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Research Article

Mosquito Surveillance in the Seaport of Cotonou, Benin: Monitoring of Species Diversity and Assessment of Susceptibility of Mosquitoes to Insecticides

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I N F O

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A B S T R A C T

Background: Seaports are one of the most important gateways for coastal countries and deserve permanent surveillance of invasive species.

Objectives: This study aims to monitor the species diversity of mosquitoes in the seaport of Cotonou and determine the phenotypic resistance profile to multiple insecticides.

Methods: The study was conducted at the Port Autonome de Cotonou (PAC) from May to August 2022. BG sentinel traps were used to capture adults which were then identified morphologically. Additionally, larvae of *Aedes* and Culex mosquitoes were collected and reared until adult emergence. Four batches of 25 adult female mosquitoes, aged 3 to 5 days, were exposed to four insecticides (bendiocarb 0.1%, pirimiphos-methyl 0.25%, permethrin 0.75% and deltamethrin 0.05%) for 60 minutes using the WHO tube test protocol. Mosquito susceptibility was determined after 24 hours.

Results: The captured adults yielded 455 adult mosquitoes, divided into six species: *Anopheles gambiae, Anopheles pharoensis, Culex quinquefasciatus, Mansonia africana, Mansonia uniformism* and *Aedes aegypti*, with *Culex quinquefasciatus* predominating (54.15%) and *Aedes aegypti* (30.66%) being the second most abundant. The females of Culex quinquefasciatus and Aedes aegypti were exposed to insecticides. Aedes aegypti showed resistance to pyrethroids but were susceptible to bendiocarb and pirimiphos-methyl, whereas *Culex quinquefasciatus* was resistant to pyrethroids and bendiocarb but susceptible to pirimiphos-methyl.

Conclusion: Mosquito surveillance in the seaport of Cotonou is essential for detection and vector control in the event of invasion by new vectors carried by boats and cargo ships.

Keywords: Mosquito Surveillance, Invasive Species, Susceptibility, Aedes

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Introduction

Invasive exotic mosquito species in new territories constitute a growing major public health concern due to their potential role in the transmission of several pathogens.[1] Rapid worldwide dispersal of mosquito vectors of human diseases is leading to an ever-increasing number of epidemic zones.[2–4]

Exotic mosquito species can be passively transported from one area to another by human activities.[5] However, the large global flow of people and commodities has greatly increased the risk of introducing invasive mosquito species.[6]

Early detection of the introduction of invasive mosquitoes is possible when a good surveillance system is implemented. This has been the case in New Zealand, the Netherlands and Portugal.[7–9] However, this has not been the case in many other countries where invasive mosquito species, such as *Aedes albopictus,* have succeeded in colonising the country.

There is strong evidence that surveillance of entry points to countries such as seaports, border crossings, and airports helps to curb the invasion and establishment of invasive mosquito species.[11]

The maritime port of Cotonou is one of the most important gateways of commodities into Benin and there is an increased flow of goods coming from and going to all the continents from here.

To deal with this threat to public health, mosquito surveillance is more than important in the maritime port of entry in Cotonou to allow the early detection and elimination of invasive mosquito vectors. Accordingly, the Port of Cotonou is equipped with a medical entomology laboratory to facilitate the monitoring and control of mosquitoes in the port area.

Our objective is to monitor the diversity of mosquito species and to assess their susceptibility to various insecticides at the seaport of Cotonou.

Methods

Collection Sites

The Port Autonome de Cotonou is located in the south of Benin, in the economic capital. It boasts a vast stretch of water covering an area of over 400,000 m^2. It serves a number of landlocked countries to the north of Benin, such as Mali, Niger and Burkina Faso, which have no maritime advantages. In the sub-region, the port of Cotonou is approximately equidistant from the ports of Lagos (Nigeria) and Lomé (Togo), at 115 km and 135 km respectively, making it the closest and fastest relay and trans-shipment port in Nigeria. It handles over 60% of Benin's trade and has been described as the country's economic lung. It is also the leading transit port for foodstuffs and manufactured goods in the Republic of Niger.

Figure 1.A Map showing the Location of the Mosquito Collection Sites in Maritime Port of Cotonou, Benin

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Adult Mosquito Collections

Mosquitoes were captured at 11 selected sites from May to August 2022 using BG sentinel traps baited with CO_2 and a human odour imitator.[12] Mosquitoes caught after 24 hours were sent to the Port laboratory and then identified morphologically using taxonomic keys.[13] Mosquito species diversity as well as the abundance of each species were assessed.

Larvae Collection and Insecticide Susceptibility Tests

Culex and *Aedes* larvae breeding sites are the most prevalent in the port. Larvae and pupae, presumed to be *Aedes* and *Culex* mosquitoes, were collected in the port area using a plastic cup and pipette. Larvae were collected from potential breeding sites and transported in well-labelled jars to the insectarium, where they were reared under the required temperature and relative humidity conditions (28 ± 2 °C and 72 ± 5%) until they reached the adult stage.

All collected immature specimens were reared in the laboratory until adults emerged. Four insecticides of interest to public health were tested according to the World Health Organization (WHO) protocol: a carbamate (bendiocarb 0.1%), an organophosphate (pirimiphos-methyl 0.25%), and two pyrethroids (permethrin 0.75% and deltamethrin 0.05%).

During a 60-minute exposure period, the sensitivity test was carried out on unfed females in batches of 25, aged 3 to 5 days on insecticide-treated paper. 25 females of both species were introduced into each tube and monitored at different time intervals (15, 30, 45 and 60 minutes), with the number of "killed" females recorded. After one hour's exposure, all mosquitoes were transferred to observation tubes and fed with sweet juice soaked in cotton. Batches exposed to untreated papers were used as the control. Mortalities were recorded after 24 hours and the susceptibility status of the population was graded according to the WHO protocol.[14] Dead and surviving mosquitoes from this bioassay were kept separately in tubes and stored in a freezer at -20 °C for subsequent molecular analysis.

Data Analysis

The resistant status of mosquito samples was determined according to the WHO criteria.[14] As per the protocol, if the mortality rate is > 98%, the population is considered totally susceptible; if the rate is between 90% and 98%, the population is suspected of being resistant, and when it is < 90%, the population is considered resistant to the insecticide tested.

Results

Species Diversity and Abundance

A total of 349 specimens of mosquitoes were collected and grouped into 6 species (Table 1). *Culex* quinquefasciatus (54.15%) and *Aedes* aegypti (30.66%) were the most abundant species caught. The other species (Anopheles pharoensis, Anopheles gambiae s.l, Mansonia africana) were poorly represented (less than 10% of the population). Large variations were observed over months in terms of the abundance of mosquitoes. May was the month with relatively more mosquitoes than the other three months.

Insecticide Susceptibility Status of Culex and *Aedes* Mosquitoes

A total of 294 females of Culex mosquitoes and 399 females of *Aedes* mosquitoes (3–5 days old) from the maritime port of Cotonou were exposed to insecticides. *Aedes* showed resistance to pyrethroids and probable resistance to bendiocarb but was found to be sensitive to pirimiphos-methyl (Figure 2). Culex mosquitoes showed resistance to deltamethrin and permethrin but were sensitive to bendiocarb and pirimiphos-methyl (Figure 3).

Table 1.Species of Mosquitoes Caught from May to August 2022 in the Maritime Port of Cotonou

Species	Number of Mosquitoes Collected				Total	Percentage
	May	June	July	August		
Anopheles gambiae s.l	5	4	5	1	15	4.30
Anopheles pharoensis	9	0	0	0	9	2.58
Aedes aegypti	44	29	21	13	107	30.66
Culex quinquefasciatus	71	50	39	29	189	54.15
Mansonia africana	8	6	6	7	27	7.74
Mansonia uniformis	2	0	0	0	2	0.57
Total	139	89	71	50	349	100.00

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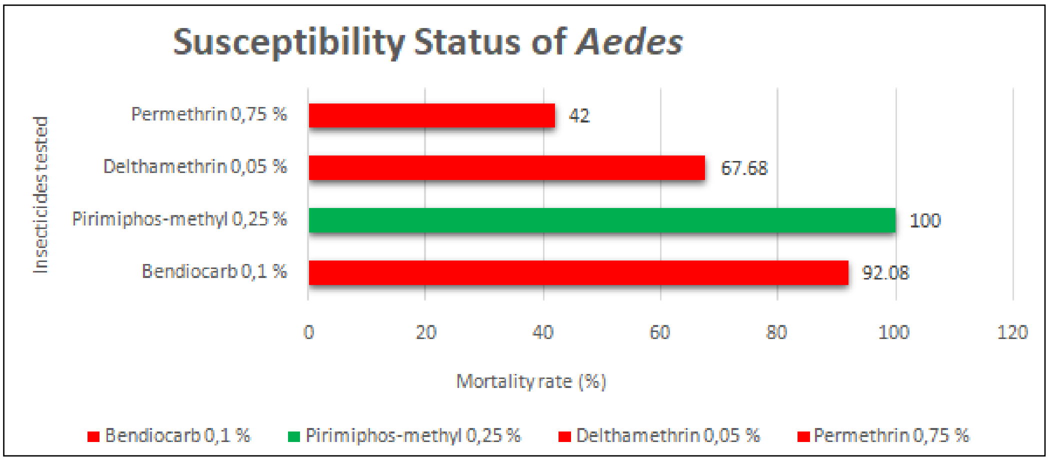

Figure 2.Susceptibility *Status* of *Aedes*

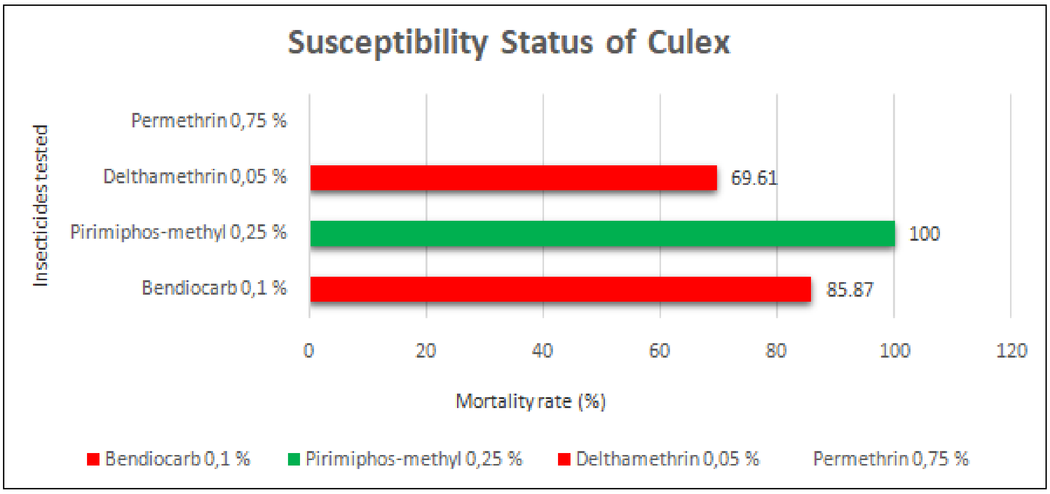

Figure 3.Susceptibility *Status* of *Culex*

Discussion

The surveillance of disease-carrying mosquito species in seaports is essential for early detection of invasive species and the pathogens they carry. The present study investigated the diversity, the abundance of mosquito species and their susceptibility to various insecticides in the maritime port of Cotonou. Our results showed that various mosquito vector species were present in great numbers in the port in this study, with *Cx. quinquefasciatus* and *Ae. aegypti* being the most abundant species. Culicid diversity was low compared with previous studies carried out in Cotonou by Padonou et al. and Agbanrin et al.[15,16] who reported 13 species of adult mosquitoes coming from larval sampling methods and 15 species of adult mosquitoes coming from different traps and HLC. This might be due to the collection method and the relatively short duration of collection. In addition, this low diversity may be due to the ecology of the environment, which was highly anthropized in line with the habitat of *Aedes* and *Culex*, sampling methods, and the mosquito collection period. Also, the lower number of mosquitoes collected can be explained by the regular indoor residual spraying activities implemented in the port.

The abundance of Aedes, particularly *Aedes aegypti,* can be explained by the existence of used tyres stored in the port, a preferred breeding site for the species. This result confirmed what was found in southern Cameroon[17] and in the Central African Republic.[18]

The results of the susceptibility test showed the populations of *Ae. aegypti* and *Cx. quinquefasciatus* to be highly susceptible to pirimiphos-methyl (mortality rate of 100%). However, they were resistant to deltamethrin. Aedes aegypti also shown resistance to permethrin. Resistance was suspected for bendiocarb (mortality rate of 92.08% for Ae. aegypti and 85.87% for Cx. quinquefasciatus). Of these four insecticides tested in the maritime port of Cotonou, it was found that pirimiphos-methyl showed promising effectiveness towards mosquito control. This has been proven by its 100% mortality rate.

The high resistance of tested mosquitoes to pyrethroids (permethrin and deltamethrin) strongly suggests that these insecticides should not be recommended for mosquito control in the port. Resistance to this class of insecticide has been reported in several studies in Benin.[19–25]

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Mosquito surveillance should be stepped up in the Cotonou seaport in the long term to not only enable early detection of invasive vector species and pathogens but also for early control of their spread outside the maritime zone.

Conclusion

Mosquito surveillance in the seaport of Cotonou is essential for the detection and vector control in the event of invasion by new vectors carried by boats and cargo ships. This study has enabled us to compile a non-exhaustive list of the species present in the port of Cotonou. In addition, it helps in decision-making concerning the insecticide selection for mosquito control operations to devise more effective control strategies to reduce populations of vector mosquito species in the port areas and decrease the risk of arbovirus introduction.

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References

1. Schaffner F, Medlock JM, Bortel WV. Public health significance of invasive mosquitoes in Europe. Clin Microbiol Infect. 2013;19(8):685-92. [PubMed] [Google Scholar]
2. Wilke AB, Beier JC, Benelli G. Complexity of the relationship between global warming and urbanization–an obscure future for predicting increases in vector-borne infectious diseases. Curr Opin Insect Sci. 2019;35:1-9. [PubMed] [Google Scholar]
3. Monaghan AJ, Eisen RJ, Eisen L, McAllister J, Savage HM, Mutebi JP, Johansson MA. Consensus and uncertainty in the geographic range of Aedes aegypti and Aedes albopictus in the contiguous United States: multi-model assessment and synthesis. PLoS Comput Biol. 2019;15(10):e1007369. [PubMed] [Google Scholar]
4. Wilke AB, Vasquez C, Carvajal A, Moreno M, Petrie WD, Beier JC. Evaluation of the effectiveness of BG-Sentinel and CDC light traps in assessing the abundance, richness, and community composition of mosquitoes in rural and natural areas. Parasit Vectors. 2022;15(1):51. [PubMed] [Google Scholar]
5. Reiter P. Aedes albopictus and the world trade in used tires, 1988-1995: the shape of things to come? J Am Mosq Control Assoc. 1998;14(1):83-94. [PubMed] [Google Scholar]
6. Tatem AJ, Hay SI, Rogers DJ. Global traffic and disease vector dispersal. Proc Natl Acad Sci U S A. 2006;103(16):6242-7. [PubMed] [Google Scholar]
7. Ammar SE, McIntyre M, Swan T, Kasper J, Derraik JG, Baker MG, Hales S. Intercepted mosquitoes at New Zealand's ports of entry, 2001 to 2018: current status and future concerns. Trop Med Infect Dis. 2019;4(3):101. [PubMed] [Google Scholar]
8. Ibañez-Justicia A, Gloria-Soria A, den Hartog W, Dik M, Jacobs F, Stroo A. The first detected airline introductions of yellow fever mosquitoes (Aedes aegypti) to Europe, at Schiphol International airport, the Netherlands. Parasit Vectors. 2017;10(1):603. [PubMed] [Google Scholar]
9. Osório HC, Ze-Ze L, Neto M, Silva S, Marques F, Silva AS, Alves MJ. Detection of the invasive mosquito species Aedes (Stegomyia) albopictus (Diptera: Culicidae) in Portugal. Int J Environ Res Public Health. 2018;15(4):820. [PubMed] [Google Scholar]
10. 10. Wilke AB, Benelli G, Beier JC. Beyond frontiers: on invasive alien mosquito species in America and Europe. PLoS Negl Trop Dis. 2020;14(1):e0007864. [PubMed] [Google Scholar]
11. Schmidt TL, Rooyen AR, Chung J, Endersby-Harshman NM, Griffin PC, Sly A, Hoffman AA, Weeks AR. Tracking genetic invasions: genome-wide single nucleotide polymorphisms reveal the source of pyrethroid-resistant Aedes aegypti (yellow fever mosquito) incursions at international ports. Evol Appl. 2019;12(6):1136-46. [PubMed] [Google Scholar]
12. Wilke AB, Carvajal A, Medina J, Anderson M, Nieves VJ, Ramirez M, Vasquez C, Petrie W, Cardenas G, Beier JC. Assessment of the effectiveness of BG-Sentinel traps baited with CO_2 and BG-Lure for the surveillance of vector mosquitoes in Miami-Dade County, Florida. PLoS One. 2019;14(2):e0212688. [PubMed] [Google Scholar]
13. Azari-Hamidian S, Harbach RE. Keys to the adult females and fourth-instar larvae of the mosquitoes of Iran (Diptera: Culicidae). Zootaxa. 2009;2078:1-33. [Google Scholar]
14. World Health Organization [Internet]. Guidelines for Testing Mosquito Adulticides Intended for Indoor Residual Spraying (IRS) and Insecticide Treated Nets (ITNs). Geneva: WHO/CDS/NTD/ WHOPES/ GCDDP/2006.3; 2006. Available from: https://www.google.com/search?q=13.+WHO%3A+Guidelines+for+Testing+Mosquito+Adulticides+Intended+for+Indoor+Residual+Spraying+(IRS)+and+Insecticide+Treated+Nets+(ITNs).+Geneva%3A+WHO%2FCDS%2FNTD%2F+WHOPES%2FGCDDP%2F2006.3%3B+2006%3A2&oq=13.%09WHO%3A+Guidelines+for+Testing+Mosquito+Adulticides+Intended+for+Indoor+Residual+Spraying+(IRS)+and+Insecticide+Treated+Nets+(ITNs).+Geneva%3A+WHO%2FCDS%2FNTD%2F+WHOPES%2FGCDDP%2F2006.3%3B+2006%3A2&aqs=chrome..69i57

j69i60.1008j0j7&sourceid=chrome&ie=UTF-8
15. Padonou GG, Sezonlin M, Gbedjissi GL, Ayi I, Azondekon R, Djenontin A, Bio-Bangana S, Oussou O, Yadouleton A, Boakye D, Akogbeto M. Biology of Anopheles gambiae and insecticide resistance: entomological study for a large scale of indoor residual spraying in south east Benin. J Parasitol Vector Biol. 2011;3(4):59-68. [Google Scholar]
16. Agbanrin R, Padonou GG, Anges Y, Attolou R, Badirou K, Govoetchan R, Gnanaguenon V, Sovi A, Anagonou R, Akogbeto M. Abundance and diversity of culicidae fauna at Cotonou in Southern Benin. Int J Curr Res [Internet]. 2015;7(3):14085-91. Available from: https://www.researchgate.net/publication/274953677_ABUNDANCE_AND_DIVERSITY_OF_CULICIDAE_FAUNA_AT_COTONOU_IN_SOUTHERN_BENIN
17. Kamgang B, Happi JY, Boisier P, Njiokou F, Herve JP, Simard F, Paupy C. Geographic and ecological distribution of the dengue and chikungunya virus vectors Aedes aegypti and Aedes albopictus in three major Cameroonian towns. Med Vet Entomol. 2010;24(2):132-41. [PubMed] [Google Scholar]
18. Kamgang B, Ngoagounj C, Manirakiza A, Nakoune E, Paupy C, Kazanji M. Temporal patterns of abundance of Aedes aegypti and Aedes albopictus (Diptera: Culicidae) and mitochondrial DNA analysis of Ae. albopictus in the Central African Republic. PLoS Negl Trop Dis. 2013;7(12):e2590. [PubMed] [Google Scholar]
19. Sagbohan HW, Kpanou CD, Osse R, Dagnon F, Padonou GG, Sominahouin AA, Salako AS, Sidick A, Sewade W, Akinro B, Ahmed S, Impoinvil D, Agbangla C, Akogbeto M. Intensity and mechanisms of deltamethrin and permethrin resistance in Anopheles gambiae s.l. populations in southern Benin. Parasit Vectors. 2021;14(1):202. [PubMed] [Google Scholar]
20. Badirou K, Osse R, Yadouleton A, Attolou R, Youssouf RA, Gnaguenon V, Akinrou B, Govoetchan R, Akogbeto M. Redistribution of kdr L1014F and ACE-1 mutations involved in the resistance of vectors to insecticides in Benin. Asian J Pharm Sci Technol. 2016;6(1):48-57. [Google Scholar]
21. Djègbè I, Boussari O, Sidick A, Martin T, Ranson H, Chandre F, Akogbeto M, Corbel V. Dynamics of insecticide resistance in malaria vectors in Benin: first evidence of the presence of L1014S kdr mutation in Anopheles gambiae from West Africa. Malar J. 2011;10:261. [PubMed] [Google Scholar]
22. Padonou GG, Gbedjissi G, Yadouleton A, Azondekon R, Razack O, Oussou O, Gnanguenon V, Rock A, Sezonlin M, Akogbeto M. Decreased proportions of indoor feeding and endophily in Anopheles gambiae s.l. populations following the indoor residual spraying and insecticide-treated net interventions in Benin (West Africa). Parasit Vectors. 2012;5:262. [PubMed] [Google Scholar]
23. Yadouleton AW, Padonou G, Asidi A, Moiroux N, Bio-Banganna S, Corbel V, N'guessan R, Gbenou D, Yacoubou I, Gazard K, Akogbeto MC. Insecticide resistance status in Anopheles gambiae in southern Benin. Malar J. 2010;9:83. [PubMed] [Google Scholar]
24. Salako AS, Ahogni I, Kpanou C, Sovi A, Azondekon R, Sominahounin AA, Tokponnon F, Gnanguenon V, Dagnon F, Iyikirenga L, Akogbeto MC. Baseline entomologic data on malaria transmission in prelude to an indoor residual spraying intervention in the regions of Alibori and Donga, Northern Benin, West Africa. Malar J. 2018;17(1):392. [PubMed] [Google Scholar]
25. Sovi A, Djegbe I, Soumanou L, Tokponnon F, Gnanguenon V, Azondekon R, Oke-Agbo F, Oke M, Adechoubou A, Massougbodji A, Corbel V, Akogbeto M. Microdistribution of the resistance of malaria vectors to deltamethrin in the region of Plateau (southeastern Benin) in preparation for an assessment of the impact of resistance on the effectiveness of Long Lasting Insecticidal Nets (LLINs). BMC Infect Dis. 2014;14:103. [PubMed] [Google Scholar]

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