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Lizards as vectors of human salmonellosis

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SUMMARY

Human infections with *Salmonella saintpaul* have become more frequent in New Zealand in recent years. Most cases now occur in Otago. It is shown that wild lizards in Otago, particularly the common skink (*Leiolopisma zelandica*), carry *S. saintpaul* and that most if not all human outbreaks of this salmonella serotype are associated with either lizards or lizard infested areas. So far as is known this is the first report incriminating lizards as the probable cause of human salmonellosis.

INTRODUCTION

Salmonella saintpaul infection in humans was first confirmed in New Zealand in 1952. Occasional sporadic cases were observed throughout both North and South Islands since that date, but from 1959 onwards a large proportion of the cases have been in the South Island and most of them have been associated with Otago. Of the 34 human index cases recorded in New Zealand, 18 have occurred in Otago. Since 1959 69 % of recorded human index cases have been associated with Otago and since 1966 the proportion has risen to 81 %. The age range has been wide but the infection has been mainly in children, particularly from 6 months to 10 years of age. Most Otago cases have occurred in the summer season from November to April, though one infection each has been recorded for July, September and October.

In 1966, after the notification of five apparently unconnected cases of *S. saintpaul* infection in Otago an intensive investigation was carried out in an endeavour to trace a common origin. Four of these cases were associated with an area of the Clutha river running through Central Otago from Alexandra to Roxburgh. Sanitary surveys were undertaken and many water, soil, domestic animal and even vegetable samples were collected in a fruitless endeavour to find the source of these cases.

In November 1967 a 3-year-old boy was admitted to Kew Hospital, Invercargill, severely ill with *S. saintpaul* infection. Examination of the faeces of every member of the household as well as two domestic helpers showed tham all to be carriers. Eighteen other human contacts and a wide variety of domestic animals and birds

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were negative on stool culture. However, a biscuit-tin, kept in the kitchen, housed five skinks (*Leiolopisma zelandica*). The excreta of these lizards yielded an almost pure culture of *S. saintpaul*. These five lizards were the survivors of some 48 lizards which had been collected the previous month near Clyde, not far from Alexandra.

Following this discovery a collection was made in December 1967 of 30 skinks (*L. zelandica*) and 20 geckos (*Hoplodactylus pacificus*) from the same valley where the original 48 lizards had been found. Three of the wild skinks were found to be excreters of *S. saintpaul*.

A review now took place of all previous notifications of human *S. saintpaul* infections occurring in Otago. It was then established that a history of association with the handling of wild lizards or with earth, rocks or vegetation in areas where lizards abounded, was present just before the onset of the human illness in every case except one in which the source of infection had been established as arising from a goose which the patient had eaten. This goose, however, was traceable to a farm in Macraes Flat, North Otago, and was known to have eaten lizards before it was killed for the table.

A review of the human *S. saintpaul* infections (supplied by the National Health Institute) occurring in other Districts of New Zealand was not so profitable. Many of the cases had either not been notified to the local Medical Officer of Health or the inquiries undertaken at the time were insufficient to establish whether an association with lizards or lizard-infested areas had been present or not before infection. In two cases, however, there was sufficient information to make it highly probable that there had been association with lizards or lizard droppings.

Table 1 summarizes the index cases of the sporadic Otago outbreaks, showing the probable area of original infection.

LIZARD INVESTIGATION

At the end of 1967 and during the early part of 1968, 120 lizards (95 *L. zelandica* and 25 *H. pacificus*) caught wild in Otago and Southland were examined bacteriologically at Kew Hospital, Invercargill. To some extent these early examinations were experimental because it was not known exactly how specimens should be taken nor from what part of the gut. However, it was soon established that it was necessary to kill the animal and take cultures from the upper gut. None of the lizards from Southland yielded *S. saintpaul*, but three of 30 *L. zelandica* from Clyde were positive for this organism. Apart from four specimens collected from Otago Peninsula and kept as pets at a primary school which were examined at Dunedin Hospital, all further animal investigations were undertaken by the Invermay Animal Health Laboratory near Dunedin. Over the summers of 1968–1970 an additional total of 372 lizards (174 *L. zelandica* and 198 *H. pacificus*) were examined at Invermay.

In the early stages, Livestock Instructors of the Department of Agriculture managed to capture lizards for the laboratory but it was soon realized that the job was very time-consuming and by no means easy, interfering considerably with

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other more urgent work. It was therefore arranged that Inspectors of Health of the District Health Office would undertake, after prior arrangement with the laboratory, a series of 'forays' for lizard-collecting. The Inspectors worked in pairs and had instructions to take only single specimens where there were many, from as wide a range over the water catchment areas as possible. The large territorial area involved, the rugged upland terrain inhabited by the lizards and the difficult road access to many of the places visited meant that each 'foray' lasted 3 or 4 days.

Table 1. *Outbreaks of* Salmonella saintpaul *infection in humans in the southern half of South Island*

Age of index case	Date	Residence	Probable area of original infection	River catchment
52 years	Apr. 1959	Dunedin	Macraes Flat	Waikouaiti
1½ years	Sept. 1961	Roxburgh	Roxburgh	Clutha
8 weeks	Nov. 1962	Roxburgh	Roxburgh	Clutha
17 years	Jan. 1965	Palmerston	Palmerston	Shag
6 months	Mar. 1965	Roxburgh	Roxburgh	Clutha
5 years	Mar. 1966	Otematata	Otematata	Waitaki
7 years	Apr. 1966	Roxburgh	Roxburgh	Clutha
1 year	Oct. 1966	Dunedin	Alexandra	Clutha
6 years	Dec. 1966	Roxburgh	Roxburgh	Clutha
19 years	Dec. 1966	Roxburgh	Roxburgh	Clutha
3 years	Nov. 1967	Invercargill	Clyde	Clutha
2 years	Feb. 1970	Dunedin	Clyde	Clutha
2½ years	Mar. 1970	Macraes Flat	Macraes Flat	Waikouaiti
41 years	Apr. 1970	Palmerston	Palmerston	Shag
72 years	Apr. 1970	Karitane	Karitane	Waikouaiti
10 weeks	Apr. 1970	Dunedin	Dansey's Pass	Taieri
11 months	July 1970	Ettrick	Ettrick	Clutha
13 years	Nov. 1970	Roxburgh	Roxburgh	Clutha

An efficient system of catching lizards and dispatching them alive to the Invermay Laboratory had to be developed. Learning the techniques of finding 'runs', observing droppings, upturning boulders as well as the acrobatics needed to secure the skinks, which run fast when uncovered, was left to the Inspectors. Since it was essential to prevent possible cross-contamination each lizard had to be 'packed' separately.

The most satisfactory and successful system developed was to put each specimen in a plastic bag with a little air, seal and label the bag with the time, date and place of collection and pack the bags in an ice-box. The cold environment encouraged an artificial hibernation and the lizards travelled alive and well by public transport for upwards of 200 miles. Fifty or more lizards could easily be packed in an ice-box of internal capacity of about 12,000 cm.^{3}. To be successful lizard-hunts were best undertaken in hot sunny weather.

On arrival at the laboratory the lizards were killed and dissected aseptically. Stomach and gut contents were cultured separately on brilliant green and MacConkey agar plates (B.B.L.*). The entire length of stomach and intestine was then

* B.B.L. = Baltimore Biological Laboratories.

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inoculated into selenite-F enrichment broth (B.B.L.) and incubated at 42° C. for 18–20 hr., then cultured on brilliant green agar. All plates were incubated at 37° C. and examined at 18 and 48 hr.

Routine biochemical confirmation was carried out by subculturing suspicious colonies on Triple Sugar Iron agar (B.B.L. and Difco) and Christensen's urea slopes. Serological confirmation was carried out using Burroughs Wellcome somatic antisera for slide agglutination tests to determine the salmonella somatic grouping. Flagellar titrations were at first done using Burroughs Wellcome flagellar antisera, later Difco Spicer Edwards pooled antisera and single factor flagellar sera were used.

RESULTS AND CONCLUSIONS

Table 2 gives the combined results of cultures undertaken by Kew Hospital and Invermay Animal Health Laboratory. Geographically by far the largest river catchments are the Clutha and the Waitaki and hence the much greater proportion of specimens collected from these two catchment areas. Among the smallest are the Opihi, the Waihao, the Shag, the Waikouaiti, the Waikawa and the Waituna.

Table 2. *Wild lizard collections from Rakaia River southwards, tested for* Salmonella saintpaul

River catchment		*Leiolopisma zealandica*	*Hoplodactylus pacificus*	Total lizards
South Canterbury	Rakaia	0/2	0/14	0/16
	Ashburton	2/17 (12)	2/10 (20)	4/27 (15)
	Rangitata	1/9 (11)	0/9	1/18 (6)
	Opihi	0/2	0/8	0/10
	Waihao	0/2	0/3	0/5
	Sub-total	3/32 (9)	2/44 (5)	5/76 (7)
Otago	Waitaki	3/49 (6)	1/103 (1)	4/152 (2)
	Shag	2/9 (22)	0/0	2/9 (22)
	Waikouaiti	4/16 (25)	0/3	4/19 (21)
	Taieri	1/19 (5)	0/6	1/25 (4)
	Clutha	11/79 (14)	1/62 (2)	12/141 (9)
	Sub-total	21/172 (12)	2/174 (1)	23/346 (7)
Southland	Waikawa	0/10	0/0	0/10
	Mataura	0/25	0/5	0/30
	Waituna	0/25	0/0	0/25
	Oreti	0/5	0/0	0/5
	Sub-total	0/65	0/5	0/70
	Totals	24/269 (9)	4/223 (2)	28/492 (5·7)

Figures indicate no. positive/no. tested, with percentages in parentheses.

Of the Otago river catchments (the Waitaki, the Shag, the Waikouaiti, the Taieri and the Clutha) each reveals an enzootic of *S. saintpaul* infection amongst the skinks and a suggestion of a much lower infection rate amongst the geckos. The broad conclusions were reached that skinks are more frequent carriers of *S. saintpaul* than geckos and that the carrier-rate amongst skinks in Otago appears higher than in the provinces to the north or south. Why this should be is as yet unknown

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and clearly a more widespread and extended survey of the *S. saintpaul* carrier state amongst New Zealand lizards should be undertaken.

A possible explanation of the largest numbers of human cases having arisen from Central Otago in recent years is that this area attracts very large numbers of summer holiday-makers, the weather and the terrain being attractive to both humans and lizards. However, the recent cases occurring in the Waikouaiti and Shag River catchments, which are not considered holiday resort areas, throw some doubt on this hypothesis.

It has been suggested that the reason for the apparently high incidence of *S. saintpaul* infection in Otago is due to greater enthusiasm amongst Otago medical practitioners for taking faecal specimens in cases of diarrhoea. However, it seems unbelievable that in other parts of the country faecal cultures are not always taken of children suffering from severe bloody diarrhoea. Otago has a relatively small population but its proportion of all recorded *S. saintpaul* infections in New Zealand is greatly in excess of what would be expected on a population basis. Moreover, it is today very unlikely that any hospital laboratory throughout the country will confuse *S. typhimurium* with *S. saintpaul*.

This paper does not pretend to show conclusive evidence that the lizards themselves are always responsible for all human cases of *S. saintpaul* infection. The evidence, however, is strong that human infection rates are highest where skink carrier rates are highest.

Undoubtedly the actual handling of lizards is not necessary for infection to be obtained. It is apparently sufficient for close contact with earth or rocks in an area abounding with lizards to cause infection. Since it has been established that lizard excreta may contain very large numbers of the organisms it is possible that *S. saintpaul* might be picked up on hands or clothing and thereby cause infection by the oral route. The work of Wilkoff, Westbrook & Dixon (1969) on the ability of *S. typhimurium* to survive for long periods on fabrics would suggest that *S. saintpaul* might also be picked up on clothing, even perhaps from sitting on the ground contaminated with lizard excreta.

DISCUSSION

The evidence presented here, while not conclusive, is sufficient to allow for prima facie belief that lizards of the species *Leiolopisma zelandica* and *Hoplodactylus pacificus* are vectors in human salmonellosis. A search of the literature on salmonellas has failed to reveal any previously recorded cases of human salmonella infection which can be attributed to infection by members of the lizard group.

Isolations of various salmonellas from lizards have been made on a number of occasions. McNeil & Hinshaw (1946) reported *S. manhatten* from a zoo iguana and *S. montevideo* from a Gila Monster. They considered their report to 'be the first true salmonellae to be reported from lizards', although drawing attention to the work by Caldwell & Ryerson (1939) on salmonellosis in reptiles. Mackey (1955) in reporting lizard isolations from 1948 to 1953 in Dar es Salaam isolated 33 different salmonella serotypes. This formidable list included *S. typhimurium* but not

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S. saintpaul and the organisms were apparently cultured and identified from droppings of two species of common house lizard, *Hemidactylus mabonia* and *Mabuya striata*. He makes the pertinent observation that 'it is difficult to understand how the lizards become infected'.

Other salmonella isolations from lizards include one serotype from the Pacific fence lizard (*Sceloporus occidentalis*), by Hinhsaw & McNeil (1947); one in African lizards (Van Oye, 1964); and ten different serotypes in Australian lizards by Atkinson (1964). The only references to the isolation of *S. saintpaul* from reptiles were found in articles by Refai & Sadek (1968) where it occurred in *Cerastes cerastes*, a horned viper, and by Iveson, Mackay-Scollay & Bamford (1969) in Western Australia where it was isolated from the two lizard species, *Varanus varius* and *Tiliqua scincoides*.

A number of authors stress the potential importance of lizards as vectors or carriers of salmonellas, e.g. Collard & Montefiore (1956) in *Agama agama* in Ibadan, and Le Minor (1964) who found 19 out of 497 *Hemidactylus bleker* and 14 out of 152 *Peripia peronii* positive for salmonellas. Outbreaks of salmonellosis in captive lizards have been reported by Lee & Mackerras (1955). Darasse, Le Minor & Lecompte (1958) point out the potential danger of contamination of drinking water supplies from lizard salmonellae.

The first isolation of *S. saintpaul* from a human in New Zealand was made in Rotorua in 1952 (Josland, 1953). That author considered it an uncommon infection in both man and animals at that time. Since then reports of *S. saintpaul* epidemics in man, such as that by Gotoff, Boring & Lepper (1966), have shown that human infections with this salmonella may be becoming commoner. The present paper would tend to support the view that human infections with *S. saintpaul* are becoming commoner in this country, particularly in Otago, and, so far as can be ascertained, is the first paper to associate infections in man with lizards as the vectors.

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