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Insecticidal activity of crude extracts of three spices and commercial botanical pesticide on oriental fruit fly under laboratory conditions

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Abstract

Background: The uses of botanical pesticides in pest management are currently on remarkable increase due to their efficacy, biodegradability, environment-friendly, and availability. Ethanol extracts of three spices (*Piper guineense*, *Aframomum melegueta*, *Zingiber officinale*) and commercial botanical pesticide AzaSol (6% azadirachtin) were assessed for contact toxicity, residual effects, and for their potential in soil application against pupariating larvae of oriental fruit fly (*Bactrocera dorsalis*) in the laboratory at ambient temperature of 27 ± 2 °C and relative humidity of 75–80%. The extracts and AzaSol were applied at 1:1.5 w/v concentration while cypermethrin was introduced as standard check and applied at 5 ml/liter of water.

Results: All the treatments were very effective against *B. dorsalis* in contact toxicity and residual effects recording 89.4–100% larval mortality at 24 h post-application. *Z. officinale* and cypermethrin had similar contact and residual effects on *B. dorsalis*, both recording 100% larval mortality at 24 h post-exposure. *Piper guineense* showed higher residual effects than contact effects, while *A. melegueta* and AzaSol showed better contact effects than residual effects against *B. dorsalis* larvae. AzaSol was the most effective among the botanicals in reducing the adult emergence and in enhancing larval mortality (96.7%) on treated soil followed by *Piper guineense* (83.3%). The efficacy of AzaSol on the treated soil was comparable to cypermethrin. All the extracts were significantly more effective than control in enhancing pupariating larvae mortality and in reducing adult emergence on treated soil.

Conclusion: Ethanol extracts of *P. guineense* and *A. melegueta* were highly promising against *B. dorsalis* on treated soil and could be adopted in soil application targeting puparia under the tree canopies as part of integrated pest management of *B. dorsalis* in orchards.

Keywords: *Bactrocera dorsalis*, Pupariating larvae, Soil treatment, Plant extracts, Mortality

Background

Globally, Tephritid fruit flies (Diptera: Tephritidae) are among the most important insect pests of fruits and vegetable causing significant economic losses to farmers (Badii, Billah, Afreh-Nuamah, Obeng-Ofori, & Nyarko, 2015). The oriental fruit fly, *Bactrocera dorsalis* (Hendel

(Diptera; Tephritidae) is one of the most important fruit fly species devastating fruit productions in sub-Saharan Africa region. Fruit production and exports in Africa is one of the agricultural sector that plays a vital role in the growth of local economy of the region, providing sources of livelihood for farmers, foreign exchange revenues, and job opportunities to many populace in the continent (Mengesha, Abate, Adamu, Zewde, & Addis, 2019). Several fruits like mango, pawpaw, avocado, and citrus are grown in Africa for exports to foreign

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countries like the USA, Europe, and Middle East. The menace of fruit fly infestations in the region has hampered the production of exportable fruits in the region for the past decades since the arrival of *B. dorsalis* in the region. *Bactrocera dorsalis* is highly polyphagous and can infest and induce a substantial economic losses to a wide variety of fruit crops like mango, guava, citrus, mandarin, peach, and pawpaw (Ekesi, Maniania, Mohamed, & Lux, 2005; Jin, Zeng, Lin, Lu, & Liang, 2011; Stephens, Kriticos, & Leriche, 2007). *Bactrocera dorsalis* causes losses to fruits through direct damage, fruit drops, and export restrictions due to quarantine regulations. Different management strategies for fruit fly species devastating crops has been developed in various parts of the world (Dias, Zotti, Montoya, Carvalho, & Nava, 2018; Mau, Jang, & Vargas, 2007; Vargas et al., 2001; Vargas, Mau, Jang, Faust, & Wong, 2008). Some of these major control strategies include farm sanitation, foliage, and soil application of insecticides, bait spray, male annihilation techniques, biological control, release of sterilized flies, and parasitoids (Clark, Steck, & Weems, 1996; Vargas et al., 2001, 2008; Vargas, Leblanc, Piñero, & Hoffman, 2014). Application of insecticides on the soil under the host trees to kill fruit fly larvae and puparia is an important part of a fruit fly suppression and eradication program (California Department of Food and Agriculture, (CDFA), 1993; Roessler, 1989; Saul, Tsuda, & Wong, 1983). However, the use of synthetic insecticides is detrimental due to its adverse effects on the environment and non-target organisms as well as toxic residues in fruits and associated restrictions in international trade on minimum residue levels (MRL) (El-Aw, Draz, Hashem, & El-Gendy, 2008). Screening plant botanicals for direct application to the soil, foliage spray, or incorporation into baits may disclose potential safer insecticide alternatives for fruit fly control. Plant extracts has been used widely and effectively in the management of several insect pests (Isman, 2006; Lengai, Muthomi, & Mbega, 2020). Fruit fly species like *B. zonata* has been reported to show repellence effects and growth inhibition by the plant extracts from Kalmus (*Acorus calamus*), neem (*Azadirachta indica*) wild rue (*Peganum harmala*), and three other plant species (Akhtar, Jilani, Mahmood, Ashfaq, & Iqbal, 2004). Acetone extracts of turmeric has been reported to show high repellent effects and growth inhibition of *B. zonata* (Siddiqi, Jilani, Rehman, & Kanvil, 2006).

Extracts from *A. indica* has been reported to show anti-feeding effects, repellence, toxicity, and anti-oviposition effects on *B. dorsalis* and melon fly, *B. cucurbitae* (Shivendra & Singh, 1998).

Aqueous extracts of *A. indica*, *Piper guineense*, *Moringa oleifera*, and *Aframomum melegueta* were found effective against oriental fruit fly larvae under laboratory condition (Ugwu & Nwaokolo, 2020). This study

therefore evaluated the efficacy of ethanol extracts of three spices (*Piper guineense* Schum and Thonn, *Aframomum melegueta* (Roscoe) K. Schum *Zingiber officinale* Roscoe, and Azasol (6% azadirachtin)) against third instar larvae and pupariating larvae of *B. dorsalis* and their potential in reducing adult emergence on treated soil.

Methods

The study was conducted in the Biology Laboratory of the Federal College of Forestry Ibadan, Forestry Research Institute of Nigeria under ambient temperature of 27 ± 2 °C and relative humidity of 70 to 80% and L12: D12 photoperiod. The extracts of spices evaluated include *P. guineense*, *A. melegueta*, *Zingiber officinale*, and Azasol (6% azadirachtin), a commercial botanical from neem plant. Dried *A. melegueta*, *P. guineense* seeds, and fresh *Z. officinale* were purchased from a local market in Ibadan while Azasol (6% azadirachtin) was procured from Arboget Company in Florida, USA

Processing and extraction of plant materials

Fresh *Z. officinale* roots were sliced into small chips and air dried for 2 weeks under ambient light conditions at fluctuating temperature of 27 ± 2 °C, 75–80% relative humidity and L12: D12 photoperiod. All the dried plant materials (*Z. officinale* roots, *P. guineense* seeds, and *A. vhmelegueta*) were ground into powder using an electric blender (Binatone blender/grinder BLG.450). The powdered plant materials were weighed out in 100 g each and extracted by Soxhlet method using 250 ml of ethanol for minimum of 8h according to Ofuya et al. (1992).

Insect culture

Populations of *B. dorsalis* were reared in the laboratory at ambient temperature of 25 ± 2 °C and relative humidity of 70 to 80% on *Irvingia gabonensis* fruits. The infested fruits of *Irvingia gabonensis* were collected from the *Irvingia* plantation at Forestry Research Institute of Nigeria and were kept in a plastic cages in the laboratory and monitored for adult emergence. The emerged adults were paired in another cage for mating and oviposition. The rearing cages were supplied with fresh *Irvingia* fruits every 2 days after the first instar larvae emergence and the culture was maintained until the end of the experiment according to Ugwu (2020)

Toxicity and residual bioassay

Ethanol extracts of the three spices were evaluated for residual action by applying 1 ml of each extracts on petri dishes lined with Whatman filter paper (90 mm). Petri dishes were left for 10 min to drain off before five third instar larvae of *B. dorsalis* were separately introduced into each petri dish. The contact toxicity of the extracts

was assessed by applying 0.1 ml of each extracts using 200 µl micropipettes on the dorsal cavity of the insect. Ethanol extracts was applied at 1:1.5 w/v (75%) concentration while liquid formulation of 6% azadirachtin (AzaSol) was prepared at 10 g/100 ml of water and applied at 1:1.5 w/v (75%) concentration. Cypermethrin was introduced as a standard check and was applied at 5 ml/liter of water, while distilled water served as control for the entire test. All assays were replicated three times in a completely randomized design (CRD).

Soil treatment with extracts and AzaSol

Soil samples were collected from the experimental farm of Federal College Forestry, Ibadan, and 100 g were weighed into each plastic cage of 14 cm × 9 cm × 7 cm. The soil was then treated with ethanol extracts and 6% azadirachtin (AzaSol) at 20 ml/100 g of soil. Cypermethrin was applied at 5 ml/l of water into 100 g of soil. Ten pupating *B. dorsalis* larvae were introduced into each cage on treated soil, covered with mosquito net, and observed for adult emergence. Each treatment was replicated three times in a CRD.

Data collection and analysis

Data on the mortality of *B. dorsalis* larvae were recorded at 20 min intervals for 24 h for both contact and residual bioassays, while adult emergence were recorded on the treated soil after 3 weeks (21 days). The pupating larvae mortality was inferred by calculating from the number of adult emergence after 21 days. Data collected were subjected to square root transformation before analysis of variance (ANOVA). Significant means were separated by Tukey's honestly significant difference (HSD) at 0.05 level of significance with ASSISTAT statistical software 7.6 beta.

Results

Contact and residual effects of the botanicals on *B. dorsalis* larvae

Plant botanicals evaluated showed contact toxicity on the third instar larvae of *B. dorsalis* under laboratory conditions. Larval mortality was observed from 60 min to 24 h post-treatments on all the botanicals (Table 1). *Piper guineense* and *Z. officinale* had similar larval mortalities with mean value of 0.67 at 60 min of post-exposure. *Aframomum melegueta* recorded highest larval mortality with mean value of 2.33 at 100 min of post-exposure, followed by 6% azadirachtin (1.33). The mortality action of *Z. officinale* was highest with mean value of 2.33 at 1440 min of post-exposure, and it was not significantly different ($p > 0.05$) from 6% azadirachtin (1.67). Synthetic insecticides (cypermethrin) were faster than all the treatments in killing all larvae at 100 min of post-exposure. All the botanicals significantly ($p < 0.05$)

Table 1 Effect of contact toxicity of botanicals and synthetic chemical on *B. dorsalis* larvae

Treatments	Time after application (min)						
	20	40	60	80	100	120	1440
<i>P. guineense</i>	0.00	0.00b	0.67a	1.33a	0.67b	1.33a	0.67b
<i>A. melegueta</i>	0.00	0.00b	0.00b	0.67b	2.33a	1.33a	0.67b
<i>Z. officinale</i>	0.00	0.00b	0.67a	0.67b	1.00ab	0.33b	2.33a
6% azadirachtin	0.00	0.00b	0.33b	1.00a	1.33a	0.67a	1.67a
Cypermethrin	0.00	2.67a	0.67a	0.33b	1.33a	0.00b	0.00b
Control	0.00	0.00b	0.00b	0.00b	0.00b	0.00b	0.00b

Means with the same letter within the same column do not differ statistically by Tukey's honestly significant difference (HSD)

caused larval mortality compared with the control where none of the larvae died.

The residual effects of the botanical followed a similar pattern with their contact toxicity. All the plant botanicals recorded larval mortality from 60 min post-exposure (Table 2). *Aframomum melegueta* recorded highest larval mortality among the extracts with mean value of 0.67 at 60 min post-exposure. *Piper guineense* and 6% azadirachtin had equal larval mortality at 80 min post-exposure with mean value of 1.69. The residual effects on the larvae continued until 24 h post-exposure. Cypermethrin commenced residual action on larvae from 40 min and gave 100% mortality at 120 min of post-exposure, while no mortality was recorded in the control throughout the period of observation.

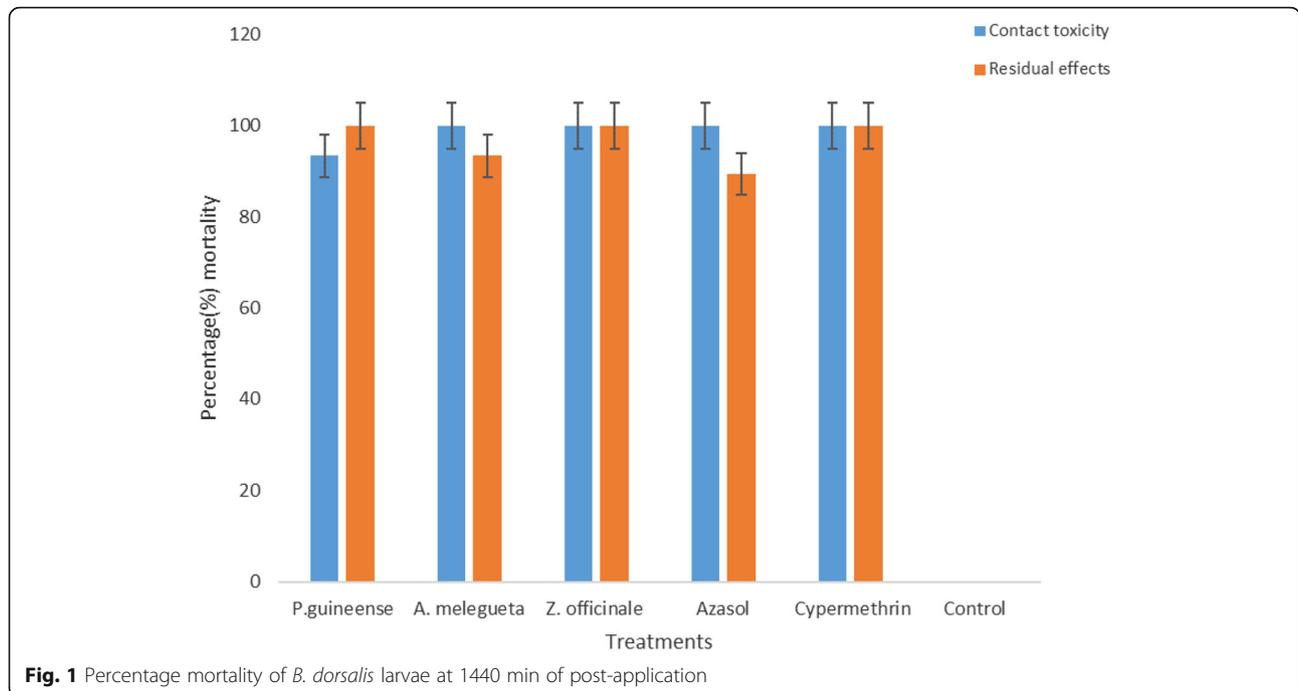
Comparison of contact toxicity and residual effects of treatments on mean percentage mortality of *Bactrocera dorsalis* larvae at 1440 min post-exposure

Contact toxicity and residual effect of treatments were compared in Fig. 1. All the treatments proved very effective against *B. dorsalis* for both contact toxicity and residual effects at 24 h post-application (Fig. 1). *Zingiber officinale* had similar contact and residual effects with cypermethrin; both caused 100% larval mortality at 24 h post-exposure. *Piper guineense* slightly had higher

Table 2 The residual effects of the botanicals and synthetic chemical on *B. dorsalis* larvae

Treatments	Time after application (min)						
	20	40	60	80	100	120	1440
<i>P. guineense</i>	0.00	0.00b	0.33b	1.67a	1.33b	0.67b	1.00b
<i>A. melegueta</i>	0.00	0.00b	0.67a	0.67b	0.67b	1.33a	1.33b
<i>Z. officinale</i>	0.00	0.00b	0.33b	0.00c	1.67a	0.67	2.33a
6% azadirachtin	0.00	0.00b	0.33b	1.67a	0.67b	1.67a	1.00b
Cypermethrin	0.00	0.67a	0.00b	1.00b	2.33a	1.00ab	0.00c
Control	0.00	0.00b	0.00b	0.00c	0.00c	0.00c	0.00c

Means with the same letter within the same column do not differ statistically ($p < 0.05$) by Tukey's honestly significant difference (HSD)



residual effects on *B. dorsalis* larvae than contact effects with 100% and 93.4% for residual and contact toxicity, respectively. *Aframomum melegueta* showed higher contact toxicity than residual effects recording 100 and 93.4% mortality, respectively. AzaSol (6% azadirachtin) had better contact effects than residual effects with 100 and 89.4%, respectively compared with *P. guineense*.

Effect of soil application of the botanicals on mortality and adult emergence of *B. dorsalis*

The results showed that all the extracts were effective in reducing emergence of *B. dorsalis* adults on treated soil (Table 3). AzaSol (6% azadirachtin) was the most effective among the botanicals in reducing the adult emergence and in enhancing larval mortality on treated soil. It caused 96.7% mortality of pupariating larvae followed by *Piper guineense* (83.3%). The effect of AzaSol on *B. dorsalis* on treated soil was comparable with

cypermethrin (100.0%). *Aframomum melegueta* caused above 70% larval mortality on treated soil. *Zingiber officinale* was least among the botanicals in causing pupariating larvae mortality (56.7%). All the extracts were significantly more effective than control in causing pupariating larval mortality of *B. dorsalis*. Adult emergence was higher on untreated soil (control) with about 80%, followed by soil treated with *Z. officinale* (43.3%), *A. melegueta* (26.7%), and *P. guineense* (16.7%) while soil treated synthetic insecticide (cypermethrin) recorded no emergence of adult *B. dorsalis*. The higher the mortality, the lower the emergence.

Discussion

All the botanicals evaluated recorded high level of *Bactrocera dorsalis* larval mortality at both contact and residual effect at 24 h of exposure. The study has demonstrated the potential of the tested plant extracts to decimate populations of *B. dorsalis* larvae. This results support several reports from previous studies on the potential of plant botanicals in the management of fruit fly infestations. Akhtar et al. (2004) reported that extracts from *Acorus calamus* Linn, *Azadirachta indica* A. Juss, *Curcuma longa* L, *Peganum harmala* L, *Saussurea lappa* (Decne)Sch.Bip., and *Valeriana jatamansi* Jones inhibited the growth of *Bactrocera zonata* in Pakistan. Similarly, Jaleel et al. (2020) reported that *Piper nigrum* has been very effective in repelling *B. correcta* and *B. dorsalis*. Furthermore, aqueous extracts of *A. indica*, *P. guineense*, *M. oleifera*, and *A. melegueta* caused contact and residual toxicity against *B. dorsalis* at

Table 3 Effect of soil application of botanicals on mortality and adult emergence of *B. dorsalis*

Treatment	Mortality (%)	Adult emergence (%)
<i>P. guineense</i>	83.3 ab	16.7 cd
<i>A. melegueta</i>	73.3 bc	26.7 bc
<i>Z. officinale</i>	56.7 c	43.3 b
6% azadirachtin (Azasol)	96.7 a	3.3 3d
Cypermethrin	100.0 a	0.0 d
Control	20.0 d	80.0 a

Mean values with the same letter within the column do not differ statistically ($p < 0.05$) by Tukey’s honestly significant difference (HSD)

varied rates under laboratory conditions (Ugwu & Nwaokolo, 2020). The insecticidal efficiency of *P. guineense* is attributed to Piperine the active ingredient in *P. guineense* (Scott et al., 2004). *A. melegueta* has been reported to possess several biological activities like anti-inflammatory, antioxidant and anti-tumor effects, and anti-microbial activity (Chung, Jung, Surh, Lee, & Park, 2001; Oloke, Kolawole, & Erhun, 1998; Tjendraputra, Tran, Liu-Brennan, Roufogalis, & Duke, 2001). Similarly, Ukeh, Birkett, Pickett, Bowman, and Luntz (2009) reported that *A. melegueta* and *Z. officinale* extracts exhibited repellent effect towards adult *S. zeamais* both in the absence and the presence of maize, *Zea mays*, grains. The biological activities of *A. melegueta* is attributed to three major compounds; (*S*)-2-heptanol, (*S*)-2-heptyl acetate, and (*R*)-linalool (Ukeh et al., 2009). Siddiqi et al. (2006) also reported that petroleum ether, acetone, and ethanol extracts of turmeric plant were effective in repelling and inhibiting growth of *B. zonata*. Several past studies have been documented on the potential of botanicals in the control of many insect pests of different crops both in the field and storage. Powders of *Z. officinale*, *A. indica*, *A. melegueta*, and other local plant materials have been found promising in protecting *Cajanus cajan* and *Vigna unguiculata* grains against *Callosobruchus maculatus* infestation in storage (Ekeh, Onah, Atama, Ivoke, & Eyo, 2013). Similarly, Idoko and Adesina (2012) reported that *P. guineense* powder has contact effects on *C. maculatus* which resulted in adult mortality and inhibition of female oviposition. The residual and contact effects of *Z. officinale* on *B. dorsalis* were comparable to cypermethrin. These results corroborate the findings by Addo (2017) who reported that aqueous extracts of *Z. officinale* were effective against major pests of cabbage (*Brassica oleracea* var. *capitata*). Giriraju and Yunus (2013) reported that 10% ethanolic extracts of *Z. officinale* possess antimicrobial potential against pathogens. The antimicrobial and other several biological activities *Z. officinale* are attributed to gingerol and paradol, shogaols, and zingerone (Rahmani, Al Shabrmi, & Aly, 2014). Several studies have shown that plant botanicals were more effective than synthetic insecticides against various insect pests of crops both in the field and storage. Basedow, Obiewatsch, Bernal Vega, Kollmann, and Nicol (2002) reported that *A. indica*-based products were more efficacious than synthetic insecticides against aphids and white flies in the field. Similarly, Ugwu, Ojo, Aderolu, and Aderemi (2014) reported that *A. indica* seed extracts showed higher efficacy than Cypermethrin against leaf roller *Sylepta derogata* on okra in the field. In this study, Azasol (6% azadirachtin) was very effective in reducing adult emergence of *B. dorsalis* on treated soil. The results supports the findings by Khan, Hossain, and Islam (2007) who

reported that high doses of *Azadirachtin*, an active ingredient of *A. indica*, were found very effective in reducing the oviposition rates of oriental fruit flies on the melon plants. The efficacy of neem-based product is attributed to their molecular component which comprises complex mixture of molecules with hydrocarbons, phenolic compounds, terpenoids, alkaloids, and glycosides (Hossain, Al-Toubi, Weli, Al-Riyami, and Al-Sabahi (2013). According to Mordue-Luntz and Nisbet (2000), neem molecules act on the life cycle of insects in various phases making it difficult for pests to resist the physiological effects of neem-based extracts. Solangi, Sultana, Wagan, and Ahmed (2011) reported that extracts of neem oil, neem seed powder solution, eucalyptus leaf, and tobacco leaf showed repellent effects against *Bactrocera zonata*. Similarly, Rehman, Jilani, Khan, Masih, and Kanvil (2009) reported that *A. indica* was very effective in reducing the oviposition rate of *B. zonata*, *B. dorsalis*, and *B. olae*. According to Isman (2000) and Cetin, Erler, and Yanikoglu (2004), plant extracts and their oils possess ovicidal, larvicidal, and repellent properties against various insect species. The most vital constituent of several plant extracts or their essential oils are monoterpenoids which possess fumigant properties making them a good element in pest management program (Ahn, Lee, Lee, & Kim, 1998; Al Qahtani, Al-Dhafar, & Rady, 2012; Regnault-Roger & Hamrouni, 1995)

The use of botanicals in pest management has shown great promise as alternatives to chemical insecticides to reduce pesticide load in the environment (Radha & Susheela, 2014).

Conclusion

The current study has proved that ethanol extracts of *Zingiber officinale*, *Piper guineense* *Aframomum melegueta*, and *Azasol* have strong insecticidal activity. *AzaSol* was most effective in reducing adult emergence of *B. dorsalis* on treated soil. Plant botanicals have proven to be environment-friendly and effective in pest management in several forms of application either as foliar spray or soil application. The commercial botanical (*AzaSol*) has proved to be more effective than ethanol extracts of the tested spices in soil application. Thus, further efforts should be made to develop these plant extracts in ready-made formulations to enhance their efficacy for several application methods.

Abbreviations

FCF: Federal College of Forestry; FRIN: Forestry Research Institute of Nigeria; GMT: Greenwich Meridian Time

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Authors' contributions

JAU conceptualized, designed, implemented the research, and drafted the manuscript. OJA analyzed the results of the research while OYA edited the manuscript draft. All authors read and approved the final manuscript.

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Availability of data and materials

All data generated or analyzed during this study are included in this published article.

Declarations**Ethics approval and consent to participate**

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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