Epidemiol. Infect. (1991), **107**, 181–187
Printed in Great Britain

Cockroaches (*Blattella germanica*) as carriers of microorganisms of medical importance in hospitals

R. FOTEDAR[1], U. BANERJEE SHRINIWAS[1] AND A. VERMA[2]

Departments of [1]Microbiology and [2]Surgery, All India Institute of Medical Sciences, New Dehli 110029, India

(*Accepted 20 February 1991*)

SUMMARY

A study was conducted to isolate and identify microorganisms of medical importance from cockroaches (*Blattella germanica*) and to ascertain their vector potential in the epidemiology of nosocomial infections. Bacteria, fungi and parasites of medical importance were isolated and identified. Important bacterial pathogens responsible for wound infections, were further studies by antibiograms. One hundred and fifty-eight out of 159 (99·4%) cockroaches collected from hospital (test) and 113 out of 120 (94·2%) cockroaches collected from residential areas (control) were carrying medically important microorganisms ($P < 0{\cdot}05$). A significantly higher ($P < 0{\cdot}001$) number of test cockroaches were carrying a higher bacterial load (1×10^4 and 1×10^5) as compared to control cockroaches. Multiple drug-resistant bacteria were isolated from test cockroaches. The diversity of drug-resistant bacterial species isolated from test cockroaches suggests their involvement in the transmission of drug-resistant bacteria. Various fungi and parasitic cysts of medical importance were also isolated from the test and control cockroaches, but the carriage rates were low. The findings suggest that cockroaches, in hospitals, can act as potential vectors of medically important bacteria/parasites/fungi.

INTRODUCTION

The medical importance of cockroaches is much greater than generally realized as they have been shown to harbour a number of pathogenic and non-pathogenic microorganisms [1, 2]. Cockroaches are known vectors of human enteropathogens as various workers have reported the isolation of various human pathogens from these insects [1, 3–5].

Cockroaches are likely to be encountered in environments which provide favourable environmental conditions and a ready source of food. Since the hospital environment provides them with suitable temperature, humidity and food, these insects are always present in varying numbers [6]. They have been shown to feed readily on faeces, sputum, skin scrapings and other human detritus as well as on a variety of foodstuffs [1, 2]. Their nocturnal and filthy habits make them ideal carriers for transmitting various pathogenic microorganisms. *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella* spp. and several other potential pathogens have been isolated from cockroaches collected from hospitals

[1, 2, 4, 5]. Since there are no studies to evaluate the possible transmission of bacteria, fungi and parasites in the hospital environment by cockroaches, the exact role played by these insects as vectors of potentially pathogenic microorganisms, remains poorly understood. Although it has been proved that cockroaches carry a large flora of pathogenic bacteria, their direct involvement in disease transmission is difficult to demonstrate [2, 4, 7]. Nevertheless, the presence of cockroaches in hospitals and their ability to carry pathogenic organisms suggests their involvement in the transmission of some infectious diseases [8].

A prospective study was therefore undertaken to ascertain the microbial profile (microorganisms of medical importance only) from external and internal surfaces of the cockroaches (*Blattella germanica*) caught from a defined area of the All India Institute of Medical Sciences (AIIMS) hospital. For comparison, during the same time period, cockroaches (*B. germanica*) were also caught from a distant residential area and their microbial flora was also studied. The bacteria commonly found to be responsible for hospital infections (*Klebsiella* spp., *Ps. aeruginosa* and *Staphylococcus aureus*) were further studied by their antimicrobial resistance to the commonly used antibiotics. Quantitative analysis of pathogenic bacteria carried by these insects, was also carried out.

MATERIALS AND METHODS

Two hundred and seventy-nine cockroaches were collected, over a period of 3·5 years (November 1985 to April 1989), 159 from a surgical ward of the All India Institute of Medical Sciences (AIIMS) hospital (test group), and 120 from a residential area (GK – I) situated 5 km from the hospital (control group). The test group of insects was collected (in day time as well as night) from the immediate environs of the patients (beds, cupboards, wooden racks, benches, stools, etc.) and floor of the wards as well as from the dressing rooms. The control cockroaches were collected from kitchens and store rooms of residential areas.

Each cockroach was collected in a sterile test tube, transported to the laboratory and anaesthetized by freezing at 0 °C for 5 min, examined under the dissecting microscope and identified using standard taxonomic keys [9].

Isolation and identification of microorganisms from external surfaces

After identification, 2 ml of sterile normal saline (0·85%) was added to the tube and the cockroach was thoroughly shaken for 2 min. A fixed volume (0·01 ml each) of the washing was cultured on blood agar, MacConkey agar, and desoxycholate citrate agar plates separately, incubated overnight at 37 °C and the colonies identified by standard bacteriological procedures [10]. Briefly, a representative colony was studied by its macroscopic morphology, Gram's stain, various biochemicals and other specific characters. Simultaneously, 0·5 ml of the washings were also inoculated in thioglycollate and Selenite-F broths, incubated for 24 h at 37 °C and subcultured on the same media. The results were read and colonies identified after overnight incubation at 37 °C.

For isolation of fungi, the washing was cultured on Sabouraud's dextrose agar with 0·5% chloramphenicol [11]. The tubes were incubated at 25 °C and the resulting growth (if any) was identified by standard mycological procedures [12].

For parasitic ova/cyst, about 1 ml of washing was centrifuged at 2000 for 5 min and the deposit examined after staining with 1% Lugols iodine under light microscopy and identified [13].

Isolation and identification of microorganisms from internal surfaces

After external washings, the cockroaches were washed in 70 percent ethyl alcohol for 5 min (to decontaminate external surfaces as 70% alcohol is bactericidal) and allowed to dry at room temperature under sterile conditions. Cockroaches were then washed with sterile normal saline for 2–3 min to remove traces of alcohol. The gut of the cockroach was dissected out and macerated aseptically in a sterile pestle and mortar in 2 ml of sterile normal saline. The resulting macerate was then processed in a similar way as described above and the results recorded.

Quantitative estimation of bacterial isolates

Quantitative analysis of medically important bacteria (*Klebsiella* spp., *E. coli*, *Proteus* spp., *Ps. aeruginosa* and *S. aureus*) isolated from external and internal surfaces of each insect was calculated by Miles and Misra's method [14]. In brief, 0·05 ml undiluted and two tenfold dilutions of 0·05 ml of washings (external and internal) were cultured on blood agar and MacConkey agar plates in duplicate. Colony-forming units (c.f.u.) were counted after overnight incubation at 37 °C and mean count of plates was taken. From this, viable count of a particular bacteria was calculated in 2 ml of washings. The overall load of bacteria carried by each insect was counted by taking into consideration both external and internal c.f.u. together.

Antibiograms

All the strains of *Klebsiella* spp., *Ps. aeruginosa*, and *S. aureus*, were tested for antimicrobial sensitivity using a modified Stokes disk diffusion method [15], using the following antibiotics (μg/disk): amikacin, 30·0; streptomycin, 100·0; gentamicin, 10·0; tetracycline, 25·0; erythromycin, 15·0; chloromycetin, 25·0; sisomycin, 100·0; cephalexin, 30·0; ampicillin, 10·0.

RESULTS

Medically important microorganisms were isolated from external and internal surfaces of 99·4% of test cockroaches and 94·2% of the control cockroaches, the difference being statistically significant ($P < 0{\cdot}05$, χ^2 test).

Figure 1 shows the quantitative estimation of pathogenic bacteria i.e. *Klebsiella* spp., *E. coli*, *Proteus* spp., *Ps. aeruginosa*, and *S. aureus* (commonly found in hospitals) isolated from cockroaches. A high bacterial load, greater than 10^4 c.f.u. was carried by 35·8% of test cockroaches as compared to 14·2% of control, the difference being statistically significant ($P < 0{\cdot}001$, χ^2 test). On the contrary, non-pathogenic bacteria were carried by 47·1% of test and 65·8% of control cockroaches, the difference was again statistically significant ($P < 0{\cdot}05$, χ^2 test, Fig. 1).

https://doi.org/10.1017/S0950268800048809 Published online by Cambridge University Press

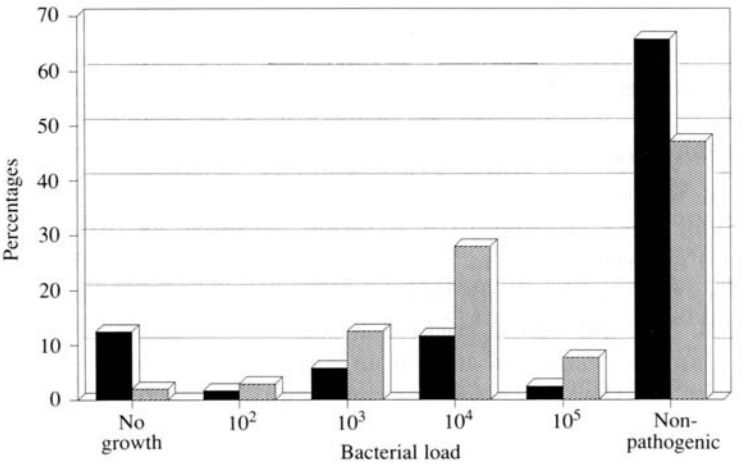

Fig. 1. Comparative bacterial loads carried by cockroaches. ■, Control; ▨, test.

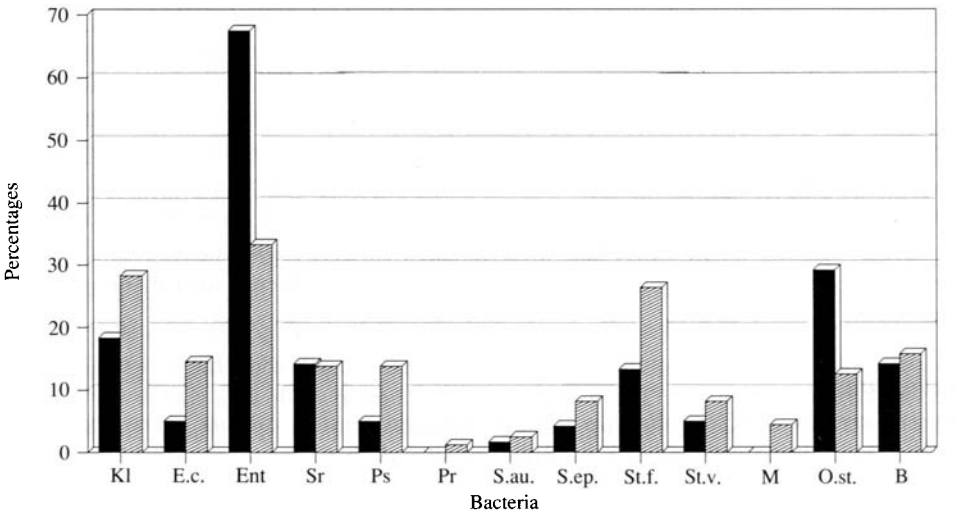

Fig. 2. Bacterial carriage rate of cockroaches. ■, Control; ▨, test. Abbreviations: Kl. klebsiella; E.c., *E. coli*; Ent. enterobacter; Sr. serratia; Ps. *P. aeruginosa*; Pr. proteus; S.au., *S. aureus*; S.ep., *S. epidermidis*; St.f., *S. faecalis*; St.v., *S. viridans*; M. micrococci; O.st., other streptococci; B. Bacillus.

Among common bacterial pathogens encountered in hospitals, *Klebsiella* spp., *E. coli*, *Ps. aeruginosa* and *S. aureus* were isolated in higher numbers from test cockroaches as compared with control cockroaches (Fig. 2). The presence of *E. coli* among cockroaches means that cockroaches have been in contact with human faeces or faeces contaminated objects [16].

Resistance to four or more antimicrobials was observed for bacterial pathogens like *Klebsiella* spp., *Ps. aeruginosa*, and *S. aureus* isolated from test cockroaches (Fig. 3). More than 85% *Klebsiella* isolates from test cockroaches showed resistance to four or more antimicrobials, whereas only 3·3% *Klebsiella* isolates from control cockroaches showed this type of resistance pattern. This difference was statistically significant ($P < 0{\cdot}001$, χ^2 test Fig. 3).

Apart from bacteria, human parasites were also isolated from test and control

https://doi.org/10.1017/S0950268800048809 Published online by Cambridge University Press

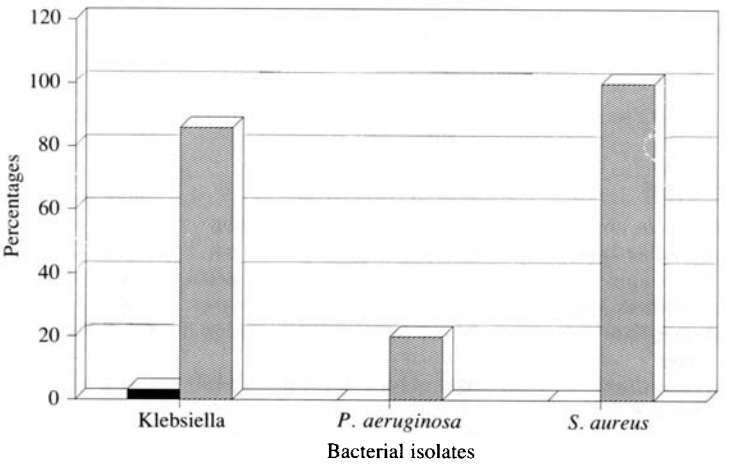

Fig. 3. Antimicrobial resistance of bacterial isolates. ■, Control; ▨, test.

Table 1. *Carriage rate of medically important parasites (cysts) from cockroaches*

Parasites isolated	Test ($N = 159$) No.	Test ($N = 159$) %	Control ($N = 120$) No.	Control ($N = 120$) %	*P* Value
Endolimax nana	10	6·2	06	5·0	N.S.
Entamoeba coli	03	1·8	02	1·6	N.S.
Entamoeba histolytica	0	—	01	0·8	N.S.

N.S., non-significant.

groups of cockroaches. Low carriage rates (statistically insignificant) for medically important parasitic cysts were observed, among test and control groups of cockroaches (Table 1).

Various medically important fungi were also isolated from test and control cockroaches (Table 2). *Candida* spp., were isolated both from test and control group of cockroaches. When carriage rates were compared, no statistically significant difference was observed among various fungal isolates except *Aspergillus niger*, which was isolated in higher number from control cockroaches (Table 2).

DISCUSSION

An impressive array of pathogenic microorganisms have been reported to be carried by the cockroaches collected from domestic, hospital and catering establishments [1, 2, 4, 5]. In this study also a high percentage of test cockroaches (99·4%) were found to carry various microorganisms (aerobic bacteria, fungi and parasites) of medical importance as compared to the control cockroaches (94·2% P value $< 0{\cdot}05$, χ^2 test). This finding suggests that almost all the cockroaches in the hospital wards carry medically important pathogenic microorganisms. Isolation of enteric bacteria from 83% of the cockroaches collected from different

https://doi.org/10.1017/S0950268800048809 Published online by Cambridge University Press

Table 2. *Carriage rate of medically important fungi from cockroaches*

Fungi isolated	Test ($N = 159$) No.	%	Control ($N = 120$) No.	%	P Value
Candida spp.	61	38·3	44	36·6	N.S.
Rhizopus spp.	01	0·6	01	0·8	N.S.
Mucor spp.	01	0·6	01	0·8	N.S.
Alternaria spp.	01	0·6	0	—	N.S.
Aspergillus niger	06	3·7	13	10·8	*
Aspergillus flavus	03	1·8	02	1·6	N.S.
Aspergillus spp.	0	—	06	5·0	N.S.

* $P < 0·05$.
N.S., non-significant.

areas of a hospital has been reported [3]. As bacteria were not isolated in the remaining 17·1% of hospital cockroaches, it appears that enteric bacteria do not inhabit cockroaches permanently but are of irregular occurrence only.

Quantitative analysis of the bacterial load carried by cockroaches of the pathogenic bacteria commonly responsible for hospital infections (*Klebsiella* spp., *Proteus* spp., *E. coli*, *Ps. aeruginosa* and *S. aureus*) revealed a higher number of test cockroaches carrying these bacteria in very high numbers as compared with control cockroaches (Fig. 1). The difference was statistically significant ($P < 0·001$, χ^2 test). There are no comparable reports in the literature, although the average colonization rate of 10^5 bacteria per mg of adult cockroach has been reported from the cockroach *Supella supellectilium* collected from hospital wards, in France [8].

It is apparent that multiple drug-resistant bacteria of medical importance were picked up by the test cockroaches from the hospital environment. These cockroaches therefore can act as mobile reservoirs of such bacteria and can start a fresh outbreak of life-threatening septicaemia/infections in neonates/intensive care units/immunocompromised patients.

It is difficult to comment upon the importance of carriage of parasitic cysts and fungi by the cockroaches, as these are carried in quite low numbers. The isolation of parasitic cysts and fungi has also been reported in low percentages by other workers [17, 18]. These insects can definitely act as carriers/vectors of these parasitic fungi/parasites.

REFERENCES

1 Roth LM, Willis ER. The medical and veterinary importance of cockroaches. Smithsonian Miscellaneous Collections 1957; **134**: 1–147.
2 Burgess NRH. The potential of cockroaches as vectors of pathogenic microorganisms. Ph. D Thesis University of London. 1974.
3 Bitter RS, Williams OB. Enteric organisms from the American cockroach. J Infect Dis 1949; **85**: 87–90.
4 Burgess NRH, Chetwyn KN. Cockroaches and the hospital environment. Nursing Times 1979; **6**: 5–7.

5 Fotedar R, Nayar E, Samantray JC, Banerjee U, Dogra V, Kumar A. Cockroaches as vectors of pathogenic bacteria. J Comm Dis 1989; **20**: 318–22.
6 Baker LF. Pests in hospitals. J Hos Infect 1981; **2**: 5–9.
7 Cochran DG, Grayson JM, Gurney AB. Cockroaches: Biology and Control. W.H.O/V.B.C. 1975; 75: 576: 1–48.
8 Guyader A Le, Rivault C, Chaperon J. Microbial organisms carried by brown banded cockroaches in relation to their spatial distribution in a hospital. Epidemiol Infect 1989; **102**: 485–92.
9 Harwood RF, James MT. In Entomology in human and animal health, 7th ed. New York; Macmillan Publishing, 1979.
10 Cruikshank R. Medical microbiology, vol. 2, 13th. ed. London: Churchill Livingston, 1978.
11 Ajello L, George LK, Kaplan W, Kaufman L. In 'CDC laboratory manual for medical mycology.' U.S. Dept of Health, Education and Welfare. Public Health Service Communicable Disease Centre. Atlanta 22, Georgia. 1963; 1–30.
12 Conant NF, Smith DT, Baker DR, Callaway JL, Martin SD. 'Manual of clinical mycology', 2nd ed. Philadelphia: Saunders and Company, 1955.
13 Chatterjee KD. In 'Parasitology (protozoology and helminthology) in relation to clinical medicine'. 11th edn. Calcutta: Sree Saraswaty Press Ltd, 1976.
14 Miles AA, Misra SS. The estimation of the bactericidal power of blood. J Hyg Camb 1938; **38**: 732–48.
15 Shriniwas, Biswas TD, Gupta UK. Evaluation of disc diffusion technique for *in vitro* antimicrobial sensitivity. Ind J Med Microbiol 1985; **3**: 215–23.
16 Frishman AM, Alcamo IE. Domestic cockroaches and human bacterial disease. Pest Control 1977; **45**: 16–20.
17 DeCoursey JD, Otto JS. Entamoeba histolytica and certain other protozoan organisms found in cockroaches in Cairo, Egypt. J NY Entomol Soc 1957; **64**: 157–63.
18 Okafor JI. Bacterial and fungal pathogens from the intestinal tracts of cockroaches. J Comm Dis 1981; **13**: 128–31.