Technoscience Journal for Community Development in Africa 2:1 (2021) 101–108
ISSN 2659-0573

Research Article

Malathion and Pirimiphosmethyl susceptibility of bendiocarb resistant *Anopheles gambiae s.l.* mosquito populations in urban Lagos, Nigeria

Adedayo O. Oduola[1], Emmanuel T. Idowu[2], Abiodun Obembe[*,3], Olubunmi A. Otubanjo[2,] Samson T. Awolola[4]

[1] Department of Zoology, University of Ilorin, Ilorin, Nigeria
[2] Department of Zoology, University of Lagos, Lagos, Nigeria
[3] Department of Biosciences and Biotechnology, Kwara State University, Malete, Nigeria
[4] Molecular Entomology and Vector Control Research Laboratory, Public Health Division, Nigerian Institute of Medical Research Lagos, Nigeria

Abstract: This study assessed susceptibility status of Anopheles mosquito populations to organophosphate insecticides in selected areas within Lagos metropolis. The study also provides an update on the earlier established *Anopheles* mosquito resistance to some insecticides such as pyrethroids and carbamate. Adult *Anopheles* mosquito populations reared from larval collections at natural breeding sites in Yaba and Lekki areas in Lagos were exposed to World Health Organization (WHO) insecticide test papers. The insecticides used for the test include: two organophosphates (Malathion and Pirimiphos methyl), two pyrethroids (deltamethrin and permethrin) and one carbamate (bendiocarb). All the mosquitoes used in this study were identified as *An. gambiae sensu lato*. Results from the study showed that *Anopheles* populations from the two sites were fully susceptible (100% mortality rates) to the organophosphates (Malathion and Pirimiphos methyl). The *Anopheles* populations exhibited resistance to pyrethroid (permethrin and deltamethrin) (≤ 60% mortality rates) and carbamate bendiocarb (≤ 38% mortality rates). The resistance levels were higher compared to the reports of earlier studies.

Keywords: Malaria; *Anopheles*; insecticide resistance; organophosphate

1. Introduction

Malaria is caused by parasites of the genus *Plasmodium* and mosquitoes of the genus *Anopheles* serve as vectors for the parasites. Globally, about 229 million malaria cases were reported by the World Health Organization in 2019 (WHO, 2020). This represents a million reported cases higher than in 2018 (WHO, 2019). Africa accounted for about 94% of the reported malaria cases in 2019, and 51% of the cases were reported in five countries - Nigeria (27%), Democratic Republic of the Congo (12%), Uganda (5%), Mozambique (4%) and Niger Republic (3%) (WHO, 2020). In addition to morbidity and mortality caused by this disease, huge economic cost of funding malaria control interventions are financial burdens on the Governments of malaria endemic countries. In Nigeria where the highest global malaria cases have been recorded, malaria accounts for about 60% of outpatient visits, 30% of hospitalizations, 10% of low birth weight and 11% of maternal mortality (NMEP, 2016). Preliminary analysis by the National Malaria Elimination Programme of Nigeria showed that US$ 2.75 billion is needed to achieve high coverage of malaria control interventions in targeted areas, and full availability of diagnosis and treatment in public health facilities (WHO, 2020).

Vector control using insecticides is a popular malaria control intervention. Currently, there are two main strategies employed for malaria vector control: use of Long-Lasting Insecticidal Nets (LLIN) and Indoor Residual Spray (IRS). Recommended classes

* Corresponding author:
Email: abiodunobembe@yahoo.com

KWASU PRESS AND PUBLISHING

of insecticides for LLIN and IRS include: pyrethroids, organochlorine, carbamates and organophosphates (WHO, 2006). Although each class of insecticide may be further categorized into different sub-classes which in turn consist of a broad range of compounds, specific ones endorsed for IRS malaria vector control based on human safety and their residual efficacy on a dwelling surface include: DDT for organochlorine; bendiocarb and propoxur for carbamate; malathion, fenitrothion and pirimiphos-methyl for organophosphates; and alphacypermethrin, deltamethrin, lambda-cyhalothrin, etofenprox, bifenthrin and cyfluthrin for pyrethroids (WHO, 2015). Pyrethroid insecticides recommended for treating mosquito bed nets include: permethrin, alphacypermethrin, deltamethrin, lambda-cyhalothrin, etofenprox and cyfluthrin (WHO, 2016). Among the insecticides recommended for bed-nets and IRS based malaria vector control, the most commonly used and tested for resistance are: deltamethrin, permethrin and alphacypermethrin (pyrethroid), bendiocarb (carbamate), DDT (organochlorine) and pirimiphos methyl (organophosphate). Insecticide-treated mosquito nets and indoor residual spraying of these recommended insecticides have contributed significantly to reductions in malaria morbidity and mortality since 2000 (Wahedi *et al*., 2020). Long-lasting insecticide-treated nets (ITNs) are more commonly used for malaria vector control because of the ease in delivering the tools through mass campaign and other routine distribution channels (Obembe *et al*., 2014). However, resistance to recommended insecticides has emerged over the years as a major threat to malaria vector control intervention programmes in many countries in sub – Saharan Africa.

The problem of insecticide resistance in malaria vectors has expanded rapidly due to the frequent use of similar insecticide families for public health and agricultural uses (Antonio-Nkondjio *et al*., 2017). Studies on insecticide resistance are required to comprehend the epidemiology, prevalence and mechanisms of resistance in order to predict strategies to overcome this problem (Sweileh *et al*., 2016). For instance, in Nigeria, a number of such studies conducted had indicated that *Anopheles gambiae sensu lato* (*s.l.*) mosquitoes exhibited resistance to some of the aforementioned recommended insecticides - pyrethroids (Oduola *et al*., 2012; Awolola *et al*., 2018; Ibrahim *et al*., 2019), organochlorine dichlorodiphenyltrichloroethane (DDT) (Oduola *et al*., 2010; Adeogun *et al*., 2017; Ononomadu *et al*., 2020) and carbamate (Oduola *et al*., 2012; Ibrahim *et al*., 2019; Ononomadu *et al*., 2020). In order to manage insecticide resistance in IRS, rotational spraying of two or more insecticide classes on a yearly basis had been recommended. Moreover, regular resistance monitoring will be crucial to establishment of a global database to track the status of insecticide resistance in malaria vectors (WHO, 2012).

For several years, over reliance on pyrethroids as the only class of insecticide approved for treating bed-nets has led to increased pyrethroids resistance in many localities (Adeogun *et al*., 2017; Ibrahim *et al*., 2019). Attempts at preserving and improving the use and effectiveness of pyrethroids for vector control purposes through LLINs have led to the development of new generation bed nets treated with a combination of pyrethroid and non pyrethroid insecticide. With the emergence of these new bed nets, there is the need to provide information on the potency of the various classes of recommended insecticides against mosquitoes in specific localities in order to prescribe effective insecticide options for both LLIN and IRS malaria vector control in such locations. Previous work on the insecticide susceptibility status of *Anopheles gambiae s.l.* in Lagos metropolis showed the occurrence of multiple resistance to organochlorine (DDT), pyrethroids (permethrin, deltamethrin and alphacypermethrin) and carbamate (bendiocarb and propoxur) insecticides (Oduola *et al*., 2010; Oduola *et al*., 2012) with no data reported on the organophosphate susceptibility profile. This study was conducted to assess the susceptibility status of *Anopheles* mosquito populations in Lagos to two recommended organophosphate insecticides and to provide an update on the earlier established *Anopheles* mosquito resistance to pyrethroids and carbamate insecticides.

2. Materials and methods

2.1. Mosquito collection, rearing and identification

Pre-adult *Anopheles* mosquitoes were collected between May and July, 2017 from natural mosquito breeding sites in Lekki (6°30′N 4°07′E) and Yaba (6.50° N, 3.38° E) in Lagos. The collected larvae and pupae were reared to adulthood (27^{o}C - 28.8^{o}C, 67.3% - 88.1%) at the insectary of Molecular Entomology and Vector Control Unit of the Nigerian Institute of Medical Research, Lagos. Details of mosquito larval collection and processing have been reported in Oduola *et al*. (2012). The emerging F1 progeny of 2-3 days old were subjected to insecticide susceptibility tests as described by WHO (2013). After

insecticide exposure, all mosquitoes were identified morphologically under a stereomiscrope following the standard morphological keys for *Anopheles* mosquito identification described by Gillies and Coetzee (1987).

2.2. Insecticide susceptibility test

Insecticide susceptibility tests were conducted with WHO test papers containing recommended diagnostic concentrations of 0.05% deltamethrin and 0.75% permethrin (pyrethroid), 0.1% bendiocarb (carbamate), 0.1% Pirimiphos methyl (organophosphate) and 5% malathion (organophosphate) using standard procedures and test kits for adult mosquitoes (WHO, 2013). To affirm the potency of the test papers before its use, *Anopheles* mosquitoes of the Kisumu susceptible strain (*Kss*) were exposed to treated papers before the experimental exposures of mosquito samples collected in the field.

For each insecticide, four replicates of 25 non-blood fed (total of 100) female *Anopheles* mosquitoes were exposed to the insecticide test papers in the test tubes for 1 hour (WHO, 2013). The insecticide papers were tested against *Anopheles* mosquitoes collected from the two different collection sites. Control experiment conducted concurrently involved two replicates of 25 mosquitoes exposed to control papers in a WHO test tube. This was set up for each insecticide tested per location. After the 1-hour exposure period, the mosquitoes were transferred into holding tubes, supplied with 10% sugar solution and monitored for mortality 24 hours after exposure (WHO, 2013). Dead (susceptible) and alive (resistant) mosquitoes were preserved separately in eppendorf tubes filled with desiccated silica gel.

2.3. Data analysis

The time taken for 50% (Kdt50) and 95% (Kdt95) of the tested mosquitoes to be knocked down were estimated by the log time probit model analysis. The WHO (2013) criteria of less than 90% mortality as resistant, 90% - 97% mortality as possible resistance and mortality ≥ 98% as susceptible was used to evaluate the resistance/ susceptibility status of the mosquitoes.

3. Results

3.1. Knock down effect of the insecticides

The time taken for 50 and 95% of the mosquitoes to be knocked down (Kdt50 and Kdt95) due to insecticide exposures are detailed in Table 1. The results from the log time probit model analysis show that the organophosphate insecticides (Malathion and Pirimiphos-methyl) required the shortest time to induce 50% (Kdt 50 of 14.75-20.90 minutes) and 95% (Kdt95 of 21.63-29.62) knock down of the mosquitoes from the two sites. Bendiocarb and permethrin insecticides required the longest time to induce 50% (Kdt50 of 55.91-168.21) and 95% (Kdt95 of 101.89-256.13 minutes) knock down of the mosquitoes from the two study sites (Table 1). The mean percentage knock down of mosquitoes after the 1 hour insecticide exposure is presented in Figure 1. Lowest percentage knock down were recorded upon exposure of the mosquitoes from the two sites to bendiocarb (22±2.31% - 24±3.27%) and permethrin (26±9.52% -37±5.03%) insecticides. Only the two organophosphate insecticides (Malathion and Pirimiphos-methyl) induced 100±0.0% knock down of all mosquitoes exposed after 1 hour (Figure 1). Results of morphological identification of all the *Anopheles* mosquito samples exposed to insecticide papers showed that the samples were all *An. gambiae s.l.*

3.2. Mortality rates of exposed mosquitoes

The result of the susceptibility assays indicated that the percentage mortalities of *Anopheles* mosquito exposed to organophosphate insecticides (Malathion and Pirimiphos methyl) were higher than those of Pyrethroid (deltamethrin and permethrin) and carbamate (Bendiocarb) insecticides (Table 2). After 24 hours post exposure period, *Anopheles* populations from both study sites were fully susceptible (100% mortality) to Malathion and Pirimiphos methyl. The lowest percentage mortalities (Lekki 36%; Yaba 38%) were obtained from exposures of mosquito populations to bendiocarb (carbamate) insecticide (Table 1). Similarly, *Anopheles* mosquito populations from both collection sites showed high level of resistance to the pyrethroid (deltamethrin and permethrin) (≤ 60% mortality) insecticides. None of the mosquito populations exposed to the organophosphate (Malathion or Pirimiphos methyl) insecticides was alive (100% mortality) after the 24 hours post exposure period (Table 2). Percentage mortality of mosquitoes from both Lekki and Yaba sites were similar upon exposures to the different insecticides. Highest percentage mortality difference (9%) was recorded upon exposures of mosquitoes from both sites to permethrin (Lekki 54%; Yaba 45%) insecticide (Table 2). Mosquitoes from both sites showed 100% mortality to the two organophosphate insecticides and only 2-3% percentage mortality difference upon exposures to deltamethrin (Lekki 57%; Yaba 60%) and bendiocarb (Lekki 36%; Yaba 38%) insecticides.

Table 1: Knockdown times of mosquitoes exposed to different insecticides

Locality	Insecticide	N	KDT_{50} (95% CI) MINS	KDT_{95} (95% CI) MINS
Yaba	Permethrin	100	62.30 (53.6-88.9)	129.14 (92.7-401.8)
	Deltamethrin	100	39.13 (32.69-46.01)	87.10 (66.12-153.68)
	Bendiocarb	100	168.21(136.1-292.4)	256.13(181.5-432.4)
	Pirimiphos-methyl	100	20.90 (19.49-23.59)	29.62 (25.57-40.17)
	Malathion	100	17.01 (11.11-26.8)	25.90 (19.61-209.75)
Lekki	Permethrin	100	55.91 (53.1-61.9)	101.89 (87.31-139.69)
	Deltamethrin	100	19.88 (18.09-21.36)	31.22 (26.60-43.61)
	Bendiocarb	100	97.11(61.2-423.23)	121.25(86.91-256.45)
	Pirimiphos-methyl	100	19.28 (18.12-21.00)	27.67 (24.47-34.39)
	Malathion	100	14.75 (12.97-15.86)	21.63 (20.05-24.81)

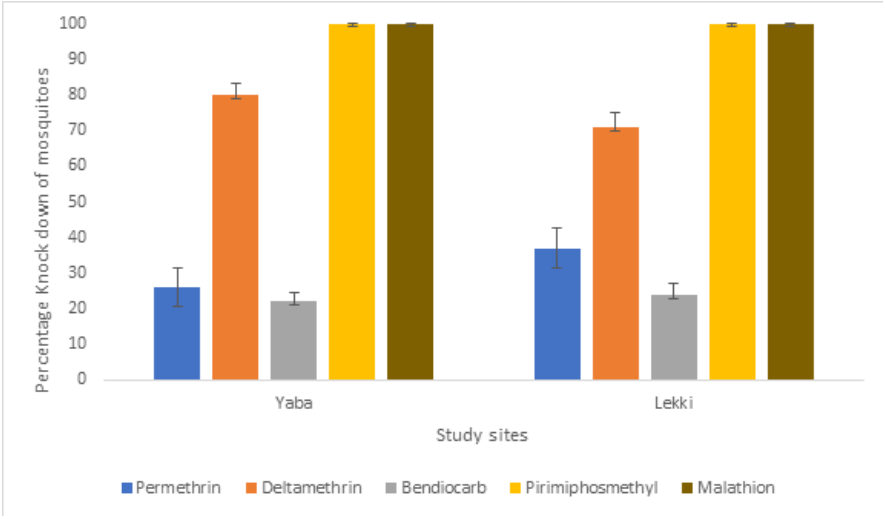

Figure 1: Mean percentage knock down of An. gambiae s.l. mosquitoes from the two study sites after 1 hour insecticide exposure.

4. Discussion

This study assessed susceptibility status of *Anopheles* mosquito populations to organophosphate insecticides in selected areas within Lagos metropolis. Such monitoring of insecticide resistance in the major local malaria vector species should be conducted regularly and in representative sites, using the insecticides already in use and those planned for use in malaria vector control. This will enable assessment of current situation for adaptation of appropriate vector-control strategies (WHO, 2018). Percentage mortality of mosquitoes from both Lekki and Yaba sites considered in the present study were similar upon exposures to the different insecticides. The similarities in mortality rates of mosquitoes from both sites indicates the same status of susceptibility to organophosphates and resistance to carbamate and pyrethroids insecticides. This suggests the prevalence of similar conditions of insecticide resistance selection pressures on the mosquitoes from both sites.

Table 2: Mortality rates and insecticide susceptibility status of Anopheles mosquito populations in urban Lagos.

Class of Insecticide	Insecticide	Community	Number of mosquitoes exposed	Mortality rates (%)	Status
Pyrethroid	Deltamethrin	Lekki	100	57	Resistant
		Yaba	100	60	Resistant
	Permethrin	Lekki	100	54	Resistant
		Yaba	100	45	Resistant
Carbamate	Bendiocarb	Lekki	100	36	Resistant
		Yaba	100	38	Resistant
Organophosphate	Pirimiphos-methyl	Lekki	100	100	Susceptible
		Yaba	100	100	Susceptible
	Malathion	Lekki	100	100	Susceptible
		Yaba	100	100	Susceptible

Susceptibility is based on ≥ 98% mosquito mortality rates (WHO, 2013).

The log time probit model analysis conducted showed high knock down times (KDT_{50} 39.13-62.30 minutes) of mosquitoes from both sites due to pyrethroid deltamethrin and permethrin exposures. Evidence of such high KDT_{50} values exhibited by field populations of *Anopheles* mosquitoes have been attributed to knock down resistance (*kdr*) genes in the mosquitoes (Chandre *et al*., 1999). Further investigations regarding the presence of *kdr* genes in the mosquito populations from both Lekki and Yaba sites are therefore desirable to unravel the mechanisms responsible for the phenotypic insecticide resistance observed in this study. The high KDT_{50} values (39.13-62.30) recorded in our study is comparable with those found in urban *An. gambiae s.l.* populations exposed to the same insecticides in Ibadan; KDT_{50} 34.0-49.5 (Ibrahim *et al*., 2013), Nigeria.

The lower knockdown and mortality rates obtained in this study indicated higher resistance of *An. gambiae s.l.* to carbamate (bendiocarb) and pyrethroid (deltamethrin and permethrin) insecticides than the results earlier reported in Lagos (Awolola *et al*., 2002; Oduola *et al*., 2010; Oduola *et al*., 2012). In contrast, this study also showed that the *Anopheles gambiae s.l.* populations in the selected areas of Lagos were fully susceptible to organophosphates (Malathion and Pirimiphos methyl). Cross-resistance between classes of insecticides had been associated with the sharing of target sites (Labbe *et al*., 2011). Both carbamate and organophosphate insecticides act on the synapse by inhibiting the action of acetylcholinesterase enzyme (Labbe *et al*., 2011). Specifically, in the *An. gambiae* mosquitoes, the acetylcholine-1^R (ace-1^R) gene had been found to cause cross-resistance between organophosphate and carbamate insecticides (Alout *et al*., 2008). Therefore, the results of carbamate resistance and 100% organophosphate insecticide susceptibility observed in this study could raise questions since the general mode of action of carbamate and organophosphate insecticide classes are similar. One valid expectation from the *An. gambiae s.l* populations tested in this study would have been an evidence of cross-resistance between carbamate and organophosphate insecticides. Nevertheless, a possible reason for the differential *An. gambiae s.l* susceptibility to carbamate and organophosphate observed in this study could be due to the type of enzymes elevated in the mosquitoes. According to WHO (2012), though ace-1^R gene has the same high likelihood in modulating for resistance to both carbamates and organophosphates, presence of elevated monooxygenases are more important in conferring resistance to carbamates than esterases while the presence of elevated esterases are more important in conferring resistance to

organophosphates than monooxygenases. Therefore, the possibility of the presence of elevated monooxygenases rather than elevated esterases could be a possible reason responsible for the carbamate resistance and organophosphate susceptibility observed in the *An. gambiae s.l* mosquito populations tested in this study. Further biochemical studies on the enzymes elevated in the resistant *Anopheles* mosquito populations from these study sites are required to confirm this possibility. Similar results of high level pyrethroid resistance, carbamate resistance and 100% susceptibility of *Anopheles gambiae* to organophosphate observed in this study have been reported in Mozambique (Cuamba *et al*., 2010), Benin republic (Aïkpon *et al*., 2013) and Burkina Faso (Namountougou *et al*., 2019).

The observation that all the samples in these collection sites were *An. gambiae s.l* further validates the earlier reported predominance of this species within the Lagos metropolis (Oduola *et al*., 2012). The already established pyrethroid resistance status of these predominant *An. gambiae s.l*. increased from ≤ 79% mortality rates (Awolola *et al*., 2002) to ≤ 73% (Oduola *et al*., 2010) and was extended further in this study to ≤ 60%. Similar sharp reductions in mortality rates of the samples exposed to bendiocarb carbamate insecticides were also observed (36-38%) compared to the previously reported ≤ 70% (Oduola *et al*., 2012). Increased resistance status of *Anopheles* mosquitoes to carbamate insecticide over time have also been reported elsewhere (Cuamba *et al*., 2010; Aïkpon *et al*., 2013).

Despite the high percentage mortality observed in the organophosphate exposure in this study, the "bad egg" smell, terseness and higher mammalian toxicity of the insecticide compared to pyrethroids still remains a challenge. However, widespread pyrethroid resistance (Toe *et al*., 2014; Awolola *et al*., 2018; Namountougou *et al*., 2019) and more recently, the increasing insecticide resistance to the carbamate alternatives (Oduola *et al*., 2012; Aïkpon *et al*., 2013; Namountougou *et al*., 2019; Ononamadu *et al*., 2020) across sub-Saharan Africa and in this study cannot be ignored. Furthermore, organophosphates like chlorpyrifos methyl have limited environmental persistence (N'Guessan et al., 2010) and the efficacy of such ordinarily more expensive insecticide has been improved through the development of microencapsulated forms (Tomlin, 2000). There is therefore the need to further encourage the use of organophosphate insecticide formulations for the control of pyrethroid and carbamate resistant *Anopheles* mosquito population in Lagos as further studies proceed on the reasons for organophosphate susceptibility despite carbamate resistance observed in the mosquitoes.

5. Conclusion

This study re-affirms evidence of increased *An. gambiae s.l.* mosquito populations' resistance to pyrethroid and carbamate insecticide in urban Lagos. The results also showed that organophosphate insecticide formulations are more effective for the control of mosquito populations in Lagos than pyrethroid and carbamate based insecticides. The reasons for the combined evidence of *An. gambiae s.l.* susceptibility and resistance to the two insecticide classes (organophosphate and carbamate) with the same mode of action as observed in this study calls for further detailed biochemical studies to unravel the role of other factors such as the types of enzymes elevated in the resistant *Anopheles* mosquito populations from these study sites and their roles in conferring phenotypic resistance to the specific insecticide classes tested. Consistency in the monitoring of insecticide resistance in this *An. gambiae s.l.* species in Lagos should be encouraged in representative sites, using the insecticides already in use and those planned for use in malaria vector control in order to assess current situations for adaptation of appropriate vector-control strategies.

Competing interests

The authors declare that they have no competing interests.

Acknowledgements

Special thanks to the Research Assistants at the Molecular Entomology and Vector Control Research Laboratory of the Nigerian Institute of Medical Research for providing technical assistance during the field and laboratory component of this research work.

References

Adeogun, A.O., Popoola, K.O., Oduola, A.O., Olakiigbe, A.K. & Awolola, T.S. (2017). High level of DDT resistance and reduced susceptibility to deltamethrin in *Anopheles gambiae*, *Anopheles coluzzii* and *Anopheles arabiensis* from urban communities in Oyo state, South-West Nigeria. *J Mosq Res*., *7*(16), 125-133.

Antonio-Nkondjio, C., Sonhafouo-Chiana, N., Ngadjeu, C.S., Doumbe-Belisse, P., Talipouo, A., Djamouko-Djonkam, L., Kopya, E., Bamou, R., Awono-Ambene, P., Wondji, C.S. (2017). Review of the evolution of insecticide resistance in main malaria vectors in Cameroon from 1990 to 2017. *Parasit Vectors*., *10*, 472.

Aïkpon, R., Agossa, F., Ossè, R., Oussou, O., Aïzoun N., Oké-Agbo, F. & Akogbéto, M. (2013). Bendiocarb resistance in *Anopheles gambiae s.l* population from Atacora department in Benin, West Africa: a threat for malaria vector control. *Parasit Vectors*., *6***,** 192.

Alout, H., Djogbe´nou, L., Berticat, C., Chandre, F. & Weill, M. (2008). Comparison of *Anopheles gambiae* and *Culex pipiens* acetycholinesterase 1 biochemical properties. *Comp Biochem Physiol Part B*., *150*, 271-277.

Awolola, T.S., Brooke, B.D., Hunt, R.H., & Coetzee, M. (2002). Resistance of the malaria vector *Anopheles gambiae s.s* to pyrethroid insecticide in south western-Nigeria. *Ann Trop Med Parasitol*., *96*, 849–852.

Awolola, T.S., Adeogun, A., Olakiigbe, A.K., Oyeniyi, T., Olukosi, Y.A., Okoh, H., Arowolo, T., Akila, J., Oduola, A., & Amajoh, C.N. (2018). Pyrethroids resistance intensity and resistance mechanisms in *Anopheles gambiae* from malaria vector surveillance sites in Nigeria. *PLoS ONE*., *13*(12), e0205230.

Cuamba, N., Morgan, J.C., Irving, H., Steven, A., & Wondji, C.S. (2010). High level of pyrethroid resistance in an *Anopheles funestus* Population of the Chokwe District in Mozambique. *PLoS ONE*., *5*(6), e11010.

Gillies, M.T., & Coetzee, M. (1987). A Supplement to the Anophelinae of Africa South of the Sahara. *S Afr Inst Med Res*., *55*, 1–143.

Ibrahim, K.T., Popoola, K.O., Adewuyi, O.R., Adeogun, A.O., & Oricha, A.K. (2013). Susceptibility of *Anopheles gambiae sensu lato* (Diptera: Culicidae) to Permethrin, Deltamethrin and Bendiocarb in Ibadan City, Southwest Nigeria. *Curr Res J Biol Sci*., *5*(2), 42-48.

Ibrahim SS, Mukhtar MM, Datti JA, Irving H, Kusimo MO, Tchapga W, Lawal N, Sambo FI & Wondji CS. (2019). Temporal escalation of pyrethroids resistance in the major malaria vector *Anopheles coluzzii* from Sahelo-Sudanian region of northern Nigeria. *Sci Rep*., 9: 7395.

Labbe,´ P., Alout, H., Djogbe´nou, L., Pasteur, N., & Weill, M. (2011). Evolution of resistance to insecticide in disease vectors. In: Michel Tibayrenc (Ed.), *Genetics and Evolution of Infectious Diseases*. Elsevier Insights (Elsevier) 363-409.

Namountougou, M., Soma, D.D., Kientega, M., Balboné, M., Kaboré, D.P.A., Drabo, S., Coulibaly, A.Y., Fournet, F., Baldet, T., Diabaté, A., Dabiré, R.K., Gnankiné, O. (2019). Insecticide resistance mechanisms in *Anopheles gambiae* complex populations from Burkina Faso, West Africa. *Acta Trop*., *197*, 105054

National Malaria Elimination Programme (NMEP). (2016). Nigeria Malaria Indicator Survey 2015: Key Indicators. Abuja, Nigeria, and Rockville, Maryland, USA: NMEP, NPopC, and ICF International. Available: https://dhsprogram.com/pubs/pdf/MIS20/MIS20.pdf.

N'Guessan, R., Boko, P., Odjo, A., Chabi, J., Akogbeto, M., & Rowland. M. (2010). Control of pyrethroid and DDT-resistant *Anopheles gambiae* by application of indoor residual spraying or mosquito nets treated with a long-lasting organophosphate insecticide, chlorpyrifos-methyl. *Malaria J*., *9*, 44.

Obembe, A., Anyaele, O.O., & Oduola, A.O. (2014). Lessons from the implementation of LLIN distribution campaign in Ilorin Kwara State, Nigeria. *BMC Public Health*., 14: 514.

Oduola, A.O., Obansa, J.B., Ashiegbu, C.O., Adeogun, A., Otubanjo, O.A., & Awolola, T.S. (2010). High level of DDT resistance in the malaria mosquito: *Anopheles gambiae s.l*. from rural, semi urban and urban communities in Nigeria**.** *J Rural Trop Pub Health*., *9*, 114–120.

Oduola, A.O., Idowu, E.T., Oyebola, M.K., Adeogun, A.O., Olojede, J.B., Otubanjo, O.A., & Awolola, T.S. (2012). Evidence of carbamate resistance in urban populations of *Anopheles gambiae s.s*. mosquitoes resistant to DDT and deltamethrin insecticides in Lagos, South-Western Nigeria. *Parasit Vectors*., *5*, 116

Ononamadu, C.J., Datit, J.T., & Imam, A,A. (2020). Insecticide resistance profile of *Anopheles gambiae m*osquitoes: a study of a residential and industrial breeding sites in Kano Metropolis, Nigeria. *Environ Health Insights*., *14*, 1–9.

Sweileh, W.M., Sawalha, A.F., Al-Jabi, S.W., Zyoud, S.H., Shraim, N.Y., Abu-Taha, A.S. (2016). A bibliometric analysis of literature on malaria vector resistance: (1996 – 2015). *Globalization and Health*., *12*, 76.

Toé, K.H., Jones, C.M., N'Fale, S., Ismail, H.M., Dabiré, R.K., & Ranson, H. (2014). Increased pyrethroid resistance in malaria vectors and decreased bed net effectiveness, Burkina Faso. *Emerg Infect Dis***.,** *20,* 1691–1696.

Tomlin, C.D.S., (2000). *The pesticide manual, A world compendium*. British Crop Protection Council, London, UK, 12 2000.

Wahedi, J.A., Ande, A.T., Oduola, A.O., Obembe, A., Tola, M., Oyeniyi, T.A., & Awolola, T.S. (2020). Dynamics of malaria vector indices in two vegetation zones within North Eastern Adamawa State, Nigeria. *Tropical Biomed*., *37*(3), 637–649.

WHO (2006). Indoor Residual Spraying. Use of indoor residual spraying for scaling up global malaria control and elimination. whqlibdoc.who.int/hq/2006/WHO_HTM_MAL_2006.1112_eng.pdf (Nov. 26, 2012)

WHO (2012). Global plan for insecticide resistance management in malaria vectors.

Geneva: World Health Organization (WHO). http://www.who.int/malaria/publications/atoz/gpirm/

WHO. (2013). Test procedures for insecticide resistance monitoring in malaria vector mosquitoes. Geneva, World Health Organization, 2013. http://who.int/iris/bitstream/10665/80139/1/9789241505154_eng.pdf

WHO. (2015). Indoor residual spraying: an operational manual for indoor residual spraying (IRS) for malaria transmission control and elimination (Second edition). Geneva: World Health Organization, 2015. https://apps.who.int/iris/bitstream/handle/10665/177242/9789241508940_eng.pdf (accessed January 15, 2021)

WHO. (2016). WHO recommended insecticide products for treatment of mosquito nets for

malaria vector control. https://www.paho.org/hq/dmdocuments/2016/2016-cha-insecticide-list-treat-malaria.pdf

WHO. (2018). Global report on insecticide resistance in malaria vectors: 2010–2016. Geneva: World Health Organization; 2018. https://apps.who.int/iris/bitstream/handle/10665/272533/9789241514057-eng.pdf?sequence=1&isAllowed=y (accessed January 15, 2021).

WHO. (2019). World Malaria Report 2019. Geneva: World Health

Organization; 2019. http://www.who.int/malaria/.../world_malaria_report_2019/

wmr2019_full_report.pdf (accessed February 6, 2020).

WHO. (2020.) World Malaria Report 2020. Geneva: World Health

Organization; 2020. http://www.who.int/malaria/.../world_malaria_report_2020/

wmr2020_full_report.pdf (accessed January 3, 2021).