

PARASITES AND PATHOGENIC BACTERIA ASSOCIATED WITH HOUSEFLIES AND THE PUBLIC HEALTH IMPLICATIONS IN OSOGBO, SOUTHWESTERN NIGERIA

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[Adeleke, M. A., Oyewole, V. O., Olabiye, K. O. & Oforka, L. C. 2017. Parasites and pathogenic bacteria associated with houseflies and the public health implications in Osogbo, Southwestern Nigeria. *Munis Entomology & Zoology*, 12 (1): 94-98]

ABSTRACT: Houseflies are known as nuisance pests due to their ubiquitous nature in the environment. The present study investigates the roles of houseflies as mechanical vectors of parasites and bacteria in Osogbo, Southwestern Nigeria. The body parts of the houseflies collected from slaughter slabs, dumping grounds and palm wine spots were examined for parasitic cysts and bacterial isolates. The bacteria were isolated using standard bacteriological procedures and the parasites were observed microscopically. The bacteria isolates were thereafter subjected to antibiotic sensitivity test using standard protocol. All the seventy (70) samples examined harbored pathogenic bacteria. The bacteria isolated include, *Salmonella typhi*, *Klebsiella pneumonia*, *Proteus vulgaris*, *Escherichia coli*, *Proteus mirabilis*, *Providencia species.*, *Enterobacteraerogenes*, *Pseudomonas aeruginosa*, *Morganella morganii*, *Vibrio cholera*, *Shigella species* and *Serratia marcescens*. Even though there was variation in diversity of the bacteria across the study locations, the difference was not statistically significant ($p > 0.05$). The cyst of *Entamoeba histolytica* and eggs of *Ascaris lumbricoides* were also recovered from few houseflies. Most of the bacteria isolates showed multiple resistance to antibiotics such as ampicillin, Cefuroxime and Ceftazidime. The isolation of these multi-drug resistance bacteria and cysts of pathogenic parasites underscore the need for constant health education on promotion of environmental sanitation in the study area.

KEY WORDS: Houseflies, bacteria, parasite cysts, multidrug resistance, Osogbo, Nigeria

Houseflies (*Musca domestica*) are the most ubiquitous insects widely distributed all over the world, but more adaptable in tropical areas (Goulson et al., 2005). Houseflies are synanthropic animals, causing serious nuisance (Subejo, 2010; Howard, 2011). Housefly could be found in filth areas, therefore serving as potential mechanical vectors to parasites and bacteria. This potential underscores their public health importance to man and farm animals (Service, 1980).

Outbreak and cases of food-borne diarrheal diseases in urban and rural areas have been associated with houseflies' abundance in tropical areas, mostly in areas with poor sanitation (Gehad & Elsherbini, 2010). Houseflies have been reported to serve as mechanical vectors of enteropathogens, which serve as causative agents of gastro-intestinal disorders to humans (Otronto & Tarsitano, 2003). Graczyk et al. (2001) reported that cysts and eggs of human parasitic protozoans and helminthes could be recovered from external body, faecal deposition and regurgitation of the houseflies.

Evidence abound that the Nigerian environment is characterized by filth, thus promoting prolific breeding of the insects such as cockroaches and houseflies (Tatfeng et al., 2005; Adeleke et al., 2012). Adeleke et al. (2012) reported the roles of cockroaches as mechanical vectors of pathogenic bacteria in Osogbo

metropolis. Up to this moment little is known on the roles of houseflies as mechanical vectors of pathogens in the metropolis. Based on this existing research gap, the present study was designed to determine the roles of houseflies in the transmission of pathogenic parasites and bacteria in Osogbo, Southwestern Nigeria.

MATERIALS AND METHODS

Study area: The study was conducted in Osogbo, the state capital of Osun State. Osogbo lies between longitude 4° 34'E and latitude 7° 46'N. The town's land mass is approximately 47 km² with a population of 156,694.

Sample collection and study sites

The study sites were categorized into three; the slaughter slabs, dumping grounds and palm wine spots. The three collection sites were duplicated, to cover different parts of Osogbo. The housefly samples were collected at different intervals, but all in the day time between the hours of 13:00 and 16:00 using sweep nets. The samples were recovered into sterile universal bottles and transported to the laboratory for further analysis.

Bacteriological examination of the samples

The house flies were kept in the universal bottles and 2ml of sterile normal saline was added to the bottles and shaken vigorously for 5 minutes and left for some minutes to dislodge debris associated with the house flies. 0.01ml of the sample was taken from each container with the use of sterile wire loop and cultured on the MacConkey agar and incubated for 24hours at 37°C. The organisms that grew on the plates were subcultured on the nutrient agar for 24hours at 37°C to obtain the pure culture for characterization. The bacteria were identified using standard microbial procedures which include macroscopic morphology, gram staining and biochemical tests as described by Baron & Finegold (1990).

Antibiotic susceptibility test

The antibiotic sensitivity test was carried out using the nutrient agar plates and antibiotic discs containing Ceftriaxone, Cephalexin, Ampicillin, Ceftazidime, Cefuroxime, Gentamycin, Ciprofloxacin, Ofloxacin, Amoxycilin Andnitrofuratoin. Inhibition diameters were measured and the zone of inhibition generated by each antibiotic disc was grouped as susceptible, resistant and intermediate by comparing the measured diameter with the manufacturer's instruction.

Parasitological analysis

2ml of normal saline was added to the universal bottles containing the houseflies. The bottles were shaken vigorously for 5 minutes. 1ml of each sample was transferred onto a clean, sterile glass slide and viewed microscopically with the 40X objective lens.

Statistical analysis

The distribution of the bacteria was expressed in percentage. The diversity of the isolates in the study locations was subjected to t-test and chi-square analysis using SPSS version 17.0

RESULTS

A total number of seventy (70) housefly samples were collected. Twenty (20) samples were collected from slaughter slabs, twenty (20) from Dumping grounds and thirty (30) from palm wine spots. The results showed that all the house flies captured from the three locations harbored microorganism. Twelve microbial

organisms were isolated namely; *Salmonella typhii*, *Klebsiella pneumonia*, *Proteus vulgaris*, *Escherichia coli*, *Proteus mirabilis*, *Providencia* spp., *Enterobacter aerogenes*, *Pseudomonas aeruginosa*, *Morganella morganii*, *Vibrio cholera*, *Shigella* spp. and *Serratia marcescens* were isolated. Even though there was variation in diversity of the isolates across the study locations, the difference was not statistically significant ($p > 0.05$) (Table 1). *S. typhii*, *K. pneumonia*, *P. vulgaris*, *E. coli*, *P. mirabilis*, *Providencia* spp. and *Enterobacter* spp. were isolated from slaughter slabs while *E. coli*, *Enterobacter* spp., *P. aeruginosa*, *M. morganii* and *V. cholera* were isolated from dumping grounds. *K. pneumonia*, *E. coli*, *Enterobacter* spp., *M. morganii*, *V. cholera* were isolated from palm wine spot.

All the isolates were susceptible to more than three (3) antibiotics, but widely resistant to Ampicillin, Ceftazidime, Cefuroxime, Amoxicillin and cephalexin. However, only one isolate was found susceptible to Ceftriaxone from the samples isolated from palm wine spot; three (3) of the organisms isolated from the dumping grounds were susceptible to Nitrofurantoin while four (4) of the organisms isolated from slaughter slabs were susceptible to Nitrofurantoin (Tables 2-4).

The parasitological analysis showed that the houseflies harboured the cyst of *Entamoeba histolytica* and *Ascaris lumbricoides* eggs (Table 5). The cyst of *E. histolytica* was found in the three study locations.

DISCUSSION

The results showed that houseflies samples collected in the present study harboured considerable number of pathogenic bacteria and parasites. This shows that house flies constitute serious public health threats in Osogbo metropolis aside their nuisance nature. All the organisms isolated during this study are of medical importance and have been implicated in many gastrointestinal and gastroenteritis disorders (Graczyk et al., 2001; Otronto & Tarsitano, 2003).

The implication of status of houseflies in transmission of helminthic eggs and protozoan cysts is alarming since the houseflies are known to live in close association with human beings. *A. lumbricoides* and *E. histolytica* are gastrointestinal parasites which usually cause chronic diarrhea, liver complications and stunted growth in the affected people (Mbanugo & Abazie, 2002; Montessoro et al., 2002; Sam-Wobo et al., 2006; Tatfeg et al., 2005; Anosike et al., 2006; WHO, 2008).

The most commonly isolated bacteria in the present study are *V. cholera*, *Enterobacter* spp. and *Escherichia coli*. *V. cholera* cause the deadly disease known as cholera, and its preponderance at palmwine spots and dumping grounds is worrisome, and this may predispose people to cholera. *E. coli* is a major cause of gastrointestinal infections with acute diarrhea (Getachew et al., 2007).

Most of the isolates were resistant to antibiotics mostly Ampicillin and Amoxicillin. These antibiotics are first choice of antibiotics administered in Nigeria probably due to the fact that they are cheap (Ehinmidu, 2003). The antibiotics resistance of pathogenic microorganisms has been reported to be plasmid mediated (Oleghe et al., 2011). There is therefore need for further studies to determine the mechanisms of resistance in the isolates.

In conclusion, the results obtained in the present study implicated houseflies as potential mechanical vectors of pathogenic bacteria and parasites in Osogbo metropolis. There is therefore an urgent need for public health education and

enforcement of sanitation laws among the food vendors and slaughtering houses in Osogbo metropolis.

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Table 1. Bacteria isolates from the body of houseflies in different study locations in Osogbo, Nigeria.

NAME OF ORGANISM	SLAUGHTER SLABS	DUMPING GROUNDS	PALM WINE SPOTS
<i>Salmonella typhi</i>	+	-	+
<i>Klebsiella pneumonia</i>	+	-	+
<i>Proteus vulgaris</i>	+	-	-
<i>Escherichia coli</i>	+	+	+
<i>Proteus mirabilis</i>	+	-	-
<i>Providencia sp.</i>	+	-	-
<i>Enterobacter sp.</i>	+	+	+
<i>Pseudomonas aeruginosa</i>	+	+	-
<i>Morganellamorganii</i>	-	+	+
<i>Vibrio cholera</i>	-	+	+
<i>Shigella sp.</i>	-	-	+
<i>Serratiamarcescens</i>	-	-	+

Legend: + Means presence of bacteria; - Means absence of bacteria (p>0.05)

Table 2. Antibiotics susceptibility of pathogenic bacteria isolated from house flies collected from slaughter slabs in Osogbo.

BACTERIA	SENSITIVE	RESISTANCE
<i>Proteus vulgaris</i>	CRO,GEN,CPR,OFL,NIT	AUG,CRX,CAZ,AMP,CL
<i>Proteus mirabilis</i>	GEN,CPR,OFL	AUG,NIT,AMP,CRX,CAZ,CL,CRO
<i>Pseudomonas aeruginosa</i>	GEN,NIT	CRO,AMP,CAZ,CRX,CL,OFL,CPR,AUG
<i>Escherichia coli</i>	GEN,CPR,OFL,NIT	CRO,CL,CAZ,CRX,AUG,AMP
<i>Enterobacteraerogenes</i>	OFL,NIT,GEN,CPR	CL,CAZ,CRX,CRO,AMP,AUG
<i>Salmonella typhi</i>	OFL,NIT,GEN,CPR	CL,CAZ,CRX,CRO,AMP,AUG
<i>Klebsiella sp.</i>	GEN,CPR,OFL	AUG,NIT,AMP,CRX,CAZ,CL,CRO
<i>Providencia sp.</i>	GEN,CPR,OFL,NIT	CRO,CL,CAZ,CRX,AUG,AMP

N:B CRO-Ceftriaxone; CL-Cephalexin; AMP-Ampicilin; CAZ-Ceftazidime; CRX-Cefuroxime; GEN-Gentamycin; CPR-Ciprofloxin; OFL-Ofloxacin; AUG-Amoxycilin; NIT-Nitrofuratoin

Table 3. Antibiotic sensitivity of bacteria isolated from houseflies collected from dumping ground in Osogbo.

BACTERIA	SENSITIVE	RESISTANT
<i>Pseudomonas aeruginosa</i>	CPR,OFL,GEN	AUG,NIT,CRX,CAZ,AMP,CL,CRO
<i>Escherichia coli</i>	GEN,CPR,OFL,NIT	CRO,CL,CAZ,CRX,AUG,AMP
<i>Enterobacteraerogenes</i>	GEN,CPR,OFL	AUG,NIT,CRX,CAZ,AMP,CL,CRO
<i>Salmonella typhi</i>	GEN,OFL,CPR,NIT	AMP,AUG,CRX,CAZ,CL,CRO
<i>Morganellamonganii</i>	CPR,OFL,GEN	AUG,NIT,CRX,CAZ,AMP,CL,CRO
<i>Vibrio cholera</i>	NIT,GEN	CRO,CL,CAZ,CRX,AUG,AMP,CPR,OFL

N:B CRO-Ceftriaxone; CL-Cephalexin; AMP-Ampicilin; CAZ-Ceftazidime; CRX-Cefuroxime; GEN-Gentamycin; CPR-Ciprofloxin; OFL-Ofloxacin; AUG-Amoxycilin; NIT-Nitrofuratoin

Table 4. Antibiotic susceptibility of bacteria isolated from houseflies collected from palm wine spots in Osogbo.

BACTERIA	SENSITIVE	RESISTANT
<i>Escherichia coli</i>	CRO,GEN,CPR,OFL	CL,AMP,CAZ,CRX,AUG
<i>Enterobacteraerogenes</i>	CRO,GEN,CPR,OFL	CL,AMP,CAZ,CRX,AUG
<i>Klebsiella sp.</i>	NIT,OFL,CPR,GEN	AUG,CRX,CAZ,AMP,CL,CRO
<i>Morganellamonganii</i>	GEN,CPR,OFL	AUG,NIT,CRX,CAZ,AMP,CL,CRO
<i>Vibrio cholera</i>	NIT,GEN,CPR,OFL,CRO	CL,AMP,CAZ,CRX,AUG
<i>Shigella sp.</i>	GEN,CPR,OFL	AUG,NIT,CRX,CAZ,AMP,CL,CRO
<i>Serrantiamarcescens</i>	CPR,GEN,OFL,NIT	CRX,CAZ,CL,AMP,CRO,AUG

N:B CRO-Ceftriaxone; CL-Cephalexin; AMP-Ampicilin; CAZ-Ceftazidime; CRX-Cefuroxime; GEN-Gentamycin; CPR Ciprofloxin; OFL-Ofloxacin; AUG-Amoxycilin; NIT-Nitrofuratoin

Table 5. Parasitological analysis of houseflies collected from various sites in Osogbo.

PARASITES	SLAUGHTER SLABS	DUMPING GROUNDS	PALM WINE SPOTS
<i>Ascaris lumbricoides</i>	+	-	-
<i>Entamoebahistoltytica</i>	+	+	+