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▣ ORIGINAL ARTICLE

Comparative Analysis of Pathogenic Organisms in Cockroaches from Different Community Settings in Edo State, Nigeria

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Abstract: Cockroaches are abundant in Nigeria and are seen to harbour an array of pathogens. Environmental and sanitary conditions associated with demographic/socio-economic settings of an area could contribute to the prevalence of disease pathogens in cockroaches. A total of 246 cockroaches (*Periplaneta americana*) in urban (Benin, n=91), semi-urban (Ekpoma, n=75) and rural (Emuhi, n=70) settings in Edo State, Nigeria were collected within and around households. The external body surfaces and alimentary canal of these cockroaches were screened for bacterial, fungal, and parasitological infections. *Bacillus* sp. and *Escherichia coli* were the most common bacteria in cockroaches. However, *Enterococcus faecalis* could not be isolated in cockroaches trapped from Ekpoma and Emuhi. *Aspergillus niger* was the most prevalent fungus in Benin and Ekpoma, while *Mucor* sp. was predominant in Emuhi. Parasitological investigations revealed the preponderance of *Ascaris lumbricoides* in Benin and Emuhi, while *Trichuris trichura* was the most predominant in Ekpoma. The prevalence and burden of infection in cockroaches is likely to be a reflection of the sanitary conditions of these areas. Also, cockroaches in these areas making incursions in homes may increase the risk of human infections with these disease agents.

Key words: cockroach, pathogen, sanitary condition, Nigeria

INTRODUCTION

Cockroaches such as *Periplaneta americana* and *Blattela germanica* have filthy habits with an ability to spoil food, transfer pathogens, and cause allergic reactions and psychological distress [1]. Typical of an urban settlement in most developing countries is overcrowding, pollution, and lack of basic services such as clean water and sanitation. These conditions could enhance the infestation rate of cockroaches to a number of pathogens [2].

Over 100 species of bacteria have been isolated from domestic cockroaches [3] of which some could be potential mechanical transmitters of pathogenic bacteria [4]. A study in Osogbo, urban Nigeria, and Ekpoma, a semi-urban area revealed an array of bacteria harboured by cockroaches [5,6]. Similarly, fungal infections have been associated with both the external body parts and feces of cockroaches [5,6]. Protozoan parasites and gastro-intestinal helminths have also been reported in cockroaches collected from both suburban and peri-urban areas [5,7] being widely implicated as reservoir hosts of medically important parasites [8,9].

The distribution of bacteria, fungi, and parasites in cockroaches could differ from one location to the other based on both environmental and sanitary conditions which could be used to assess the likely infectious diseases that pose great risk as we consider the capacity of cockroaches in spreading diseases in a complementary manner. Hence, this paper reports a comparative analysis of the prevalence of pathogenic organisms present in cockroaches sampled from urban, semi-urban, and rural areas of Edo State, Nigeria.

MATERIALS AND METHODS

Study Area

This study was carried out in urban (Benin), Semi-urban (Ekpoma), and rural (Emuhi) areas in Edo State, Nigeria between February and July 2013. The capital of the state is Benin. Benin (61°9'N, 5°36'E) is a city approximately 25 miles north

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of the Benin River, and it is the center of Nigeria's rubber industry, with a traditional industry of palm nuts processing for oil production. Ekpoma (64°5'N, 60°8'E), a semi-urban settlement, is the headquarters of the Esan West Local Government Area and a host to a tertiary institution (Ambrose Alli University). It is mostly populated by the Esan people and students from various parts of Nigeria. Sanitary conditions in this area are almost underdeveloped. Fecal disposal and water supply are very elementary in the sense that the majority of the population relies on pit toilets for fecal disposal and hand-dug wells for water storage. Emuhi (60°40'N 60°10'E), a rural community [10] is also under the administration of Esan West Local Government Area. The community is popularly reputed in the region for the production of pineapple fruit. Fecal and waste disposal are done in crude manners due to the lack of modern toilet and waste disposal facilities.

Sample collection and analysis

A total of 246 adult cockroaches (*P. americana*) were trapped from different homes in Benin (n=91), Ekpoma (n =85), and Emuhi (n=70) for bacterial, fungal, and parasitological studies. Cockroaches were collected in different areas of individual homes using sweep net and insecticide spray from the parlours, bedrooms, verandas, kitchens, toilets, and bathrooms, and then stored in sterile universal containers and transported to the laboratories.

Pathogens on the external body of cockroaches were isolated by filling the containers with 5 ml of sterile normal saline and then shaken vigorously. Afterwards, a loopful of each suspension was cultured for bacteriological and fungal investigations. Furthermore, the alimentary canal of each cockroach was emptied in a different container of 5 ml of saline for another round of bacteriological and fungal identification. Plates with blood and McConkey agar were used for bacteria culture, while Sabouraud and potato dextrose agar were used for fungi culture. These plates were incubated at 37℃ for 24 hr or more. The organisms were identified using standard bacteriological and fungal techniques as described by Mackie and McCartney [11]. A 10-fold dilution was carried out on each suspension to determine the total viable counts per millilitre on each cockroach using the pour-plate method. These counts were made on plates showing discrete and evenly distributed colonies.

For parasitological studies, samples were transferred to a conical test tube and spun at 3,000 rpm for 5 min. The deposits were observed on clean glass slides and viewed under a microscope. The eggs of the parasites were identified and the number counted per ml. Results were entered in excel spread sheet, and InStat statistical package was used to analyze the data.

RESULTS

Table 1 shows the different pathogens isolated from both the external body surface and alimentary canals of cockroaches. A total of 246 cockroaches were examined, and it was demonstrated that more microbes and parasites were collected in the alimentary canal than the body surface of cockroaches.

A comparison of the distribution of bacteria in cockroaches collected from urban, semi-urban, and rural communities are shown in Table 2. In Benin, 10 species of bacteria were isolated, whereas fewer numbers were identified in Ekpoma (7 species) and Emuhi (8 species). Clearly, *Bacillus* sp. was the most prevalent bacterial infection with Emuhi community showing the highest infection rate in cockroaches (OR, 8.23; $P<0.05$).

Table 1. Pathogens isolated from the body surface and internal organs of cockroaches (n=246)

Bacteria		Fungus		Parasite (eggs)	
Body surface	Alimentary canal	Body surface	Alimentary canal	Body surface	Alimentary canal
Bacillus sp.	*Bacillus* sp.	*Aspergillus niger*	*Aspergillus niger*	*Ascaris lumbricoides*	*A. lumbricoides*
Escherichia coli	*E. coli*	-	*Candida* sp.	*Trichuris trichiura*	*T. trichiura*
Proteus mirabilis	*P. mirabilis*	*Mucor* sp.	*Mucor* sp.	-	*Enterobius vermicularis*
-	*Psuedomonas aeruginosa*	-	*Rhizopus*	-	*Schistosoma haematobium*
-	*Proteus vulgaris*	*Saccharomyces cerevisiae*	*S. cerevisiae*	*Coccidia*	-
Staphylococcus aureus	*Staphylococcus aureus*	-	*Fusarium* sp.	-	*Balantidium coli*
-	*Staphylococcus epidermis*	-	*Penicillium* sp.	*Entamoeba histolytica*	*E. histolytica*
-	*Enterococcus faecalis*				
Citrobacter freundi	-				
	Salmonella sp.				

Table 2. Prevalence and bacterial load in cockroaches from urban, semi-urban, and rural communities

Bacteria	Urban (Benin)		Semi-urban (Ekpoma)		Rural (Emuhi)	
	N (%)	Load (organisms/ml)	N (%)	Load (organisms/ml)	N (%)	Load (organisms/ml)
Bacillus sp.	47 (51.6)	8.0×10^9	38 (41.2)	9.0×10^9	56 (80.0)	3.1×10^9
Escherichia coli	7 (7.7)	10×10^9	13 (15.3)	3.3×10^7	12 (17.1)	5.2×10^9
Proteus mirabilis	10 (1.1)	4.0×10^9	4 (4.7)	5.2×10^9	2 (2.9)	3.8×10^9
Pseudomonas aeruginosa	7 (7.7)	6.0×10^9	4 (4.7)	3.4×10^9	7 (10.0)	3.5×10^9
Proteus vulgaris	1 (1.1)	11×10^9	1 (1.2)	2.4×10^6	-	-
Staphylococcus aureus	7 (7.7)	4.0×10^9	4 (4.7)	5.3×10^9	10 (14.3)	3.9×10^8
Staphylococcus epidermidis	2 (2.2)	3.4×10^9	-	-	1 (1.4)	3.0×10^9
Enterococcus faecalis	2 (2.2)	5.0×10^8	-	-	-	-
Citrobacter freundi	2 (2.2)	6.0×10^9	2 (2.4)	3.4×10^9	2 (2.9)	4.4×10^5
Salmonella sp.	2 (2.2)	3.0×10^9	-	-	1 (1.4)	4.9×10^5

Table 3. Prevalence and fungal load in cockroaches from urban, semi-urban, and rural communities

Fungi	Urban (Benin)		Semi-urban (Ekpoma)		Rural (Emuhi)	
	N (%)	Load (fungi/ml)	N (%)	Load (fungi/ml)	N (%)	Load (fungi/ml)
Aspergillus niger	30 (33.0)	5.2×10^9	21 (24.7)	4.3×10^9	4 (5.7)	7.8×10^9
Candida sp.	5 (5.5)	3.0×10^7	1 (1.2)	7.0×10^9	1 (1.4)	9.7×10^8
Mucor sp.	17 (18.7)	5.7×10^8	14 (16.5)	9.1×10^9	12 (17.1)	4.3×10^9
Rhizopus sp.	5 (5.5)	3.9×10^9	-	-	-	-
Saccharomyces cerevisiae	5 (5.5)	8.1×10^6	1 (1.2)	2.0×10^6	7 (10.0)	6.6×10^8
Fusarium sp.	3 (3.3)	4.0×10^9	3 (3.5)	5.2×10^9	2 (2.9)	4.5×10^9
Penicillium sp.	2 (2.2)	3.1×10^8	5 (5.9)	8.2×10^8	5 (7.1)	5.7×10^9

Table 4. Prevalence and parasitic load in cockroaches from urban, semi-urban, and rural communities

Parasites	Urban (Benin)		Semi-urban (Ekpoma)		Rural (Emuhi)	
	N (%)	Load (eggs/ml)	N (%)	Load (eggs/ml)	N (%)	Load (eggs/ml)
Ascaris lumbricoides	12 (13.2)	17	3 (3.5)	20	2 (2.9)	5
Trichuris trichiura	4 (4.4)	39	4 (4.7)	18	-	-
Enterobius vermicularis	3 (3.3)	30	1 (1.2)	4	1 (1.4)	6
Schistosoma haematobium	3 (3.3)	4	1 (1.2)	6	-	-
Coccidia	3 (3.3)	2	-	-	-	-
Balantidium coli	1 (1.1)	15	1 (1.2)	44	-	-
Entamoeba histolytica	2 (2.2)	4	1 (1.2)	3	-	-

Similarly, our data shows that *Escherichia coli* was the second most dominant bacterial isolate, with Emuhi recording the highest infection rate. *Enterococcus faecalis* could not be isolated in cockroaches from Ekpoma and Emuhi.

Table 3 summarizes a comparative data of fungal infections in cockroaches. Seven fungi were isolated in Benin, while 6 each for Ekpoma and Emuhi. Comparatively, *Aspergillus niger* had the highest prevalence in Benin and Ekpoma than other fungi except for the preponderance of *Mucor* sp. in cockroaches collected from Emuhi. *Rhizopus* sp. were seen in cockroaches from Benin only.

Parasites were isolated from cockroaches in urban, semi-urban, and rural settlements as presented in Table 4. Our record shows that *Ascaris lumbricoides* was predominant in the 3 areas and particularly highest in cockroaches from Benin (OR= 3.12; $P<0.05$). In addition, *Trichuris trichiura* was the most frequently occurring parasites in Ekpoma. We did not isolate *A. lumbricoides* and *Enterobius vermicularis* eggs in Emuhi, whereas coccidian parasites were recovered from Benin only.

DISCUSSION

This study isolated 10 species of bacteria from Benin; seven and 8 species from Ekpoma and Emuhi respectively. There was the predominance of *Bacillus* sp. and *E. coli* in the cockroaches collected from the 3 settlements. Also, the prevalence of *Staph-*

ylococcus aureus was higher in Emuhi than in the other areas. Additionally, *E. faecalis* was not isolated in cockroaches trapped from Ekpoma and Emuhi but was seen in Benin. The distribution of bacterial organisms in Ekpoma was not so different from the report of Tatfeng et al. [5] except that, in this work, *Bacillus* sp. was the most frequently seen microorganism in cockroaches which was not the case in the previous investigation from Ekpoma. *Bacillus* sp. and *E. coli* being harboured by cockroaches can cause diarrhea [12]. *S. aureus* causes nosocomial infections, and cockroaches have been implicated as carriers [13,14]. Generally, spread of *S. aureus* is through human-to-human contact, although recently some veterinarians discovered that the infection can spread through pets [15]. The presence of *E. faecalis* in Benin aligns with the assertion that this organism survives highly unfriendly environmental conditions which characterizes some urban cities in Nigeria [16].

A total of 7 fungal species were isolated from cockroaches in Benin, while 6 species were present in cockroaches collected in Ekpoma and Emuhi. The most prevalent species in the 3 areas were *A. niger* and *Mucor* sp. *A. niger* causes a disease called black mold on certain fruits and vegetables, and it is ubiquitous in soil. Being regarded as an opportunistic pathogen [17], *A. niger* has been associated with otomycosis infection among Nigerians [18]. *Mucor* is a microbial genus of approximately 3,000 species of moulds commonly found in soil, digestive systems, plant surfaces, and rotten vegetable matter. However, some *Mucor* sp. have been reported as infectious agents, although their inability to grow at temperatures above 32℃ raises doubt as to their validity as human pathogens, and their pathogenic role may be limited to cutaneous infections [19,20]. *Rhizopus* organism was not seen in Ekpoma and Emuhi but present in Benin. This could be attributed to the number of cockroaches analyzed against previous investigation of relatively larger sample size in Ekpoma where this fungus was isolated in cockroaches [5]. *Rhizopus* spp. are found on a wide variety of organic substrates while some are opportunistic agents of human zygomycosis.

More parasites were recovered in Benin in comparison with the other communities. Also, we observed that *A. lumbricoides* was the most occuring parasite across the sampled areas; and this is similar to the observation of Etim et al. [7]. This also corroborates previous reports regarding the epidemiology of intestinal helminths in Edo State where *A. lumbricoides* was recorded to be the most prevalent intestinal parasite across age groups [21,22]. *Schistosoma haematobium* ova were identified in a few cockroaches. We strongly suspect that some dwellers may be infected with this parasite which might have been picked up by these cockroaches probably in the toilet or bathroom. A study in Sokoto, Nigeria has also isolated *S. haematobium* eggs from cockroaches [23]. Furthermore, coccidian parasites were isolated in Benin only, an indication of cockroaches having contact with fecal droppings of birds, dogs, and/or cats. Poor fecal and garbage disposal systems observed in Benin City may have contributed in no small way to the parasitic contamination of these cockroaches.

The urban area of Benin had the highest burden of disease agents in cockroaches. This is quite instructive in the sense that it simply shows the sanitary situation of this area. Interestingly, this is contrary to our expectation considering the circumstances that rural dwellers are subjected to lack/inadequate basic infrastructural facilities which include waste disposal facilities. Consequently, it is assumed that these rural dwellers exhibit relatively poorer sanitary habits that predispose the environment to a higher proliferation of disease agents. Additionally, rural areas are being adjudged to be least developed socioeconomically, with its attendant challenges of non-provision of waste disposal facilities, resulting in the habit of indiscriminately disposing of waste materials leading inevitably to generation of pockets of litter-spots that obviously constitute breeding sites for most disease agents; and when cockroaches visit these sites, they pick up all kinds of pathogens. However, the results we have for Benin is strongly adduced to some urban challenges other authors have considered as risk factors to disease outbreaks [2,24]. Moreover, we observed that in Benin City, there is inadequate waste disposal facility and agency, and therefore wastes are allowed to remain for a long time at collection points before they are disposed of. We observed therefore that between the period when the waste bins are filled to capacity and the time of disposal, cockroaches feast within these exposed bins and pick up all manners of pathogens that might contaminate food materials, kitchen utensils and other household items through its activities of fecal droppings [25] or mere body contacts with these items, thereby posing great health risks to dwellers.

In conclusion, cockroaches in Edo State could be mechanical transmitters of medically important pathogens, and inhabitants of urban centres are at the greatest risk of infection. Our results also indicate that the burden of disease pathogens in cockroaches could be related to both environmental conditions and sanitary habits of dwellers. Since cockroaches col-

lected from a representation of urban, semi-urban, and rural communities had issues with disease pathogen infestation, it is strongly recommended that control measures should be greatly improved especially by driving adequate and effective institution of waste management systems in Edo State.

CONFLICT OF INTEREST

We declare that we have no conflict of interest related to this work.

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