showed the high yield and a difference in the chemical composition of the essential oils from *C. citratus* and *C. nardus* leaves. The two essential oils showed different biological activities towards *D. porcellus*. The results revealed the good potential of *C. citratus* as both antifeedant and fumigant toxic agent against *D. porcellus*. Whereas, *C. nardus* essential oil could be recommended as repellent. Both essential oils constitute viable alternatives for control of *D. porcellus*. However, further studies are required to evaluate the insecticidal activity of both Cymbopogon essential oils under real yam chips storage conditions and to develop a good formulation to improve their efficacy, and stability as biopesticides.

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Authors' contributions LYLE and MFS contributed to the study conception and design. KP, ACA and TJ performed the experiments. LYLE analysed the data and drafted the manuscript. GB, DV, KO, DSL, SMF, TM and GF corrected the manuscript. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest No potential conflict of interest was reported by the authors.

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