

Cockroaches (*Blatta* and *Periplaneta* species) as reservoirs of drug-resistant salmonellas

S. J. N. DEVI¹* AND C. J. MURRAY²

¹Laboratory of Developmental and Molecular Immunity, National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, MD 20892 USA

²Salmonella Reference Laboratory, The Institute of Medical and Veterinary Science, Adelaide, S. Australia 5000

(Accepted 24 April 1991)

SUMMARY

A total of 221 cockroaches (*Blatta* and *Periplaneta* spp.), collected in hospitals, houses, animal sheds, grocery stores and restaurants, in various parts of South Kanara District, a south-west coastal region of India, were studied bacteriologically for the presence of various salmonellas. Salmonellas were isolated from 4.1% of these cockroaches. Nine strains of salmonellas were recovered, belonging to five serotypes – *Salmonella* *bovismorbificans*, *S. oslo*, *S. typhimurium*, *S. mbandaka* and *S. braenderup*, the former two being the commonest serotypes. All salmonellas were resistant to one or other of 11 antibacterial drugs used in the susceptibility test. Isolation of salmonellas from cockroaches collected from the livestock premises and human dwellings suggested that they may act as significant reservoirs of salmonella in nature. Recovery of serotypes, phage types and R-types that were commonly isolated from humans and animals of this locality, suggested a transmission role for cockroaches. By harbouring potentially pathogenic, drug-resistant salmonellas, these wandering arthropods may pose dangerous infective hazards to humans and animals.

INTRODUCTION

A great variety of insects are incriminated as carriers of several pathogens [1]. Arthropods are often recognized as harbouring a wide spectrum of microorganisms. Cockroaches are known to carry a diverse pathogenic bacterial flora, although their role in the direct transmission of infection is seldom established [2–8]. Cockroaches have been identified in the past as carriers of salmonella [9–13]. The free wandering movements of cockroaches from one location to another and their possible contribution to disease transmission prompted this study, the purpose of which was to determine the extent of salmonella carriage by cockroaches captured

* Address for correspondence and reprint requests: Dr Sarvamangala Devi, J.N., LDML, NICHD, National Institutes of Health, Bldg 6, Room 1A05, Bethesda, Maryland 20892, USA.

at various ecological niches, to assess the serotype, phage type and antibiotic sensitivity patterns of the isolates and to determine their role in the epidemiology of salmonellosis in South Kanara District, South India.

MATERIALS AND METHODS

Collection of specimens

During the period of this survey from January 1981 to December 1983, a total of 221 adult cockroaches was collected in different houses, animal sheds, restaurants, grocery stores and hospitals in various parts of South Kanara District. The cockroaches were trapped in the kitchen, bathroom, storage areas of houses, the corridors, patient rooms and cafeterias of hospitals and the kitchen, storage areas, sewers and garbage cans of restaurants. These cockroaches were identified as belonging to *Blatta* and *Periplaneta* species.

Processing of specimens

After transport to the laboratory, the cockroaches were killed with diethyl ether and their digestive tracts were dissected using autoclave-sterilized instruments. The guts were processed with and without enrichment, on MacConkey's agar, brilliant green agar and eosin methylene blue agar. Selenite broth and tetrathionate broth [14] were used as enrichment media and were incubated at 37 °C for 24–30 h. The culture plates were incubated at 37 °C for 24–72 h.

Identification of salmonella

Non-lactose-fermenting colonies were identified by subjecting them to a battery of biochemical tests [15]. The biochemical identification was confirmed by slide agglutination and coagglutination [16] with salmonella O and H antisera, A to E. The final reconfirmation and phage typing [17] was performed at the National Salmonella Center, Adelaide, Australia.

Antibiotic sensitivity test

Kirby-Bauer's agar disk diffusion technique [18] was used to test the susceptibility of salmonella strains to 11 antibacterial drugs: ampicillin (A, 10 µg/ml), chloramphenicol (C, 30 µg/ml), furadantin (Fu, 300 µg/ml), gentamicin (G, 10 µg/ml), kanamycin K, 30 µg/ml), streptomycin (S, 10 µg/ml), sulphadiazine (Su, 300 µg/ml), tetracycline (T, 30 µg/ml), co-trimoxazole (Tm, 25 µg/ml), nalidixic acid (Nx, 30 µg/ml), and polymixin B (Pb, 300 units/ml).

RESULTS

Of 221 cockroaches screened, 4.1% were found to harbour salmonellas in their gut. Three of nine salmonella strains recovered were *S. bovis/morbificans* belonging to phage types 11, 13 and 23, all of which were resistant strains. Two *S. typhimurium* strains, both resistant, belonged to UDNC phage type. Two strains of *S. oslo*, and one each of *S. mbandaka* and *S. braenderup* were also recovered (Table 1).

Table 1. Serotype and drug resistance patterns of salmonellas recovered from cockroaches (*Blatta* and *Periplaneta* sp)

Source	Numbered cultured	Number culture positive	Serotype (number)	Phage type	R-pattern*
Hospital	21	3	<i>S. typhimurium</i> (1)	UDNC	FuT
			<i>S. bovismorbificans</i> (1)	23	FuSuT
			<i>S. oslo</i> (1)	—	SuT
Grocery stores	49	2	<i>S. bovismorbificans</i> (1)	13	SuNx
			<i>S. mbandaka</i> (1)	—	SuT
Animal sheds	82	2	<i>S. braenderup</i> (1)	—	Nx
			<i>S. oslo</i> (1)	—	SuT
Houses	44	1	<i>S. bovismorbificans</i> (1)	11	Su
Restaurants	25	1	<i>S. typhimurium</i> (1)	UDNC	FuSu

* See Materials and Methods for details.

DISCUSSION

The potential role of invertebrate vectors in the spread of several infectious diseases is undeniable. Cockroaches carry several pathogenic bacteria, but do not suffer from any disease [2]. Species of *Citrobacter*, *Enterobacter*, *Klebsiella* and *Serratia* represent the normal flora of cockroaches. *Acinetobacter* sp., *Pseudomonas fluorescens*, *Ps. putida*, *Ps. aeruginosa* and *Escherichia coli* have been isolated from cockroaches captured in hospitals [8]. Bacteria were also introduced into the gut of cockroaches experimentally through their diet [9, 13, 19, 20]. Although many animals and insects carry salmonellas in their gut, cockroaches become particularly significant because of their nocturnal wandering, feeding and defaecation habits.

The isolation of appreciable numbers of salmonellas (4.1%) from cockroaches captured in human dwelling areas and livestock premises indicates that these domestic insects can pose a problem. Except for *S. braenderup*, all other serotypes, R-types and various phage types of *S. typhimurium* and *S. bovismorbificans* have been isolated from human, animal and environmental sources in this south-west coastal region of South India between 1981 and 1983 [21], forming circumstantial evidence for the contribution of cockroaches to the occurrence and persistence of salmonella infections in this area. That *S. braenderup* was recovered for the first time in India during this study [22], indicates that cockroaches may introduce rare serotypes into the community.

Isolation of drug-resistant salmonellas from cockroaches of the region encompassed in this study was not surprising, because similar observations were made previously among salmonella strains recovered from frogs [23]. As revealed by a 3-year epidemiological study, there was a constant circulation of drug-resistant salmonellas in the community, both amongst humans and animals and in the environment [21]. This is most likely due to the uncontrolled availability of antibiotics in the pharmacy and the indiscriminate use of antibiotics in human and animal populations. The ubiquitous existence of cockroaches in nature, in close association with man and animals, and their ability to carry pathogenic drug-

resistant salmonellas are suggestive of their involvement in the transmission of infections, especially in hospitals. Drug-resistant salmonellas may be spread in the hospital environment through contaminated foods, pharmaceutical, medical devices, hospital equipment and aerosols.

Cockroaches probably acquire salmonellas naturally from the animal- and human-polluted environment. As they feed indiscriminately on both the food and faecal matters of human and animal origin, they may acquire salmonellas in sewers, toilets and latrines and carry the pathogens to food storage and food handling areas such as kitchens, cafeterias, canteens or restaurants. They may also carry salmonellas on their exterior body parts including bristly legs and may act as potential mechanical vectors in the dissemination of infections. *S. typhi* was isolated from the legs and faeces of cockroaches captured in the homes of people who suffered from typhoid fever [24]. Hospital outbreaks of salmonellosis due to *S. typhimurium* [25] and *S. bovismoribificans* [26] were traced to cockroaches. Salmonellas can remain viable in the faeces of cockroaches for varied lengths of time. By experimental infection, *S. oranienberg* was shown to remain viable in the faeces of *Periplaneta americana* for as long as 85 days to 6 months under humid conditions [27]. This suggests that contamination of food and water sources with cockroach faeces may lead to epidemics.

The presence of salmonellas in cockroaches becomes important in many respects. Cockroaches form the natural prey for a variety of animals and birds; they have been found in the stomach contents of several amphibians, reptiles, rodents, domestic and wild birds and animals [2]. While cockroaches form a part of human food in certain countries and cultures, they are also used for medicinal purposes by others [2].

Frogs and many reptiles have been identified as natural reservoirs of salmonella in the south-west coastal region of India [21, 23]. This study provides information on yet another reservoir of salmonella in nature and the possible mode of transmission of salmonella infections in the community. The transmission of salmonellas from these natural reservoirs to humans and animals or vice versa was substantiated by the finding of identical serotypes, phage types and R-types in the human, animal and environmental sources of this locality [21, 23].

Thus, cockroaches appear to play a significant role in the epidemiology of salmonellosis in this geographical area. Salmonella carriage in cockroaches, which infest human and animal habitats, increases the risk of potential salmonella contamination, particularly in hospitals, houses and restaurants. By forming the natural prey for a wide spectrum of domestic and wild animals and birds, cockroaches help distribute salmonellas among different host species. Greater attention has to be paid to these insects particularly in areas of poor sanitation.

ACKNOWLEDGEMENTS

The first author acknowledges the laboratory facilities provided by Dr P. G. Shivananda, Department of Microbiology, Kasturba Medical College, Manipal, India. The assistance of Ms U. Girija in the collection of specimens is gratefully acknowledged.

REFERENCES

1. Steinhaus EA. A study of the bacteria associated with thirty species of insects. *J Bacteriol* 1941; **42**: 757–90.
2. Roth LM, Willis ER. The medical and veterinary importance of cockroaches. *Smithsonian Miscellaneous Collections* 1957; **134**: 1–147.
3. Burgess NRH, McDermott SN, Whiting J. Aerobic bacteria securing in the hind-gut of the cockroach, *Blatta orientalis*. *J Hyg* 1973; **71**: 1–7.
4. Cochran DG, Grayson JM, Gurney AB. Cockroach: Biology and control, WHO vector biology and control series, Geneva, 1–53.
5. Frishman AM, Alcamo IE. Domestic cockroaches and human bacterial disease. *Pest Control* 1977; **45**: 16–20.
6. Cornwell PB, Mendes MF. Disease organisms carried by oriental cockroaches in relation to acceptable standards of hygiene. *Int Pest Control* 1981; May–June, 722–74.
7. Cruden DL, Markovetz AJ. Microbial ecology of the cockroach gut. *Ann Rev Microbiol* 1987; **41**: 617–43.
8. Le Guyader A, 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. Mackerras IM, Pope P. Experimental salmonella infection in Australian cockroaches. *Austral J Exp Biol Med Sci* 1948; **26**: 465–70.
10. Singh SP, Sethi MS, Sharma VD. The occurrence of salmonellae in rodent, shrew, cockroach and ant. *Int J Zoonoses* 1980; **7**: 58–61.
11. Panhotra BR, Agnihotri V, Agarwal KC, Batta RP. Isolation of salmonellae from hospital food and vermin. *Indian J Med Res* 1981; **74**: 648–51.
12. Saxena SN, Mago ML, Rao Bhau LN, Ahuja S, Singh H. Salmonella serotypes prevalent in India during 1978–81. *Indian J Med Res* 1983; **77**: 10–18.
13. Ash N, Greenberg B. Vector potential of the German cockroach (*Dictyoptera blattellidae*) in dissemination of *Salmonella enteritidis* serotype *typhimurium*. *J Med Entomol* 1980; **17**: 417–23.
14. Cruickshank R, Duguid JP, Marmion BP, Swain RHA. Medical microbiology. 12th ed. Edinburgh, London: E. & S. Livingstone, 1975: 126–8.
15. Edwards PR, Ewing WH, eds. Identification of Enterobacteriaceae. 3rd ed. Minneapolis: Burgess Publishing Co. 1972: 21–47.
16. Devi SJN, Shivananda PG. Coagglutination technique for identification of salmonella. *Indian J Med Res* 1983; **78**: 459–64.
17. Callow BR. A new phage-typing scheme for *Salmonella typhimurium*. *J Hyg* 1959; **57**: 346–59.
18. Bauer AW, Kirby WMM, Sherris JC, Tenckhoff M. Antibiotic susceptibility testing by a standardized single disk method. *AM J Clin Pathol* 1966; **45**: 493–7.
19. Burgess NRH, McDermott SN, Whiting J. Laboratory transmission of Enterobacteriaceae by the oriental cockroach *Blatta orientalis*. *J Hyg* 1981; **71**: 9–14.
20. Klowden MJ, Greenberg B. Salmonella in the American cockroach: evaluation of vector potential through dosed feeding experiments. *J Hyg* 1976; **77**: 105–11.
21. Devi SJN. An investigation into the incidence of salmonellosis in and around South Kanara District, India. *Arogya J Hlth Sci* 1989; **15**: 80–4.
22. Devi SJN, Murray CJ, Shivananda PG. Isolation of *Salmonella braenderup* (6,7:e,h:e,n,z15) for the first time in India. *Indian J Med Sci* 1986; **40**: 177–8.
23. Devi SJN, Shivananda PG. Frogs as natural reservoirs of salmonella. *Indian J Med Res* 1983; **78**: 465–70.
24. Antonelli G. La blatta nella igiene domestica. *Riv Soc Ital, Igiene, Milan* 1930; **52**: 132–42.
25. Graffar M, Mertens S. Le rôle des Blattes dans la transmission des salmonelloses. *Ann Inst Pasteur* 1950; **79**: 654–60.
26. Mackerras MJ, Mackerras IM. Salmonella infections in Australian cockroaches. *Aust J Sci* 1948; **10**: 115.
27. Olson TA, Rueger ME. Experimental transmission of *Salmonella oranienberg* through cockroaches. *Pub Hlth Rep* 1950; **65**: 531–40.