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REGULAR ARTICLE

MICROBIAL LOAD AND MULTIPLE DRUG RESISTANCE OF PATHOGENIC BACTERIA ISOLATED FROM FEACES AND BODY SURFACES OF COCKROACHES IN AN URBAN AREA OF SOUTHWESTERN NIGERIA

Monsuru Adebayo Adeleke[*1], Hilda Abiola Akatah[1], AbdulWasiu Oladele Hassan[2], Wasiu Olalekan Adebimpe [3]*

[1] Public Health Entomology and Parasitology Unit, Department of Biological Sciences, P.M.B 4429, Osun State University, Osogbo, Nigeria

[2]Department of Microbiology and Parasitology, Ladoke Akintola University Teaching Hospital, Osogbo, Nigeria

[3]Department of Community Medicine, College of Health Sciences, Osun State University, Osogbo

*Corresponding author: healthbayom@yahoo.com.

ABSTRACT

This study investigates the microbial load and antibiotic susceptibility pattern of pathogenic bacteria isolated from the faeces and body surfaces of cockroaches in Osogbo, Southwestern Nigeria. The cockroaches collected from residential areas and hospital vicinities were screened for microbial load and antibiotic susceptibility pattern using standard protocols. A total of twenty- three microorganisms namely *Klebsiella aerogenes, Bacillius cereus, Proteus spp, Staphyloccocus aureus, S. saprophyticus, Enteroccocus faecalis, Staphylococus epididermis, E. coli, Listeria monoctogene, Proteus mirabilis, Citrobacter species, Pseudomonas aeruginosa,* Psuedomonas species, *Seretia mensence, Candida albicans, Candida spp., Aspergilius spp., A. flavus, A. fumigates, Mucor species and Penicilium species* were isolated. The microbial load of the microorganisms was significantly higher in the isolates from hospital as compared with the residential area ($p<0.05$) with the exception of *Canidida* species, *Mucor* and *Penicillium* which had higher or equal microbial load at the residential areas. All the pathogenic bacteria isolated had multiple resistance to antibiotics

most importantly, Ampicillin, Augumentin, Amoxicillin and Septrin (30µg). Efforts geared towards controlling the insects will be indispensable in curbing the wide spread of multi-drug resistant pathogens in the study area.

Keywords: cockroaches, microbial load, antimicrobial, multi-drug resistance, Nigeria

INTRODUCTION

Antimicrobial resistance of pathogenic microorganisms has assumed a worrisome dimension with recent trend of resistance of pathogenic bacteria to common antibiotics. This increase in antibiotic resistance was premised on the drug pressure as a result of abuse of common antibiotics by the users and uncomplimentary fake drugs in circulation **(Ehinmidu 2003, Tachebe *et al*., 2006, Oleghe *et al.*, 2011)**. The unwholesome behaviour has led to the genetic response of the microorganisms to microbial therapy which has now become an issue mitigating the control of pathogenic microorganisms in different parts of the world **(Oleghe *et al.*, 2011).**

It has most often been assumed that the drug resistance in clinical isolates usually results from the contamination of resistant bacteria from the drug pressurized environment **(Oleghe *et al.,* 2011).** The bacteria contaminant could be from water, food or contact with the vectors harbouring the pathogens. Cockroaches stay in filthy environments in the house, shops and even hospitals where both clinical and environmental samples coincide **(Fortedor *et al*., 1992)**. Therefore, their roles in promoting drug resistance in pathogenic microorganisms cannot be overlooked.

Though, previous studies have implicated cockroaches as potential carriers of microorganisms and drug resistant microbes in different parts of the world **(Cloarec *et al*., 1992; Fortedor *et al*., 1999; Padro *et al*., 2002; Tatfeng *et al*., 2005; Tachebe *et al*., 2006)** there was little or no information on anti-microbial susceptibility status of the micro-organisms harboured by the cockroaches in Osogbo in particular and Nigeria in general.

The objective of the present study was to investigate the microbial load and anti-microbial susceptibility of pathogenic bacteria isolated from the feaces and cockroaches in Osogbo, Southwestern Nigeria.

MATERIAL AND METHODS

Collection of cockroaches

The study was carried out in Osogbo, Osun State Nigeria. Osogbo lies latitude of $7^{o}49^{'}$N and a longitude of $4^{o}37^{'}$E. The faeces and body surfaces of cockroaches collected from four randomly selected residential areas and two hospital vicinities; Ladoke Akintola University Teaching Hospital Osogbo and State Hospital Asubiaro between November 2011 and February 2012 were screened for microbial load and susceptibility pattern to antibiotics. The cockroaches were trapped with sterile hand gloves and transferred to sterile universal containers. The cockroaches were kept in the bottles until they defeacate. The cockroaches and the feaces were then transferred to separate sterile universal bottle for analysis.

Screening for pathogenic organisms

The cockroaches and the feaces were kept in the universal containers and 2ml of sterile normal saline (0.9%) was added to the universal containers and vigorously shaken for 2 minutes. 0.01ml of the sample was then taken from each container and cultured on the MacConkey , Sabouraud's dextrose agar and chocolate agar plate and incubated overnight at 37^{o}C.

Identification of bacteria

The colonies were identified by standard bacteriological procedures; macrosopic morphology, biochemical and gram staining in accordance with **Cowan and Steel (1975)** .

Gram stain

Gram's stain was performed to determine if the organism is gram negative or gram positive. The staining was performed on the isolates according to the known procedures. A smear of the test organism was made on a clean slide, dried and covered with crystal violet for 30-60 seconds. It was washed off with clean water and covered with Lugol's iodine for 30-60 seconds and washed off with clean water. The slide was decolourized with acetone-alcohol, and washed immediately with clean water and covered again with neutral red stain for 2

minutes, and washed off with clean water.The back of the slide was wiped clean and placed in a draining rack for the smear to air dry. The slide was examined microscopically with the oil immersion lens after the application of the oil on the slide. Gram positive bacteria gave a dark purple colour while gram negatives give a red colour.

Biochemical tests

Several biochemical tests were performed on the isolates for identification purposes as described by **Baron and Finegold (1990)**. The catalase test was done to differentiate the bacteria that produce the enzyme catalase, such as staphylococci from the non-catalase producing bacteria such as streptococci. Citrate utilization test was done to identify enterobacteria, the test is based on the ability to use citrate as its only source of carbon. Using Simmon's citrate agar, slopes of the medium was prepared in bijou bottles as recommended by the manufacturer. With the aid of a sterile straight wire, the slope was streaked with a saline suspension of the test organism, and the butt was stabbed.
It was incubated at 35^{o}C for 48hours. A bright blue colour in the medium indicates a positive citrate test (e.g *Klebsiella pneumoniae*), while no change in the colour of the medium gives a negative citrate test (e.g *E.coli*). Coagulase test was done to identify *S.aureus* which produces the enzyme coagulase. A drop of distilled water was placed at the end of a slide, a colony of the test organism is emulsified on it to make a suspension. A loopful of plasma was added to it and mixed gently, presence of clumping within 10 seconds indicates the presence of *S. aureus*, while absence indicates presence of *E.coli* or *S.epidermidis*. Oxidase test was used to identify *Pseudomonas, Neisseria, Proteus, Brucella* and *Pasteurella* species. A piece of filter paper was placed in a clean sterile petri dish, and 2 or 3 drops of freshly prepared oxidase reagent was added to it. Using a piece of stick, a colony of the test organism was smeared on the filter paper. A blue-purple colour within 10 seconds shows an oxidase positive test. Urease test was done to differentiate enterobacteria, e.g *Proteus* strains **(Chaichanawongsaroj *et al*.,2004)**. The test organism is inoculated in a bijou bottle containing 3ml sterile Cristensen's modified urea broth and incubated at 35-37^{o}C for 3-12hours. A pink colour in the medium gives a positive test result **(Baron and Finegold, 1990).** Other biochemical tests for detection and identification of various types of bacteria encountered in this study include; carbohydrate utilization tests (sugar test). - lactose , sucrose , mannitol , maltose , xylose and dextrose, indole test, methyl red test ,voges proskuer test, triple sugar iron test, lead acetate test, mannitol motility test, oxidation - fermentation test, and amino acid degradation test. The

enterobacteriaecae group and other group of bacilli were identified in accordance with their characteristics by comparing with standard table **(Chaichanawongsaroj *et al*.,2004).**

Fungi identification

The fungi isolates were identified by microscopic examination of the actively growing mould using morphological characters such as, the absence or presence rhizoid, colour, and micro-morphology of their sporulating structures and conida **(Evans and Richrdson, 1989**; **Onions, *et al*., 1991).**

Total viable count

A ten-fold dilution was carried out on each suspension to determine the total viable count of each cockroach using the pour plate method counts were made on plates showing discrete colonies. A quantitative analysis of bacteria was calculated as described by **Salehzadeh *et al*., (2007)**. The overall load of bacteria carried by each insect was counted and expressed as colony forming unit (c.f.u).

Antibiotic sensitivities of isolated pathogenic bacteria

The Bauer-Kirby procedure was performed on the identified isolates using nutrient agar plate and antibiotic discs containing chloramphenicol, 30µg; septrin, 30µg; sparfloxacin, 10 µg; ciprofloxacin, 10 µg; amoxacillin, 30 µg; augmentin, 30ug;streptomycin, 30 µg; gentamicin 10 µg; pefloxacin, 30 µg; ofloxacin 10 µg; ; ampicillin/ cloxacillin, 30µg; ofloxacin, 5 µg; erythromycin, 10µg;gentamicin, 10 µg; ciprofloxacin, 5 µg; clindamycin, 10 µg; cephalexin,30 µg; Flucloxacillin 30 µg; augumentin, 30 µg; and Septrin, 50µg. Inhibition diameters were measured and the zone of inhibition generated by each antibiotic disc was grouped susceptible or resistance by comparing the measured diameter with the standard given in the manufacturer's instruction. These antibiotics were tested with 0.1ml of 0.5McFaland standard of overnight pure culture of *E.coli ATCC 2592* as control organism for the sensitivity as described by Bauer-Kirby **(Thomas *et al*., 2012).**

Statistical Analysis

The comparison of data obtained from the feaces and body surface of the cockroaches were analyzed with t-test using SPSS version 16.0.

RESULTS

The results of the microbial analysis of the feaces and body surfaces of the cockroaches revealed that the body surface of all the cockroaches caught at both hospitals and residential areas were positive for microorganisms while 83.6% and 95.5% of the feaces from residential areas and hospitals were positive respectively (Table 1). The variation in the occurrence of the microorganisms between body surface and feaces was not significant ($p>0.05$). A total of twenty- three microorganisms namely *Klebsiella aerogenes, Bacillius cereus, Proteus spp, Staphyloccocus aureus, S. saprophyticus, Enteroccocus faecalis, Staphylococus epididermis, E. coli, Listeria monoctogene, Proteus mirabilis, Citrobacter species, Pseudomonas aeruginosa,* Psuedomonas species, *Seretia mensence, Candida albicans, candida spp, Candida spps, Aspergilius spp, A. flavus, A. fumigates, A, Mucor species and Penicilium species* were isolated. All the microbial isolates were found in the body surface of the cockroaches while ten of the twenty three isolates were found in the feaces of cockroaches (Table 2). The microbial load of the microorganisms was significantly higher in the isolates from hospital as compared with the residential area ($p<0.05$) with the exception of *Canidida* species, *Mucor* and *Penicillium* which had higher or equal microbial load at the residential areas (Table 3).

Tables 4 and 5 present the results of susceptibility of the isolates to the antibiotics. The results revealed that all the isolates from residential area were resistant to more than two antibiotics with the exception of *Proteus* species. Moreover, virtually all the isolates were resistant to Ampicillin while majority were susceptible to Streptomycin. However, all the isolates from hospital vicinity had multiple antibiotic resistance and their susceptibility varies with different antibiotics. Majority of the isolates were resistant to Augumentin, Amoxicillin and Septrin (30 µg).

Table 1 Occurrence of the microorganisms in the faecal pellets and body surfaces of the cockroaches at the study area

Sources	No of cockroaches examined		No positive (%)	
	Feaces	Body surface	Feaces	Body surface
Residential	55	55	46 (83.6)	55(100)
Hospital	45	45	43 (95.5)	45 (100)

Table 2 Microbial diversity in the body surfaces and feacal pellets of the cockroaches at the study area

Name of Isolates	Feacal pellets	Cockroach surface
Bacteria isolates		
Baccillus species	+	+
Klebsiella aerogenes	+	+
Proteus species	-	+
Staphyloccocus aureus	+	+
Baccillus cereus	-	+
Staphyloccocus saprophyticus	-	+
Enteroccocus feacalis	+	+
Staphylococus epididermis	+	+
Escherichia coli	+	+
Listeria monoctogenes	+	+
Proteus mirabilis	-	+
Citrobacter species	-	+
Psuedomonas species	-	+
Psuedomonas aeruginosa	-	+
Seretia mensence	-	+
Fungi isolates		
Candida Spcies	+	+
Candida albicans	+	+
Aspergillus flavus	-	+

Aspergillus fumigates	-	+
Aspergillus species	+	+
Mucor Species	-	+
Penicillum species	-	+

- Means absent
+ Means present

Table 3 Microbial load of bacteria and fungi encountered on the body surfaces and feacal pellets of the cockcroaches

Organisms	**Residential**		**Hospital**	
	No of isolates	**Mean load**	**No of isolates**	**Mean load**
Bacillius cereus	11	12.5×10^3	6	15.5×10^9
E. coli	4	12.5×10^6	2	12.5×10^7
Pseudomonas spp	1	1.0×10^4	8	12.5×10^7
Canidida spp	5	12.5×10^{10}	3	12.5×10^9
Aspergillus	1	1.2×10^2	2	1.2×10^3
Mucor	2	1.0×10^1	Not found	
Penicillum	1	1.2×10^2	1	1.2×10^2
Aerobic spore bearer	24	15.58×10^{10}	28	15.7×10^{11}
Total coliforms	10	12.0×10^7	21	12.0×10^9
Standard plate count	55	12.5×10^5	68	12.5×10^6

Table 4 Antibiograms of pathogenic bacteria isolated from cockroaches trapped at the residential areas

Bacteria	**Gram positive Bacteria Isolates**	
	Sensitive	**Resistant**
Bacillus spp	SP, OF, E, AM, CN, AU, CPX, PEF	S, CO, CH
S.aureus	OF, CIP, CN, , E	FX, AP, CO, AU
Enterococcus faecalis	OF, CIP, AU, CN,	FX, CO, AP, PEF
Bacillus cereus	CIP, GN, OF	E, CX, CO, FX, AP, AU
S. saprophyticus	CIP, CN, OF, E, AU	FX, CO, AP
S. epidermidis	CIP, OF, AU, E, CN	CO, AP, FX, CX
Aerobic spore bearer	GN, CIP, AP	AU, E, CO, FX, AP, CD
Listeria monocytogenes	E, CIP, GN	CX, CO, CD, AP
	Gram Negative Bacteria Isolates	
Proteus mirabilis	OFX, AU, CN, SP, CPX, AM	S, CO
Klebsiella spp	CO, AU, CIP, AM	OF, E, AP
Klebsiella aerogenes	CO, AU, CIP, AM	OF, E, AP
E. coli	OF, CIP, GEN	AU, CO, CD, CX, AP, FX
Klebsiella spp	CPX, SP, CO, S, OFX, PEF	CN, AU, CH, AM
Citrobacter spp	CPX, SP, PEF, OFX	AM, AU, CN,S, CO, CH
Pseudomonas aeruginosa	PEF, OFX, CPX	S, CO, AM, AU, CH

OF, Ofloxacin (5µg); E, Erythromycin; CIP/CPX, Ciprofloxacin; CD, Clindamycin; GN/CN, Gentamicin; CX, Cephalexin; CO, Septrin (50 µg); AP, Ampicillin/Cloxacillin; FX, Flucloxacillin; AU, Augumentin; SXT Septrin (30 µg); CH, chloramphenicol; SP, sparfloxacin; AM, Amoxicillin; PEF, perfloxacin; OFX, Ofloxacin (10 µg), S, Streptomycin.

Table 5 Antibiograms of pathogenic bacteria isolated from cockroackes trapped at the hospital vicinity

Bacteria	**Gram Positive isolates**	
	Sensitive	**Resistant**
Serratia marcescens	CPX, SP, PEF, OFX, SP	CN, SXT, CN, AU, AM, CH
S. saprophyticus	PEF, OFX, S, SP, CPX	AU, CN, AM, SXT, CH
S.aureus	CN, PEF, OFX,S, SP, CPX	SXT, AM, AU,CH
Bacillus cereus	SP, OFX, PEF, CPX	S, AU, AM, SXT, CH, CN
Enterococcus faecalis	OF, E, GN, CD, CX	FX, AP, AU
Listeria monocytogenes	E, CIP, GN	CX, CO, FX, AP, CD
	Gram Negative isolates	
Klebsiella spp	CN, PEF, OFX, CH, SP, CPX	AM, AU,SXT, S
Klebsiella aerogenes	S, CN,PEF, CPX	SXT, CH, AM, AU
Proteus spp	SP, CPX, AM, CN, PEF, OFX	AU, S, SXT, CH
E.coli	PEF, OFX, S, SP, CPX	AU, CN, AM, SXT, CH
Pseudomonas aeruginosa	PEF, OFX, CPX	S, SXT, AM, AU, CH
Citrobacter spp	CPX, SP, PEF, OFX	AM, AU, CN, S, SXT, CH

OF, Ofloxacin (5µg); E, Erythromycin; CIP/CPX, Ciprofloxacin; CD, Clindamycin; GN/CN, Gentamicin; CX, Cephalexin; CO, Septrin (50 µg); AP, Ampicillin/Cloxacillin; FX, Flucloxacillin; AU, Augumentin; SXT Septrin (30 µg); CH, chloramphenicol; SP, sparfloxacin; AM, Amoxicillin; PEF, perfloxacin; OFX, Ofloxacin (10 µg), S, Streptomycin.

DISCUSSION

The present results clearly indicated that nearly all the cockroaches in residential areas and hospital vicinity harboured pathogenic microorganisms. This high prevalence of the microorganisms harboured in the body and feaces of the cockroaches portends public health risks to the people in residential areas and transmission of nosocomial infections in the hospitals at the study area. Most of the bacterial isolated from the cockroaches in the present study are highly pathogenic and have been implicated in many nososocomial and gastroenteric infections (**Tatfeg *et al*., 2005, Lamiaa *et al*., 2010).** The isolation of *Candida species* from the cockroaches signifies the contact of the cockroaches with blood stream

infections **(Salehzadeh *et al.*, 2007)**. The isolation of such fungi from cockroaches in hospital is alarming and could worsen the infection morbidity and mortality in the hospitals, most especially for immune compromised patients who are already overwhelmed by infections **(Hong e*t al.*, 2003)**.

Moreover, the isolation of *A. flavus* from cockroaches in residential areas poses threat when considering the public health importance of this species. *A. flavus* has been known to produce mycotoxins which is one of the leading causes of food poisoning **(Tatfeng *et al.*, 2005; Salehzadeh *et al* 2007)**. The significant higher distribution of microorganisms and the microbial load in cockroaches from hospitals in comparison with residential areas is in consonance with the previous studies **(Fortedor *et al.*, 1999; Salehzadeh *et al* 2007)**.

Multiple drug resistance patterns were observed in all the isolates from hospitals and residential areas with the exception of *Proteus* species which showed double resistance in residential area. This observation contradicts the results of **Salehzadeh *et al* (2007)** but agrees with report of **Fortedor *et al.*, (1999)**. Though, most of the previous studies have reported the role of cockroaches as vectors of multi-drug resistant bacteria **(Fortedor *et al.*, 1999, Padro *et al.*, 2002)**, our results in the present study also showed multiple drug resistance in isolates from residential areas. This possibly introduces new dimension to the episode of drug resistant pathogens at the study area, as wide spread and contamination of isolates earlier susceptible to antibiotics could be observed within the shortest time in the metropolis. The above speculation is possible when considering the clustering plan of the residential areas in the metropolis and the unrestricted mode of movement of cockroaches at night **(Pai *et al.*, 2003; Pai *et al.*, 2004; Gracyk *et al.*, 2005; Pai *et al.*, 2005)**

The high prevalence of multi-drug resistant pathogens isolated from the cockroaches in residential areas in the present study may be pointing to the fact there has been a prolong drug pressure/ abuse of most of these antibiotics by the residents. Most of the antibiotics which had multiple resistance patterns in the isolates (such as Ampicillin, Augumentin, Amoxicillin and Septrin) were the first choice of antibiotic drugs in Nigeria probably due to the fact that they are cheap **(Ehinmidu, 2003).** These drugs would have been seriously abused by the people due to self-medication and over-dose. In Nigeria, the antibiotic resistance in pathogenic organisms has been reported to be plasmid mediated **(Oleghe *et al.*, 2011)**. While it is acknowledged that the focus of the this study is not geared towards investigating the mechanisms of resistance in the isolates, further studies concentrating on the mechanisms of the resistance may be of valuable information in understanding the origin of resistance observed in the isolates most importantly in the residential areas.

CONCLUSION

Our results showed high prevalence of pathogenic organisms in the body and feaces of cockroaches in Osogbo and reported the multiple resistance of the pathogenic bacteria to antibiotics. The multiple resistance of the isolates most importantly, the isolates from residential areas showed that the surveillance on pattern and origin of antimicrobial drug resistance should not be limited to only clinical isolates. It is therefore pertinent to educate the people at the resident areas on the danger inherent in harbouring the cockroaches in residential areas and hospital vicinities at the study area.

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REFERENCES

BARON, E. J. – FINEGOLD, S. M. 1990. Bailey and Scott's diagnostic microbiology, 7th ed., In St. Louis: New York 1990, p. 323 – 861.

CHAICHANAWONGSAROJ, N. – VANICHAYATANARAK, K. – PIPATKULLACHAT, T. – POLROJPANYA, M. – SOMKIATCHAROEN, S. 2004. Isolation of gram-negative bacteria from cockroaches trapped from urban environment. In *Southeast Asian Journal of Tropical Medicine and Public Health,* vol. 35, 2004, no.1, p. 681-684.

CLOAREC, A. – RIVAULT, G. – FONTAINE, F. – LEGUYADER, A. 1992. Cockroaches as carriers of bacteria in multifamily dwellings. In *Epidemiology and Infection*, Vol.109, 1992, p. 483–490.

COWAN, S. T – STEEL, J. L. 1975. Manual for the identification of Medical Bacteria, 2nd Ed., In Cambridge University Press, England 1975, p. 45-114.

EHINMIDU, J.O. 2003. Antibiotics susceptibility patterns of urine bacteria isolates in Zaria, Nigeria. In *Tropical Journal of Pharmaceutical Research,* vol. 2, 2003, no.2, p. 223-228

EVANS, E. G. V. – RICHRDSON, M. D. 1989. Medical mycology: a practical approach. Oxford University Press: Oxford 1989, 154p.

FORTEDOR, R. – BAREIJER, U. – VERMA, A. 1991. Cockroaches (*Blattella germanica*) as carriers of micro-organisms of medical importance in hospitals. In *Epidemiology and Infection,* vol. 107, 1991, p. 181-187

GRACZYK, T. K. – KNIGHT, R. – TAMANG, L. 2005. Mechanical transmission of human

protozoan parasites by insects. In *Clinical Microbiology Reviews*, vol. 35, 2005, p. 128–132.

HONG, W. – WEN, H. – LIAO, W. 2003. Fungal infection in organ transplant patients. In *Chinise Medecal Journal,* vol. 116, 2003, p. 1421–1425.

LAMIAA, B. – ANTONIO, S. – AMIN, L. – LEBBADI, M. – AARAB, A. – GUTIERREZ, J. 2010. Antibiotic resistance patterns of bacterial strains isolated from *Periplaneta americana* and *Musca domestica* in Tangier, Morocco. In *Journal of Infection in Developing Countries*, vol. 4, 2010, no. 4, p. 194-201.

OLEGHE, P. O. – ODIMEGWU, D. C. – UDOFIA, E. – ESIMORE, C.O. 2011. Multidrug resistant bacteria isolates recovered from herbal medicinal preparations in Southeastern Setting, Nigeria**.** In *Journal of Rural and Tropical Public Health,* vol. 10, 2011, p. 70-75

ONIONS, A. H. S – ALLSOPP, A. – EGGINS, H. O. W. 1981. Introduction to industrial mycology, Edward Arnold: London 1981 , 389p.

PAI, H. H – KO, Y. C. – CHEN, E. R. 2003. Cockroaches (*Periplaneta americana* and *Blattella germanica*) as potential mechanical disseminators of *Entamoeba histolytica*. In *Acta Tropica,* vol.87, 2003, p. 355–359.

PAI, H. H. – CHEN, W. C. – PENG, C. F. 2004. Cockroaches as potential vectors of nosocomial infections. In *Infection Control and Hospital Epidemiology,* vol. 25, 2004, p. 979–984.

PAI, H. H. – CHEN, W. C. – PENG, C. F. 2005. Isolation of bacteria with antibiotic resistance from household cockroaches (*Periplaneta americana* and *Blattella germanica*). In *Acta Tropica*, vol. 93, 2005, 259–265.

PRADO, M. A. – PIMENTA, F .C. – HAYASHID, M. – SOUZA, P. R. – PEREIRA, M.S. – GIR, E. 2002. Enterobacteria isolated from cockroaches (*Periplaneta americana*) captured in a Brazilian Hospital. In *Pan Americana Journal of Public Health,* vol. 11, 2002, p. 93–98.

SALEHZADEH, A. – TAVACOL, P. – MAJHUB, H. 2007. Bacteria, fungi and Parasitic contaminants of cockroaches in Public Hospitals of Hamedah, Iran, In *Journal of Vector Borne Dis*eases, vol. 44, 2007, p. 105-110

TACHEBE, J. – ERKU, W. – GERBE-MICHAEL, T. – ASHENAFI, M. 2006. Cockroach-associated food-borne bacteria from hospital and restaurant in Addis Ababa, Ethopia: Distribution and antibiograms. In *Journal of Rural and Tropical Public Health,* vol. 5, 2006, p. 34-41

TATFEG, Y. M. – USUANLE, A. – ORUKPE, A. – DIGBAN, A. K. – OKODUA, A. – OVIASOGIE, F. – TURRAY, A. A. 2005. Mechanical transmission of pathogenic organisms: the role of cockroaches, In *Journal of Vector Borne Diseases*, vol. 42, 2005, p. 129-134

THOMAS, B. T. – EFFEDUA, H. I. – MUSA, O. S. – ADEYEMI, M. T. – ADESOGA, K. O. – OGUNDERO, O. – OLUWADUN, A. 2012. Enumeration of Microorganism in Dried cassava Powder (Garri); a Comparative Study of Four Methods. In *New York Science Journal*, vol.5, 2012, no. 1, p. 1-7.