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Article

Diversity and Abundance of Mosquitoes in the Seaport Area of San Pedro (Côte d'Ivoire)

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I N F O

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A B S T R A C T

Introduction: As part of the implementation of arbovirus vector surveillance at the Seaport of San Pedro by the International Health Regulations, entomological surveys were conducted from March to July 2018 to determine the diversity and abundance of putative arbovirus vectors.

Method: Mosquitoes were collected as immatures and adult stages in the activity and residential areas of the San Pédro Seaport platform.

Results: A total of 7 and 14 species of mosquitoes were collected in the activity and residential areas respectively, with a dominance of the genera *Culex* species (5 in the activity area and 10 in the residential area). Only *Aedes aegypti* had a high frequency of occurrence with 47.29% in the activity area and 61.97% in the residential area. This species was accessory in activity areas and regular in residential areas. The diversities indices were higher in the residential area with 1.27, 0.64, and 0.48 respectively for Shannon's Index, Simpson's Index, and Equitability Index. *Aedes aegypti* accounted for 78.42% of the mosquitos collected in activity area and 49.47% of those collected in the residential area with an exophilic respective of 88.32% and 75.21%.

Conclusion: Aedes aegypti abundance at the Seaport of San Pedro constitutes a permanent health threat that requires appropriate control and surveillance actions.

Keywords: Diversity, Abundance, Mosquitoes, San Pedro, Seaport Area, Côte d'Ivoire

Introduction

Vector-borne diseases are a public health problem in the world, particularly in tropical areas. They are responsible for almost 20% of the global burden of infectious diseases.[1] In recent decades, and particularly since the 1980s, these arbovirus diseases have increased worldwide.[2]

Arthropods (sandflies, lice, bugs, ticks, and mosquitoes) are responsible for the transmission and maintenance of the life cycle of these arboviruses.[3] However, the main vectors are *Aedes albopictus* (Skuse, 1894) and *Aedes aegypti* (Linnaeus, 1762), with the ability to transmit dengue fever, yellow fever, chikungunya and Zika.[3-5] The spread of these arboviruses and their vectors has been facilitated by globalisation and increased trade in goods and people.[6] Thus, today, no region is immune to the risks of importation.[7]

In response to this ongoing threat, the International Health Regulations recommend establishing vector surveillance systems at points of entry and developing evidence-based vector control strategies to prevent the spread of the diseases they transmit.[8] Its implementation has enabled New Zealand, Netherlands, and Portugal to prevent the invasion of their territories by mosquito species.[9]

In Côte d'Ivoire, the health authorities, aware of the threat posed by the discovery of *Aedes albopictus* in the seaport of Abidjan,[10,11] decided to take stock of the situation in the country's second port. Within this framework, this study was initiated in the port of San Pedro to strengthen global health security by implementing the IHR recommendations. The study required knowledge of the diversity and abundance of mosquitoes in the seaport of San Pedro.

Methodology

Study Area and Period

A study, descriptive and transversal, was conducted at the seaport of San Pédro (4° 44′ 41″ North and 6° 38′ 23″ West), in the southwest of Côte d'Ivoire, 368 km from Abidjan.[12] The climate is sub-equatorial and composed of two dry seasons (one from December to February and the other from July to August) and two wet seasons (one from March to June and the other from September to November). Annual rainfall and temperature ranged from 1,200 to 1,500 mm and 24 to 27 °C respectively.[13] Spread over 2,000 hectares, the seaport of San Pedro is divided into three sectors, namely the commercial port sector, fishing port sector, and industrial and commercial sector.[14]

Port activities consist mainly of exports, with average annual growth rates of 11.31%.[15] It is the second-largest seaport in the country and the first cocoa export port in the world. The San Pedro port does 67% of its trade with Europe, 25% with America, and 8% with Asia.[13] Mosquitoes were collected monthly from March to July 2018 as immature forms by inspecting waterbodies and as adults (resting catch) from the activity and residential areas, which were approximately at a distance of 400 m from each other. Six study sites were selected of which four were in the activity area (SEPBA, fishing port, sub-customs, and health centre) and two were in the residential area (General Manager city and Rade city) (Figure 1).

Larval Surveys

Larval surveys were conducted at each site. All aquatic habitats were examined for mosquito larvae, and pupae in the described habitats and sampled using a 5 ml pipette for small waterbodies and standard dipping methods for medium and large waterbodies to determine larval and pupae densities. The immature stages of mosquitoes collected were preserved in jars and labelled according to the breeding site, type of water bodies, and date. These immature populations contained in the jars were brought back to the insectary of the Institut National d'Hygiène Publique (INHP) and reared to the adult stage and identified to genera and species using morphological identification keys of Edwards[16] and Huang.[17]

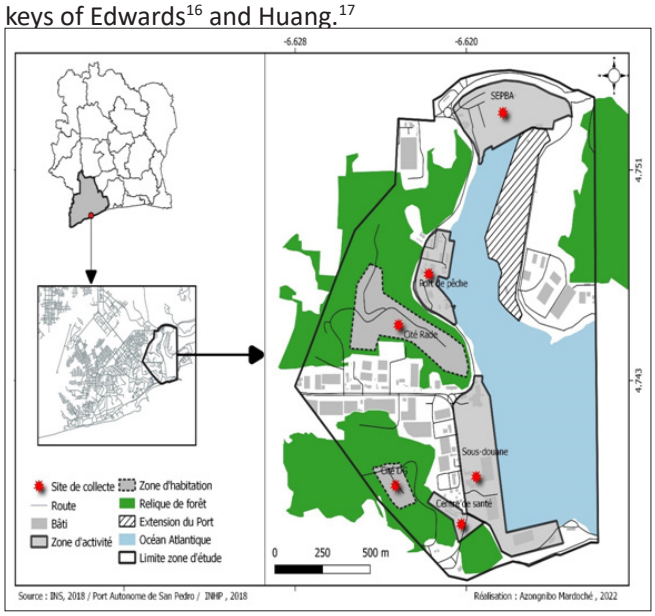

Figure 1.Geographical Location of the San Pedro Port Study Sites

Resting Fauna Catch

Adult mosquitoes were collected from 06:00 to 08:00 am at the resting habitats from each study site. Mosquitoes were collected by Prokopack, for 5 minutes for two consecutive days per site. Collected Culicidae was identified as genera and species using morphological identification keys of Edwards[16] and Huang.[17]

Data Analysis

Three ecological structure indices (Shannon Index, Simpson Index (1-D), and equitability) and the ecological composition indices (Species richness (S) and relative abundance (C))

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were determined using Past software version 4.02. The Shannon diversity index (H' = -∑ ((Pi) × $\log_2$ (Pi)) quantifies the heterogeneity of the biodiversity of a study area. This index varies between 0 (a single species encountered) and 4.5 (very high diversity).[18] Simpson's index (D = Σ ni(ni-1))/N (N-1) evaluates the probability that 2 individuals, drawn at random from a population, belong to the same species.[19] This index ranges from 0 (maximum diversity) to 1 (minimum diversity).[19] The equitability index (E' = H'/$\log_2$(S)), also known as regularity, measures the fair distribution of taxa. This index ranges between 0 (one species dominates) and 1 (all species have the same abundance).[19] The species richness (total number of species sampled in an area; S)[19] and the variation in relative abundance (C = ni/N*100)[20] between areas were compared using R software version 3.6.2. Consistency or frequency of occurrence is the number of records that contain the species of interest concerning the total number of records. Six categories of species are found: rare species (C < 5%), incidental species (5% ≤ C < 25%), accessory species (25% ≤ C < 50%), regular species (50% ≤ C < 75%), constant species (75% ≤ C < 100%), and ubiquitous species (C = 100%).[21] A Pearson correlation test was used to determine the strength of the association between certain variables.

Results

Culicidae Composition

Overall, 14 species belonging to five genera (*Aedes, Anopheles, Culex, Eretmapodites*, and *Mansonia*) were collected. Of these, 10 belonged to the genera *Culex* (*Cx. albiventris*, *Cx. fraseri*, *Cx. horridus*, *Cx. nebulosus*, *Cx. poicilipes*, *Cx. quinquefasciatus*, *Cx. rima*, *Cx. tigripes*, *Cx. Theileri,* and *Cx. weschei*). The other genera *Aedes* (*Ae. aegypti*), *Eretmapodites* (*Er. quinquevittatus*), *Mansonia* (*Ma. uniformis*) and *Anopheles* (*An. gambiae*) were each represented by one species. In the activity area, 7 species were collected, including 5 of the genera *Culex* and one each of the genera *Aedes* and *Anopheles*. However, in the residential area, all 14 species of the five genera were collected (Table 1).

Table 1.Distribution of Culicidae in the Activity Area and Residential Area

Family	Genera	Species	Activity Area	Residential Area
Culicinae	*Aedes*	*Ae. aegypti*	+	+
		S1	1	1
	Culex	*Cx. albiventris*	+	+
		Cx. fraseri	+	+
		Cx. horridus	-	+
		Cx. nebulosus	-	+
		Cx. poicilipes	-	+
		Cx. quinquefasciatus	+	+
		Cx. rima	-	+
		Cx. theileri	-	+
		Cx. tigripes	+	+
		Cx. weschei	+	+
		S2	5	10
	Eretmapodites	*Er. quinquevittatus*	-	+
		S3	0	1
	Mansonia	*An. uniformis*	-	+
		S4	0	1
Anophelinae	*Anopheles*	*An. gambiae*	+	+
		S5	1	1
Total number of species			7	14

Table 2.Frequency of Occurrence of Culicidae in the Activity Area and Residential Area

Family	Genera	Species	Activity Area		Residential Area	
Culicinae	*Aedes*	*Ae. aegypti*	47.29	Accessory	61.97	Regular
	Culex	*Cx. albiventris*	2.33	Rare	5.63	Incidental
		Cx. fraseri	2.33	Rare	1.41	Rare
		Cx. horridus	0.00	-----	4.23	Rare

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		Cx. nebulosus	0.00	------	1.41	Rare
		Cx. poicilipes	0.00	-----	1.41	Rare
		Cx. quinquefasciatus	20.93	Incidental	29.58	Accessory
		Cx. rima	0.00	-----	1.41	Rare
		Cx. theileri	0.00	-----	2.82	Rare
		Cx. tigripes	0.78	Rare	2.82	Rare
		Cx. weschei	0.78	Rare	5.63	Incidental
	Eretmapodites	*Er. quinquevittatus*	0.00	-----	1.41	Rare
	Mansonia	*An. uniformis*	0.00	-----	4.23	Rare
Anophelinae	*Anopheles*	*An. gambiae s.l.*	7.75	Incidental	5.63	Incidental
Total			57.36	Regular	74.64	Regular

Table 3.Abundance of Culicidae in the Activity Area and Residential Area

Species	Activity Area	Residential Area
	N (%)	N (%)
Ae. aegypti	985 (78.42)	744 (49.47)
An. gambiae	20 (1.59)	17 (1.13)
Cx. albiventris	14 (1.11)	133 (8.84)
Cx. fraseri	15 (1.19)	3 (0.20)
Cx. horridus	0 (0.0)	19 (1.26)
Cx. nebulosus	0 (0.0)	1 (0.07)
Cx. poicilipes	0 (0.0)	1 (0.07)
Cx. quinquefasciatus	219 (17.44)	491 (32.65)
Cx. rima	0 (0.0)	2 (0.13)
Cx. theileri	0 (0.0)	2 (0.13)
Cx. tigripes	1 (0.08)	1 (0.07)
Cx. weschei	2 (0.16)	8 (0.53)
Er. quinquevittatus	0 (0.0)	1 (0.07)
Ma. uniformis	0 (0.0)	81 (5.39)
Total	1256 (100)	1504 (100)

Frequency of Occurrence of Culicidae

The encountered species had higher rates of occurrence in the residential area than in the activity area, except for *Culex fraseri* and *Anopheles gambiae* which retained their category (accidental) regardless of the area. *Aedes aegypti* moved from the incidental species in the activity area to the regular species in the residential area with a respective occurrence frequency of 47.29% and 61.97%. *Culex quinquefasciatus* was an accidental (20.23%) and incidental (29.58%) species in the activity and residential areas respectively. *Culex albiventris* and *Culex weschei* were both moved from the rare species of 2.33% and 0.78% to the incidental species of 5.63% and 5.63% respectively from the activity area to the residential area (Table 2).

Culicidian Diversity

In the activity and residential areas, the diversity indices were respectively 0.68 and 1.27 for the Shannon index, 0.35 and 0.64 for the Simpson index, and 0.35 and 0.48 for the equitability index (Figure 2). The Shannon and Simpson indices showed that more mosquito species were collected in residential areas than in activity areas. Furthermore, the equitability index showed that in the activity area, *Aedes*

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aegypti (78.42%) dominated all species. However, in the residential area, despite the dominance of *Aedes aegypti* (49.47%), mosquitoes were significantly better distributed within species. In both the activity and residential areas, *Aedes aegypti* was statistically more abundant than the other species encountered (p value < 0.01) (Table 3).

Resting Density of Mosquitoes

A total of 635 resting mosquitoes had collected at the port of San Pedro, of which 50.08% (n = 318) were *Aedes aegypti,* and 49.92% were other mosquitoes. Of the mosquitoes collected, 273 (42.99%) were obtained in the activity area and 362 (57.01%) in the residential area. Mosquitoes collected in the residential area were statistically more abundant than those collected in the activity area (Chi-square = 12.47 and p value = 0.0004) (Table 4). In the activity area, *Aedes aegypti* accounted for more than two-thirds (72.16%; n = 197) of the mosquitoes collected with a marked abundance (88.32%) outdoors. In contrast, in the residential area, *Culex quinquefasciatus* (60.22%; n = 218) was more abundant followed by *Aedes aegypti* (33.43%; n = 121) (Chi-square = 27.75, and p value < 0.0001). Despite the low rate of *Aedes aegypti* collected at rest in the residential area, this species was statistically more abundant outdoors (75.21%) than indoors (24.79%) (Chi-square = 30.75 and p value < 4.32e-8) (Table 4).

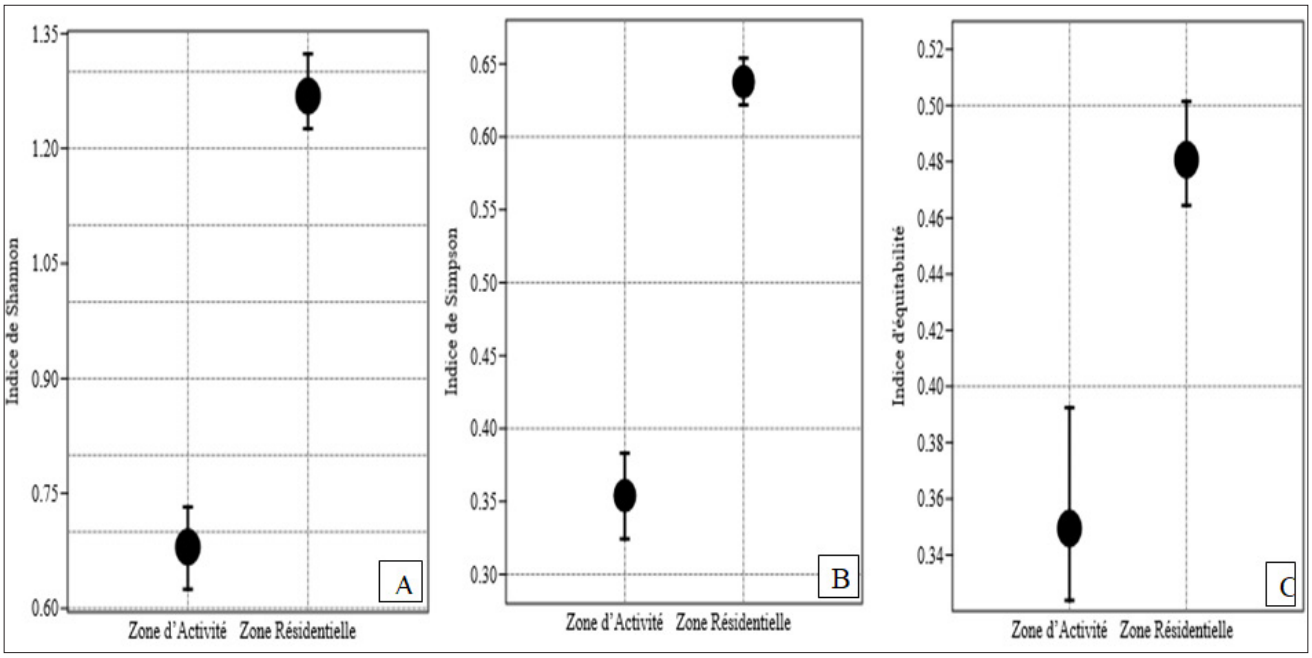

Figure 2.Diversity of Culicidae in the Activity Area and Residential Area

Table 4.Resting Density of Mosquitoes in the Activity Area and Residential Area

Area	Species	Number	Outdoor		Indoor		Chi-square	p Value
			N	%	N	%		
Activity area	*Ae. aegypti*	197	174	88.32	23	11.68	115.74	< 0.001
	An. gambiae sl	3	1	33.33	2	66.67	---	---
	Cx. albiventris	0	0	0.00	0	0.00	---	---
	Cx. fraseri	0	0	0.00	0	0.00	---	---
	Cx. horridus	0	0	0.00	0	0.00	---	---
	Cx. poicilipes	0	0	0.00	0	0.00	---	---
	Cx. quinquefasciatus	72	27	37.50	45	62.50	4.5	0.033
	Cx. theileri	0	0	0.00	0	0.00	---	---
	Cx. weschei	1	1	100	0	0.00	---	---
	Ma. uniformis	0	0	0.00	0	0.00	---	---
	Subtotal 1	273	203	74.36	70	25.64	64.90	< 0.001

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Residential area	*Ae. aegypti*	121	91	75.21	30	24.79	30.75	< 0.001
	An. gambiae sl	5	5	100	0	0.00	---	---
	Cx. albiventris	1	1	100	0	0.00	---	---
	Cx. fraseri	2	2	100	0	0.00	---	---
	Cx. horridus	8	8	100	0	0.00	---	---
	Cx. poicilipes	1	0	0.00	1	100	---	---
	Cx. quinquefasciatus	218	57	26.15	161	73.85	49.61	< 0.001
	Cx. theileri	1	1	100	0	0.00	---	---
	Cx. weschei	3	0	0.00	3	100	---	---
	Ma. uniformis	2	2	100	0	0.00	---	---
	Subtotal 2	362	167	46.13	195	53.87	2.16	0.1411

Table 5. Triangular Matrix comparing the Variables studied in the Activity Area and Residential Area

		Tem	RS	Shan Index	Simp Index	Equi Index	Ab Culic	Ab *Ae. aegypti*	Ab *Cx. quinque*	Fre Cul	Fre *Ae. aegypti*	Fre *Cx. quinque*
Precipitation	Correlation	-0.40	0.21	0.30	0.10	0.10	0.650	0.70	0.30	0.44	0.40	-0.322
	p value	0.25	0.37	0.31	0.44	0.44	0.235	0.06	0.31	0.45	0.50	0.597
Temperature	Correlation		-0.71	-0.60	-0.80	0.20	-0.38	-0.10	-0.90	-0.76	-0.51	-0.62
	p value		0.178	0.14	0.05	0.37	0.521	0.44	0.02	0.13	0.38	0.26
Species richness	Correlation			0.72	0.82	-0.10	0.363	0.36	0.67	0.78	0.67	0.98
	p value			0.09	0.04	0.43	0.548	0.28	0.11	0.11	0.21	0.004
Shannon index	Correlation				0.90	0.60	0.531	0.30	0.20	0.87	0.79	0.72
	p value				0.02	0.14	0.357	0.31	0.37	0.06	0.11	0.16
Simpson index	Correlation					0.30	0.414	0.00	0.50	0.83	0.64	0.67
	p value					0.31	0.488	0.50	0.20	0.08	0.24	0.22
Equitability index	Correlation						0.022	0.10	-0.60	0.16	0.11	-0.08
	p value						0.972	0.44	0.14	0.80	0.86	0.90
Abundance Culicidae	Correlation							0.90	0.10	0.80	0.92	0.18
	p value							0.02	0.44	0.10	0.029	0.77
Abundance *Ae. aegypti*	Correlation								0.00	0.36	0.614	-0.26
	p value								0.50	0.55	0.270	0.67
Abundance *Cx. quinquefasciatus*	Correlation									0.47	0.201	0.51
	p value									0.42	0.745	0.38
Frequency Culicidae	Correlation										0.94	0.66
	p value										0.01	0.23
Frequency *Ae. aegypti*	Correlation											0.53
	p value											0.35

Correlation between the Variables Studied

The Pearson correlation test was used to evaluate the relationships between several variables studied. The temperature was negatively correlated with species richness, Simpson's index, and culicid abundance. The Simpson index was positively correlated with species richness and the Shannon index. The frequency of occurrence of culicids was positively correlated with the frequency of occurrence of *Aedes aegypti* (Table 5).

Discussion

The entomological surveys carried out at the port of San

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Pedro to determine the diversity and abundance of Culicidae resulted in the collection of 7 and 14 mosquito species in the activity and residential areas, respectively. In both areas, 70% of the identified species belonged to the genera *Culex*. The species richness of the *Culex* genera can be explained by their ability to adapt to several types of breeding sites. These results differ from those of Konan et al.[10] and Yao et al.[11] at the port of Abidjan in Côte d'Ivoire and those of Bhadauriya et al.[6] at the port of Kandla in India. In these works, carried out in different ports, mosquitoes were collected with very few species of the genera *Culex*. The specific variation in mosquito genera from one study to another could be explained by different collection methods used. In their studies, the authors used either trap-and-lay or larval surveys respectively. The majority of the mosquito species collected had a higher frequency of occurrence in the residential area than in the activity area. These results show that mosquitoes were present in more abundance in residential areas than in activity areas. The regular and accessory categories of *Aedes aegypti*, and *Cx. quinquefasciatus*, respectively, would be related to the urban nature of these two species.[22] The results are in agreement with those of Braks et al.,[23] who found the presence of *Aedes aegypti* in more than 55% of the surveys in Rio de Janeiro. Also, in the work of Merabeti et al.,[24] and Bouabida et al.[25] the majority of species encountered were in the rare and accidental categories, which was the case in this study. The Shannon (1.27), Simpson (0.64), and equitability (0.48) diversity indices showed that more species of mosquitoes were collected in the residential area than in the activity area. The low diversity of mosquito species observed in the activity area can be explained by the fact that in this area, breeding sites are present at a specific time of the year, which is not the case in the residential area. Results are in agreement with those of Konan et al.,[26] and Cornet et al.[27] Authors have indicated that sites with low culicid diversity do not offer favourable living conditions for culicid settlement, unlike sites with high diversity. *Aedes aegypti* accounted for 78.42% and 49.47% of mosquitoes collected in activity and residential areas respectively. The abundance of these species may be associated with the multiplicity of such breeding sites. Indeed, port activities create suitable breeding sites for *Aedes aegypti*.[28] According to Cornet et al.,[27] urbanisation leads to a variation in the composition of species of genera *Aedes* mosquitoes causing a dominance of *Aedes aegypti*. It was found by Bhadauriya et al.[6] that the proliferation of *Aedes aegypti* was related to the mismanagement of fire extinguisher buckets. The results agree with those of Konan et al.[10] and Yao et al.[11] at the port of Abidjan, Côte d'Ivoire where *Aedes aegypti* accounted for over 83% of the mosquito species collected. In both areas, 88.32% and 75.21% of *Aedes aegypti* collected at rest were outdoors, respectively. The exophilic behaviour of *Aedes aegypti* would be explained by the fact that mosquitoes seek out shady and moist areas for shelter during the day. Therefore, the behaviour will be associated with the availability of outdoor breeding sites.[29] These results are similar to those of Bagny.[30] That found *Aedes aegypti* to be exophilic in Réunion.

Conclusion

At the port of San Pedro, 14 species of mosquitoes, including 10 of the genus *Culex*, were identified during the entomological investigations. More species of mosquitoes were collected in the residential area as compared to the activity area. Also, species were continuously present in the residential area as compared to the activity area. *Aedes aegypti* accounted for over half of the specimens collected in both areas. The presence of this species constitutes an important threat, which is permanent in residential areas and periodic in activity areas. This study is one of the first of its kind at the port of San Pedro. It is useful for the development of control strategies and the establishment of adequate entomological surveillance in the port of San Pedro.

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Conflict of Interest: None

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