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The epidemiology of leptospirosis in North Queensland

I. General survey of animal hosts

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INTRODUCTION

The east coast of North Queensland (Fig. 1) differs from the rest of the Australian continent in combining high rainfall with tropical temperatures. Leptospirosis was first recognized here in 1933 in the sugar-cane fields of Ingham (18° 39′ S., 146° 10′ E.) by Morrissey (1934), and the laboratory investigations were reported by Cotter & Sawers (1934). Lumley (1937) named the first two serotypes isolated, and three others had been identified (Johnson, 1950) by the time the Institute's Field Station was established at Innisfail (17° 32′ S., 146° 02′ E.) in 1951. Subsequent investigation of cases of fever increased the number of serotypes known to infect man in the area to fourteen (Sinnamon *et al.* 1953; Smith *et al.* 1954; Smith & Brown, 1955; Broom & Smith, 1956; Addamiano, Babudieri & Smith, 1960; Alexander & Smith, 1962), and their general epidemiology was analysed by Derrick *et al.* (1954) and Derrick (1956). A fifteenth (*Leptospira bindjei*) has been recognized since our work was completed, and is reported in Part II (Battey, Smith & Barrow, 1964).

The association of rodents and marsupial bandicoots with canefield leptospirosis was observed in the early investigations (Cotter & Sawers, 1934). Leptospires seen in the kidneys of bandicoots were not isolated and identified, but *Leptospira zanoni* (= *australis B*) was recovered from *Rattus rattus*, and *L. australis* (= *australis A*) was isolated in the Ingham district from *R. sordidus conatus*, which was recorded at that time as *R. culmorum* (Cotter, 1935; Sawers, 1938). A later series of animals was reported by Doherty, Emanuel & Moore (1956), and is included in the present survey. These authors found that 11 out of 16 *R. s. conatus* from the Babinda and Mirriwinni areas (Fig. 1) were infected with *L. australis*, nine being urinary carriers, and that antibodies to *L. australis* and other serotypes were present in the sera of bandicoots.

Nearly half the cases of leptospirosis in the area proved to have no occupational relation to the canefields (Derrick, 1956), and it soon became apparent that a knowledge of the associations between the serotypes and their animal hosts was essential before the epidemiology of the disease could be fully understood. There

was a further incentive to pursue this line of inquiry. Leptospirosis causes an appreciable amount of illness and economic loss (largely in payment of compensation for lost wages) in North Queensland, but it is rarely fatal, and people do not fear it enough to exert themselves seriously to prevent it. Burning the cane in the

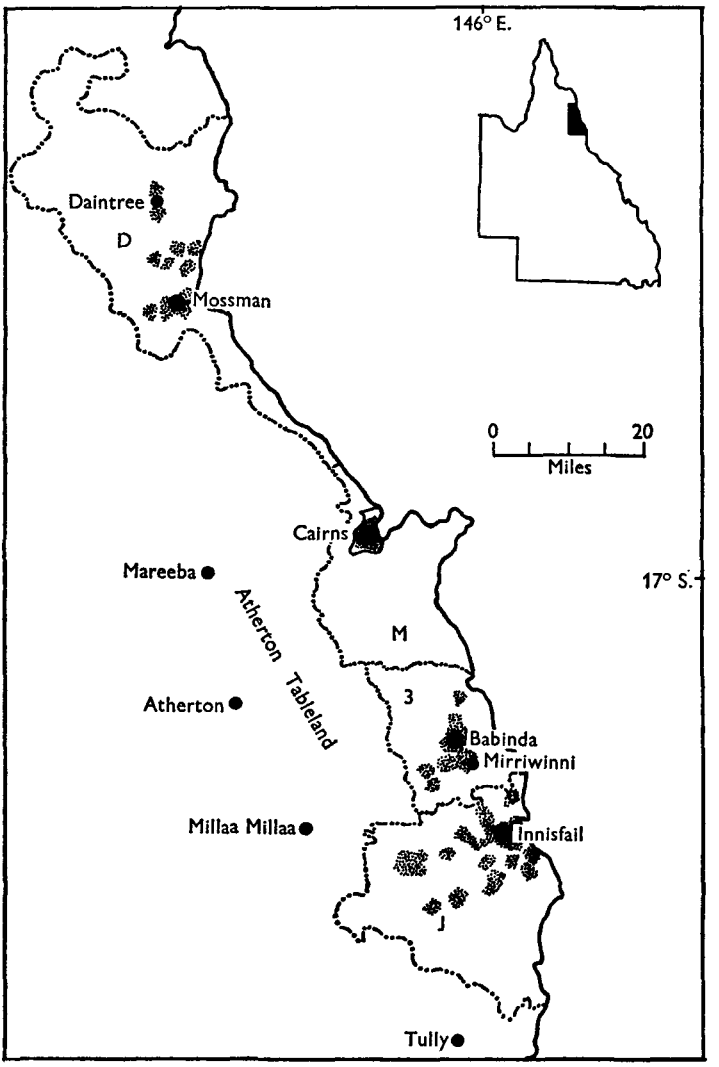

Fig. 1. Map showing trapping areas (stippled) in North Queensland. Shires are indicated by: J, Johnstone; D, Douglas; 3, Division 3 of M, Mulgrave.

fields before it is cut is the only community measure that can be enforced, and it eliminates neither rats nor leptospires (Emanuel & Harrison, 1961; Doherty *et al.* 1956); protective clothing will not be worn; while vaccination, if it were effective, would meet apathy and possibly obstruction. It is difficult to escape the conclusion that the only method of prevention that might be socially and economically practicable would be some form of ecological control, and that could not even be

thought about without a clear understanding of the hosts, their ecology, and the conditions which determine the incidence of infection in them.

This phase of the research was therefore developed in two stages. In the first, which is reported here and in Part II, the plan was to examine as many animals of as many species as possible, over a relatively wide range of country, in an attempt to assess the significance of different species as hosts of different serotypes. The second stage grew out of the ecological studies of the mammals, which proceeded concurrently (Harrison, 1962). Natural leptospiral infections were followed in the course of mark–recapture observations on the animals, and information was obtained on duration of infection, persistence of antibodies, and the relationship between density of host populations and the incidence of infection in them. These data are still being analysed.

MATERIALS AND METHODS

Most of the material in the general survey was collected by the staff of the Field Station between June 1953 and May 1961. It comprised 5 monotremes, 463 marsupials, 1350 rodents, 67 bats, 295 domestic mammals, 30 birds, 28 reptiles, and 21 amphibians. It has been supplemented, where appropriate (notably in Tables 3 and 4), by records from an additional 180 bandicoots and 1005 rodents that were collected in the mark–recapture areas.

Collection of animals

The topography of the area has been described by Derrick *et al.* (1954) and Harrison (1962). It presents a series of mountain ranges and tablelands, separated from the Pacific Ocean by a narrow coastal plain. Except for a lower range running approximately parallel to the coast, the land between the mountain chain and the coast is used mainly for growing sugar cane.

Most of the wild animals examined were obtained from the Johnstone, Mulgrave (Division 3), and Douglas shires, including the towns of Innisfail, Babinda, and Mossman (Fig. 1). The average annual rainfall in these areas varies from approximately 90–150 in. In addition, 28 animals from Cairns were examined, and 61 sera from the Atherton Tableland, including 46 samples from marsupials collected in 1945 for Professor F. J. Fenner.

The types of trap used at the Field Station allowed small mammals to be caught alive. Local residents trapped most of those from the Mulgrave shire and several from other areas. In the earlier work, an attempt was made to trap areas in which human infections had occurred, generally in or close to canefields.

An outbreak of leptospirosis in dairy cattle was investigated in conjunction with veterinary officers of the Queensland Department of Agriculture and Stock. Additional cattle sera, pig sera, and a few sheep sera were obtained from abattoirs or from veterinarians, and specimens from wild pigs were collected by men on hunting parties.

Methods of examination

Wild animals were given serial numbers, and species, sex, sometimes weight, and usually whether the animal was young or mature, were recorded. The animals were

killed by ether or chloroform, pinned out, and swabbed with a 1:1000 solution of 'Zephiran'. After dissection of the chest wall, heart blood was taken by sterile syringe or Pasteur pipette, and allowed to clot. The abdomen was dissected, and small pieces of kidney cortex were placed in culture media, using fresh sterile instruments for each stage of the dissection.

Kidneys, and sometimes other tissues, were cultured in 2·5 ml. quantities of Schüffner's and Fletcher's media containing 20% rabbit serum dispensed in $\frac{1}{4}$ oz. McCartney bottles. The cultures were incubated at 30° C., and were examined at intervals for 28 days. When leptospires were seen, subcultures were made.

Material for dark-ground examination was obtained by scraping the cut surface of the kidney with a sterile platinum loop, and rubbing up the material in one or two drops of saline on a glass slide. A 22 mm. square coverslip was applied, and the whole area searched for leptospires, using an 8 mm. objective. Freshly voided urine was examined similarly when it was not desired to kill the animal.

Tissues from some wild animals were inoculated into laboratory mice. Quantities of 0·5 or 1 ml. of emulsion of kidney alone, or a mixture of liver, spleen and kidney, were inoculated intraperitoneally into each of a group of two or four mice. Guinea-pigs were occasionally used in addition to mice.

Sera only were obtained from most of the domestic animals. All blood specimens were allowed to clot at room temperature, and, after separation, the serum was stored in the refrigerator until transported to Brisbane.

Serology and identification of strains

Agglutination tests on the sera and identification of the leptospires isolated were carried out at the Laboratory of Microbiology and Pathology, Brisbane, by the methods described by Smith *et al.* (1954) and Smith & Brown (1955); except that the technique of the cross-absorption test was changed during the latter half of the investigation, when formalized suspensions were substituted for suspensions of living organisms, and serum was absorbed for 18 hr. at 30° C. Thirteen serotypes reported from man in North Queensland were used as antigens in the agglutination test: *L. icterohaemorrhagiae*, *L. canicola*, *L. zanoni*, *L. robinsoni*, *L. australis*, *L. bratislava* (= *esposito* of earlier publications), *L. pomona*, *L. grippotyphosa*, *L. medanensis*, *L. kremastos*, *L. mini*, *L. hyos*, and *L. celledoni*. In addition, most of the sera were tested against *L. autumnalis*, and some against *L. bataviae*. *L. cynopteri* was included in the tests on some bat sera.

Interpretation of serological results

Tabulation of the serological results from the animals from which leptospires were isolated showed that most serotypes normally produced a characteristic pattern of serological response which was independent of the host species. However, there was sometimes appreciable overlap, so the unsupported serological findings must be interpreted with caution. Thus, other serotypes sometimes induced production of heterologous antibodies to *L. icterohaemorrhagiae* to such high titres that it would be unwise to identify this serotype on serological evidence alone;

therefore it and its possible hosts are recorded with question marks in the tables. The serological response to infection also fails to differentiate adequately between *L. zanoni* and *L. robinsoni*, between *L. australis* and *L. bratislava*, between *L. medanensis*, *L. kremastos* and *L. mini*, and between *L. canicola*, *L. broomi* and *L. bindjei*. Reactions to these serotypes will therefore usually be referred to as *pyrogenes*, *australis*, *hebdomadis*, and *canicola* serogroups, respectively.

In the absence of proof by isolation or dark-ground observations, titres of 1:100, with normal spread of reactions, have been accepted as reasonable evidence of past infection in wild animals. This policy is conservative, but analysis of the titres recorded in infections proved by isolation of the organism suggests that the incidence of infection was not seriously underestimated. Concurrent high titres to serotypes that do not usually overlap were accepted as evidence of more than one past infection. Antibody patterns in the rodents indicated relatively few multiple infections, but reactions to two or more unrelated serogroups were demonstrated in 28 % of the infected bandicoots.

Interpretation of the antibody patterns in the domestic animals is more difficult. Titres of 1:100 are recorded, but possibly antibody levels only of 1:1000 or higher in cattle and pigs should be accepted as reliable evidence of infection. The scatter of positive reactions at serum dilutions of 1:100 and less, without the clear patterns generally found in the wild animals, may be due to previous leptospiral infections, but it raises the suspicion that the findings may be non-specific. Thus, Babudieri & Gaspardis (1959) suggested that the agglutination reactions found in bovine sera may sometimes be paraspecific, and Schebitz & Dedié (1955) found that non-specific stimuli may stimulate or increase antibody formation against leptospires in horses.

Comparison of survey methods

Table 1 shows the percentage of infected animals according to species in 1498 rodents and bandicoots which were examined by all three methods of kidney culture, dark-ground examination, and serology.

Dark-ground and culture. Overall results show that dark-ground examination was inferior to cultivation for the detection of kidney infections. Of the animals in Table 1, leptospires were seen on dark-ground examination in 53 (3·5 %), and were grown in culture from 74 (5 %). The number of animals showing kidney infections by one or both methods was 86 (5·7 %). Dark-ground examination was especially useful in the series of *R. assimilis*, in which no successful culture was obtained from four of the animals in which leptospires had been observed in the kidneys.

Animal inoculation. Material from relatively few wild animals was inoculated into laboratory mice and guinea-pigs. However, this procedure increased the number of strains isolated from 74 to 77. One strain of *L. celledoni* from *R. assimilis* was isolated from the kidneys of an inoculated laboratory mouse, and two strains of *L. australis* in *R. s. conatus* from a mouse and a guinea-pig respectively. In all three wild animals, leptospires were observed on dark-ground examination of the kidney.

Demonstration of leptospires and serology. Of 1498 animals in Table 1, leptospires

were demonstrated by one or both methods of culture and dark-ground examination in 86 (5·7 %), and antibody titres of 1:100 or more were recorded in 311 (21 %). The total number of animals showing infections, as determined by either demonstration of leptospires or antibody titres of 1:100 or more, was 328 (22 %).

Table 1. *Comparison of survey methods in bandicoots and rodents examined by all three methods*

Species	No. examined	Infection rates determined by: Culture (%)	Dark-ground (%)	Serology (%)	All methods (%)
R. norvegicus	13	61	23	46	69
P. nasuta	48	10	2	60	60
I. macrourus	325	2	0·6	45	45
R. s. conatus	108	18	19	30	33
H. chrysogaster	48	0	0	25	25
M. musculus	130	16	16	9	17
R. assimilis	137	0·7	4	13	14
R. rattus	411	3	0·2	12	12
U. caudimaculatus	26	0	0	8	8
Melomys spp.	252	0·8	0	1	1
Total	1498	5·0	3·5	21	22
			5·7		

Serological results for the 86 animals in which leptospires were demonstrated show that 69 (80 %) had agglutination titres of 1:100 or more. The percentage of serologically positive animals was appreciably different from the total percentage of infected animals in only two species, *R. norvegicus* and *Mus musculus* (Table 1). Five out of eight *R. norvegicus* in which leptospires were demonstrated showed titres of 1:100 or more, one showed a titre of 1:30, and two 1:10 against *L. zanoni*. Eleven out of 21 *M. musculus* excreting *L. zanoni* showed titres of 1:100 or more, nine showed titres of 1:30, and one showed a titre of 1:10 to the homologous serotype. In addition, the following animals in which leptospires were demonstrated showed antibody titres less than 1:100 against the homologous serotype: two *R. s. conatus* with no detectable antibodies to *L. australis*, one *R. s. conatus* with a titre of 1:10 to *L. zanoni*, one *R. assimilis* with a titre of 1:30 to *L. celledoni*, and one *R. assimilis* with none to *L. hyos* (but 1:1000 to *L. pomona*).

INFECTIONS IN MARSUPIALS AND RODENTS

The species examined

Bandicoots and rodents are the most abundant mammals in the area, and they received most attention, the traps used being particularly adapted to capture them. Consequently, there is some bias against the larger or arboreal marsupials, which do not appear to be particularly numerous in the coastal strip, but they too were examined whenever an opportunity arose. Brief notes on the species are given below, and a more detailed account of them can be found in the paper by Harrison (1962), whose nomenclature is followed here.

Marsupials

Antechinus flavipes godmani (Thomas), marsupial mouse. Insectivorous; confined to rain forest.

Perameles nasuta Geoffroy, long-nosed bandicoot. Common in vegetation of all types; an animal about a foot long, which hunts and roots for its insect food, sleeping in a nest of a heap of leaves.

Isoodon macrourus (Gould), short-nosed bandicoot. Similar to *P. nasuta*, but usually confined to cultivated areas or secondary woodland, where it is very numerous.

Trichosurus vulpecula (Kerr), common brush-tailed possum. Arboreal; specimens from open eucalypt forest have been examined, but none of the rain-forest form.

Hypsiprymnodon moschatus Ramsay, musky rat-kangaroo. An insectivorous animal of the floor of rain forest; rather like a bandicoot.

Aepyprymnus rufescens (Gray), rufous rat-kangaroo. A small, kangaroo-like animal of the open eucalypt forest.

Dendrolagus lumholtzi Collett, Lumholtz's tree kangaroo. A rain-forest species adapted for tree climbing.

Thylogale stigmatica Gould, red-legged pademelon or scrub wallaby. A wallaby (i.e. a small kangaroo) of the floor of rain forest.

Protemnodon agilis (Gould), wallaby. An open-country species occurring in the cultivated areas.

Rodents

Hydromys chrysogaster Geoffroy, water rat. A large, carnivorous species associated with watercourses, but ranging far afield.

Melomys cervinipes (Gould), naked-tailed rat. A climbing rat of rain forest and woodland generally.

M. lutillus littoralis (Lönnberg), naked-tailed rat. Very similar to the last, and often difficult to distinguish from it, but smaller, and confined to grassland, becoming a pest of sugar cane.

Uromys caudimaculatus (Krefft), giant naked-tailed rat. A very large, climbing rat of rain forest.

Rattus assimilis (Gould), allied rat. A moderately large, native rat of the floor of rain forest, eating both insects and vegetable matter.

R. sordidus conatus Thomas, canefield rat. A native rat of grassland or open forest; here found almost exclusively in canefields.

R. rattus (Linnaeus), house rat. Common in houses and gardens, and occurring also in cultivated fields and the edges of woodland.

R. norvegicus (Berkenhout), Norway or brown rat. A cosmopolitan house rat; confined in northern Queensland to the centres of coastal towns.

Mus musculus Linn., house mouse. Common in houses, and occurring also in gardens and cultivated fields.

Table 2. *Leptospiral infections in marsupials and rodents—general survey*

Species	No. examined	Total animals infected	Serogroups	No. of infections	
				Total	Urinary
Marsupials					
Perameles nasuta	67	34	*?icterohaemorrhagiae*	1	—
			canicola	1	—
			pyrogenes	10	—
			australis	15	—
			pomona	1	—
			hebdomadis	14	5
			hyos	3	—
			celledoni	4	—
Isoodon macrourus	362	150	*canicola*	29	2
			pyrogenes	56	1
			australis	47	1
			pomona	19	—
			hebdomadis	36	2
			celledoni	4	1
Antechinus flavipes godmani	5	0	—	—	—
Trichosurus vulpecula	15	1	*hebdomadis*	1	—
Aepyprymnus rufescens	6	0	—	—	—
Hypsiprymnodon moschatus	2	0	—	—	—
Dendrolagus lumholtzi	1	0	—	—	—
Thylogale stigmatica	2	1	*grippotyphosa*	1	—
Protemnodon agilis	3	0	—	—	—
Rodents					
Hydromys chrysogaster	68	15	*?icterohaemorrhagiae*	1	—
			pyrogenes	5	—
			australis	7	—
			hyos	3	—
Uromys caudimaculatus	41	3	*pyrogenes*	2	—
			australis	1	—
			hyos	1	—
Melomys cervinipes	80	1	*celledoni*	1	1
Melomys lutillus	248	2	*canicola*	1	1
			australis	1	—
			celledoni	1	—
Rattus sordidus conatus	122	37	*pyrogenes*	5	2
			australis	34	22
Rattus assimilis	157	21	*?icterohaemorrhagiae*	1	—
			pyrogenes	3	—
			australis	1	—
			pomona	12	3
			hyos	6	2
			celledoni	1	1
Rattus rattus	471	51	*?icterohaemorrhagiae*	2	—
			canicola	1	1
			pyrogenes	29	12
			australis	12	—
			pomona	8	—
			grippotyphosa	3	—
Rattus norvegicus	13	9	*pyrogenes*	9	8
Mus musculus	150	22	*pyrogenes*	21	21
			australis	4	1

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Host distribution of the serotypes

It is necessary to distinguish between the broader results that were derived principally from serology, and the more precise ones that were obtained when the leptospires were isolated and identified. The serogroups recorded from the 1813 marsupials and rodents examined in the survey series are set out in Table 2. Leptospiral infections were detected in all nine species of rodents and the two bandicoots in sufficient numbers to indicate that these small, predominantly terrestrial, fossicking animals were reservoirs of at least some representatives of all the major serogroups known from North Queensland. Antibodies were found also in two other marsupials (at 1:300 to the *hebdomadis* serogroup in a *Trichosurus vulpecula* from the Atherton Tableland, and at 1:100 to *L. grippotyphosa* in a *Thyogale stigmatica* from near Innisfail), but their significance is not so clear.

Table 3. *Leptospires isolated from bandicoots and rodents*

Serotypes	Numbers of strains	Hosts
L. broomi	3	*I. macrourus* (2) *R. rattus* (1)
L. bindjei	1	*M. lutillus* (1)
L. zanoni	47	*I. macrourus* (3) *M. lutillus* (1) *R. s. conatus* (2) *R. assimilis* (2) *R. rattus* (12) *R. norvegicus* (7) *M. musculus* (20)
L. robinsoni	2	*U. caudimaculatus* (1) *R. s. conatus* (1)
L. australis	39	*P. nasuta* (2) *I. macrourus* (1) *R. assimilis* (5) *R. s. conatus* (29) *R. rattus* (1) *M. musculus* (1)
L. kremastos	5	*P. nasuta* (4) *I. macrourus* (1)
L. mini	1	*I. macrourus* (1)
L. hyos	4	*U. caudimaculatus* (1) *R. assimilis* (3)
L. celledoni	3	*I. macrourus* (1) *M. cervinipes* (1) *R. assimilis* (1)

The information about the 105 strains of leptospires that have been identified is given in Table 3, which is set out differently from Table 2, because it includes isolations made in the mark–recapture experiments as well as from the survey series. There is also a series of urinary infections in *R. assimilis* from which the organisms have not been isolated. Most of these were encountered in a mark–

recapture experiment in rain forest. Sera from the rats reacted strongly with *L. pomona*, and the organisms from the urine of several were transmitted to laboratory mice, which also developed antibodies to *L. pomona*, but all attempts to recover them in culture have failed. Presumably they are either a mutant, which has lost its capacity to grow in Schüffner's and Fletcher's media, or a closely related serotype.

Most of the identifications of the commoner serotypes were from the survey series, so it follows that the infections recorded in Table 2 on serological grounds as *pyrogenes* serogroup would have been caused predominantly by *L. zanoni*, and those recorded as *australis* serogroup predominantly by *L. australis*. The hosts of *L. broomi* are also indicated fairly clearly by the isolations. The *pomona*, *grippotyphosa*, *hyos*, and *celledoni* serogroups are each represented in Australia by a single serotype, so reactions to them may be accepted as specific, with the reservation that there may still be undiscovered variants or near relatives.

The errors introduced into the analysis by accepting these seven specific identifications seem likely to be small, but the remaining serogroups cannot be treated in the same way. Comparison of our findings with Derrick's (1956) analysis of frequency of occurrence and association with canefields suggests that *I. macrourus* may prove to be a better host for *L. mini* than *P. nasuta*, and that *U. caudimaculatus* and *R. s. conatus* are probably not the only hosts of *L. robinsoni*. It may be inferred too, by exclusion, that bandicoots are probably the hosts of *L. canicola*. We have only the sketchiest indication of possible hosts of *L. icterohaemorrhagiae*, and none at all of those of *L. bratislava*.

Thus, there is evidence that nearly all of the serotypes that have been isolated from man in the area occur also in this section of the animal population. Conversely, no serotype that is not known from the human population was recovered from the animals. Few serological tests were done with representatives of serogroups that have not yet been recorded in Australia, but cultures were made from more than 1600 of the animals in this series, so any undetected serotypes would have been uncommon or ill-adapted to grow on the media used.

Multiple infections

The numbers of infections recorded in Table 2 exceed the numbers of infected animals, reflecting the fact that some animals had had more than one infection. It is difficult to recognize concurrent infections with two or more serotypes (Smith & Doherty, 1956; Alexander *et al.* 1957). Two (both with *L. australis* + *L. hyos* in *R. assimilis*) have been demonstrated by isolation in the mark–recapture experiment referred to above, but none in the survey series, and it is not possible to indicate whether the infections with different serogroups recorded on serological grounds in the following animals were concurrent or successive:

In 13 *P. nasuta:* 1 *?icterohaemorrhagiae* + *australis*, 1 *canicola* + *australis*, 2 *pyrogenes* + *australis*, 1 *pyrogenes* + *hebdomadis*, 1 *pyrogenes* + *celledoni*, 4 *australis* + *hebdomadis*, 1 *australis* + *hyos*, 1 *pyrogenes* + *australis* + *hebdomadis*, 1 *hebdomadis* + *hyos* + *celledoni*.

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In 39 *I. macrourus*: 4 *canicola* + *australis*, 2 *canicola* + *hebdomadis*, 12 *pyrogenes* + *australis*, 2 *pyrogenes* + *pomona*, 10 *pyrogenes* + *hebdomadis*, 2 *australis* + *hebdomadis*, 1 *australis* + *celledoni*, 1 *pomona* + *hebdomadis*, 1 *pomona* + *celledoni*, 2 *hebdomadis* + *celledoni*, 2 *pyrogenes* + *australis* + *hebdomadis*.

In 1 *H. chrysogaster*: *pyrogenes* + *australis*.

In 1 *U. caudimaculatus*: *pyrogenes* + *australis*.

In 1 *M. lutillus*: *canicola* + *celledoni*.

In 3 *R. assimilis*: *pyrogenes* + *pomona*, *australis* + *hyos*, *pomona* + *hyos*.

In 4 *R. rattus*: 3 *pyrogenes* + *australis*, *pomona* + *grippotyphosa*.

In 3 *M. musculus*: *pyrogenes* + *australis*.

The numbers of multiple infections in most species are too small for statistical tests, and all that can be said is that they are of the order to be expected if the chances of infection with different serogroups are independent of one another. With *I. macrourus*, numbers are large enough to show that, while this is true of most multiple infections, the numbers with *pyrogenes* + *australis* serogroups ($\chi^2 = 8{\cdot}4$, $P < 0{\cdot}01$) and with *pyrogenes* + *hebdomadis* serogroups ($\chi^2 = 10{\cdot}0$, $P < 0{\cdot}01$) are significantly in excess of chance expectation, but not the numbers with *australis* + *hebdomadis* serogroups. This is a curious finding that is difficult to interpret; it may be a manifestation of focality.

Maintaining and incidental hosts

Audy (1958) has developed the important concept of maintaining hosts, which ensure the perpetuation of a particular local population of parasites without the intervention of other, incidental hosts. The qualities that distinguish the two classes will be discussed later; we are concerned here only to find out whether they can be recognized in the associations that we have studied.

It is evident, from Table 2, that one species of mammal may be host to more than one serotype, and that individual serotypes may infect more than one species of host. There are, however, some definite associations, which become clearer when the numbers of infections are reduced to rates per 100 animals of the species, and the data are arranged in a form to facilitate comparison, as in Table 4, which includes infections recorded at first examination of mark–recapture animals as well as those in the survey series. The frequency with which leptospires are excreted in the urine is of crucial importance in transmission and maintenance, so this information is included also, isolation from the kidneys being regarded equally with actual detection of leptospires as an indicator of urinary excretion.

The frequency of excretion can be used to provide two different kinds of information, and two different ratios are therefore defined. One is the excretion rate, which is the number of urinary excretors per 100 of the whole population examined; it is shown, for the associations in which it can be calculated, as the left-hand figure in each column in Table 4. The other is the *excretion index*, which is the ratio of the number of animals excreting leptospires to the total number known to have been infected; it is expressed as a decimal fraction rather than a percentage, because a different notation should reduce the risk of confusion between the two ratios.

The most striking association observed was that between the *hebdomadis* sero-

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Table 4. *Frequency of leptospiral infections per hundred animals in the wild populations*

Species	No. examined	(*?icterohaemorrhagiae*)		*canicola* serogroup		*pyrogenes* serogroup		*australis* serogroup		*pomona*		*grippotyphosa*		*hebdomadis* serogroup		*hyos*		*celledoni*	
		L	T	L	T	L	T	L	T	L	T	L	T	L	T	L	T	L	T
P. nasuta	124	—	2	—	0·8	—	12	2	18	—	0·8	—	—	**4**	**16**	—	3	—	3
I. macrourus	485	—	—	0·4	6	0·4	14	0·2	11	—	4	—	—	**0·6**	**8**	—	—	0·2	0·8
H. chrysogaster	92	—	1	—	—	—	8	—	8	—	—	—	—	—	—	—	4	—	—
U. caudimaculatus	83	—	—	—	—	1	5	—	5	—	2	—	—	—	—	1	4	—	—
M. cervinipes	233	—	—	—	—	—	—	—	—	0·5	2	—	—	—	—	—	—	0·5	0·5
M. lutillus	520	—	—	0·2	0·2	0·2	0·6	—	2	—	—	—	—	—	—	—	—	—	0·2
R. s. conatus	433	—	—	—	—	2	4	**12**	**19**	—	—	—	—	—	—	—	—	—	—
R. assimilis	306	—	0·3	—	—	0·3	2	2	4	**4**	**12**	—	—	—	0·6	1	3	0·3	0·3
R. rattus	520	—	0·4	0·2	0·2	**3**	**7**	0·8	5	—	1	—	0·6	—	—	—	—	—	—
R. norvegicus	13	—	—	—	—	**61**	**69**	—	—	—	—	—	—	—	—	—	—	—	—
M. musculus	155	—	—	—	—	**13**	**13**	0·6	2	—	—	—	—	—	—	—	—	—	—

L, Leptospires seen or isolated; T, total infections.
The most clearly defined associations are shown in bold type.

group and the marsupials, particularly the bandicoots, which showed a combined excretion index of 0·14 (609 examined, 59 infected, 8 excreting). No rodents were infected with this serogroup in the survey series, but two *R. assimilis* with titres of 1:100 were encountered in a mark–recapture experiment. Infections with the *canicola* serogroup were also significantly more frequent in the bandicoots than in the rodents ($\chi^2 = 107{\cdot}6$, $P < 0{\cdot}01$). The excretion index was 0·07 in the bandicoots, but it might have been higher for *L. broomi* alone, and the picture is somewhat obscured by the possibility that *L. bindjei* may be maintained by *Melomys*.

The introduced rodents are the principal hosts of *L. zanoni* in the sample, the frequency of excretion of leptospires being significantly higher in them than in any other species ($P < 0{\cdot}01$). Among the native rats, *R. s. conatus* stands out as the principal host of *L. australis*, total and excretion rates being significantly higher than in any other species ($P < 0{\cdot}01$ for both rates), while the excretion index (0·6) is notably high. The figures are, to an extent, inflated by inclusion of a heavily infected series collected during an outbreak of leptospirosis near Babinda (Doherty *et al.* 1956); nevertheless, they confirm an association that has been known since leptospirosis was first recognized in this country. That *R. s. conatus* may not be the only maintaining host of *L. australis* is indicated by the discovery of an apparently self-contained focus of infection in *R. assimilis* in the rain-forest experiment already mentioned. An even clearer rain-forest association, however, is that between *L. pomona* (or a variant) and *R. assimilis*, with an infection rate of 12 % and an excretion index of 0·3.

These associations have all been encountered so much more frequently than would have been expected by chance that the species or groups of animals involved may be presumed, at least provisionally, to be maintaining hosts for the particular serotypes. The situation is not so clear for two other predominantly rain-forest infections. Both *L. hyos* and *L. celledoni* are rather evenly distributed among their host species, and the principal suggestions of maintenance come from their relatively high excretion indices.

Many of the infections included in Table 4 do not fit into the patterns described above. They are scattered among the host species, usually in relatively small numbers, and they generally show a low excretion index, which suggests that most of them are transitory. We would regard these as probably representing casual infections in incidental hosts. Thus, both bandicoots, *R. rattus*, and *R. assimilis* seem to be able to acquire casual infections with most of the serotypes that they do not normally maintain, and the large rats, *H. chrysogaster* and *U. caudimaculatus*, appear to serve as incidental hosts for a more limited range of serotypes rather than as maintaining hosts for any. It is more difficult to decide whether *R. s. conatus* is an incidental or focal maintaining host for *L. zanoni* and *M. musculus* for *L. australis*, and it seems likely that the status of some of the species may vary with local circumstances.

Geographical distribution of infections

Derrick (1956) analysed 219 cases of leptospirosis in man diagnosed in North Queensland, of which two were from the Atherton Tableland and 188 from the

coastal shires of Johnstone, Mulgrave, and Douglas, within which he treated Division 3 of Mulgrave (Fig. 1) as a separate area. The incidence of leptospirosis was significantly higher, and infections with *L. zanoni*, *L. australis*, and *L. hyos* significantly more frequent, in Mulgrave 3 than elsewhere; *L. australis* preponderated significantly over other local serotypes in Mulgrave 3, and *L. zanoni* in Johnstone; and *L. medanensis* was found only in the northern part of the coastal strip, *L. robinsoni* in the southern. We have attempted a similar analysis of the infections listed in Table 2.

Atherton Tableland. This area lies at 2000–3000 ft., has a rainfall less than half that of the coastal strip, and is used mainly for dairying and growing tobacco. Derrick recorded *L. zanoni* and *L. hyos*, and we have found antibodies to *L. hyos* in *U. caudimaculatus* and *R. assimilis*, to *L. celledoni* in *P. nasuta*, and to the *hebdomadis* serogroup in *T. vulpecula*, as well as to *L. pomona* and *L. hyos* in cattle and pigs. All that can be said of the area is that at least five serotypes can survive there.

Table 5. *Relative frequency (per cent) of infections in bandicoots and rodents in the geographical divisions*

Serogroups	Johnstone, 200 infections in 1117 animals	Mulgrave 3, 162 infections in 282 animals	Douglas, 51 infections in 255 animals
icterohaemorrhagiae	?2	?0·5	?2
canicola	0	18	6
pyrogenes	50	22	8
australis	24	41	14
pomona	6	0	53
grippotyphosa	0	0	6
hebdomadis	11	15	8
hyos	5	0·5	0
celledoni	2	3	4

The coastal strip. Dissection of the records showed variations in total incidenc of infection of the same order as those found by Derrick, the relative proportions in the Johnstone, Mulgrave 3, and Douglas areas being 1:3·3:1·2 for the animals and 1:4·4:1·9 for man. The incidence of infection in bandicoots was also significantly higher in Mulgrave 3 than in the other areas ($\chi^2 = 21{\cdot}1$, $P < 0{\cdot}01$), and in *R. s. conatus* significantly higher in Mulgrave 3 than in Johnstone ($\chi^2 = 26{\cdot}2$, $P < 0{\cdot}01$). The relative frequency of the serotypes within each area is shown in Table 5. As in man, the *pyrogenes* group predominated in Johnstone and the *australis* group in Mulgrave 3, and the only notable discordance is a high incidence of *L. pomona* (in *I. macrourus* and *R. rattus*) in Douglas. When the species were examined separately, the only other striking difference observed was in *R. rattus*, which showed *L. zanoni*, *L. australis*, *L. pomona*, and ?*L. icterohaemorrhagiae* infections in Johnstone, *L. broomi* and *L. australis* in Mulgrave 3, and *L. pomona* and *L. grippotyphosa* in Douglas.

Population studies have not been undertaken outside the Johnstone shire, and no reason can be suggested for the differences that have been observed between the

three coastal areas. The points of immediate relevance that emerge from the study are that the infections in man and the animals show a degree of parallelism that is compatible with an association between them, and that the maintaining hosts, at least of *L. zanoni*, may not be the same in all parts of the coastal strip.

Infections in town, field, and forest

Urban infections. Infections in towns can be divorced from those in adjacent canefields only in Cairns (population 21,000) and Innisfail (7000). Derrick recorded 1 infection with *L. celledoni* from Cairns, and 4 with *L. zanoni* from Innisfail. Five *I. macrourus*, 1 *H. chrysogaster*, 21 *R. rattus*, and 1 *M. musculus* were examined from Cairns; all were negative. The Innisfail series comprised 9 *P. nasuta*, 13 *I. macrourus*, 16 *H. chrysogaster*, 1 *M. lutillus*, 3 *R. assimilis*, 104 *R. rattus*, 13 *R. norvegicus*, and 7 *M. musculus*. Evidence of infection with *L. zanoni* was found in 3 *P. nasuta*, 6 *I. macrourus*, 1 *H. chrysogaster*, 9 *R. rattus*, 9 *R. norvegicus*, and 1 *M. musculus*, while 2 *R. rattus* might have been infected with *L. icterohaemorrhagiae*. In one small area along the river bank, infections were found in 8 out of 8 *R. norvegicus* (7 excreting), in 3 out of 25 *R. rattus* (1 excreting), and in 1 *I. macrourus*. No other serotypes were detected in these animals, and thus the area apparently provided a pure focus of infection with *L. zanoni*. It is not surprising that infections have occurred in the human population.

Rural infections. The coastal area is dominated by rain forest on the hills and sugar cane on the plains, and each form of vegetation has its own characteristic fauna, which, excluding stray animals, is listed below (Harrison, 1962).

	Canefields	Rain forest
Marsupials	*I. macrourus* *P. nasuta*, less frequent	*P. nasuta* (3 other species of minor importance)
Rodents	*M. lutillus* *R. s. conatus* *R. rattus* *M. musculus*	*M. cervinipes* *R. assimilis* *U. caudimaculatus*
	H. chrysogaster in wet places anywhere	

Other forms of vegetation, such as secondary forest, grazing land, and swamp, constitute a minor part of the vegetation pattern, and the species found there also inhabit canefields or rain forest, depending on propinquity.

Derrick (1956) examined the relationship of the infections in man to occupation in canefields, and his findings can be summarized in the following statements, omitting *L. icterohaemorrhagiae*, *L. pomona*, and *L. medanensis*, for which there were insufficient data:

Positively associated with canefields: *L. australis*, *L. mini*.

Negatively associated with canefields: *canicola* serogroup.

Without predominance in or away from canefields: *L. zanoni*, *L. robinsoni*, *L. kremastos*, *L. hyos*, *L. celledoni*.

By reason of their wider distribution but more limited range of movement, the small ground mammals are better indicators than infected people of the places in

Table 6. *Infections in rodents and bandicoots from different habitats*

Species	Canefields						Rain forest						Other habitats						
	No. examined	No. of infections: *zanoni*	*robinsoni*	*australis*	*pomona*	*hebdomadis* gp.	*hyos*	No. examined	No. of infections: *zanoni*	*australis*	*pomona*	*hebdomadis* gp.	*hyos*	*celledoni*	No. examined	No. of infections: *?icterohaemorrhagiae*	*zanoni*	*australis*	*pomona*	*hebdomadis* gp.	*hyos*	*celledoni*
P. nasuta	22	4	—	6	—	7	—	13	—	2	1	4	2	1	8	—	1	1	—	1	—	—
I. macrourus	62	12	—	6	—	8	—	0	—	—	—	—	—	—	26	—	2	—	—	2	—	—
H. chrysogaster	21	2	—	4	—	—	1	3	1	—	—	—	—	—	5	—	—	—	—	—	—	—
U. caudimaculatus	0	—	—	—	—	—	—	10	1	1	—	—	—	—	26	—	1	—	—	—	—	—
M. cervinipes	5	—	—	—	—	—	—	48	—	—	—	—	—	1	21	—	—	—	—	—	—	—
M. lutillus	99	—	—	1	—	—	—	0	—	—	—	—	—	—	70	—	—	—	—	—	—	—
R. assimilis	3	—	—	—	—	—	—	86	1	—	10	—	1	—	47	1	2	1	1	—	4	1
R. s. conatus	83	4	1	11	—	—	—	0	—	—	—	—	—	—	3	—	—	—	—	—	—	—
R. rattus	174	18	—	11	1	—	—	0	—	—	—	—	—	—	43	—	2	—	—	—	—	—
M. musculus	127	20	—	4	—	—	—	0	—	—	—	—	—	—	6	—	—	—	—	—	—	—

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which populations of leptospires are maintained. The records for the Johnstone shire are divided according to habitat in Table 6, from which it is apparent that there is a wider range of serotypes in each of the rural habitats than in the urban infections recorded earlier, and that the 'other habitats' served, in general, as areas of overflow from the canefields or rain forest for the leptospires as well as for the mammals.

L. zanoni (60 infections), *L. australis* (43), and *hebdomadis* serogroup (15) were the commonest leptospires in the canefields, as is also indicated by the records of their occurrence on eight widely scattered cane farms in the district, namely: *L. australis* in 8, *L. zanoni* in 6, *hebdomadis* serogroup in 5. When the figures were corrected for uneven sampling, it was clear that *R. s. conatus* predominated among the ground rats and *L. australis* among the serotypes on most of the farms sampled; but both the populations of particular species and the incidence of infection varied considerably, and Table 7 illustrates a situation in which *M. musculus* and *L. zanoni* had been unusually abundant. It is also evident, from all the data, that *L. zanoni* was more strongly associated with canefields in this district than Derrick's figures would have suggested.

Table 7. *Infections found on one cane farm*

Species	No. examined	No. infected	Serotypes	No. of infections: Urinary	No. of infections: Total
I. macrourus	13	5	*zanoni*	—	2
			australis	—	3
			mini or *kremastos*	—	1
R. s. conatus	26	7	*zanoni*	1	1
			australis	4	6
R. rattus	41	2	*zanoni*	—	1
			australis	—	1
M. musculus	92	17	*zanoni*	16	16
			australis	1	3
Melomys spp.	4	0	—	—	—

L. pomona (11 infections) and the *hebdomadis* serogroup (4), associated respectively with *R. assimilis* and *P. nasuta*, were the commonest leptospires in the rain-forest animals. A similar occurrence of *L. hyos* and *L. celledoni*, with *H. chrysogaster* and *M. cervinipes* added to the hosts, is less apparent, because 6 of the infections were in animals collected in gallery forest along small water courses, and they consequently appear under 'other habitats' in Table 6. The mark–recapture experiment to which several references have already been made has shown that a focus of infection with *L. pomona*, and probably *L. australis* and *L. hyos*, was being maintained in rain forest by *R. assimilis*, while the *hebdomadis* serogroup was being maintained by *P. nasuta*, which was also serving as an incidental host of other serotypes. In this experiment, 40 out of 103 *R. assimilis* were found to be infected at first examination, an incidence that would rival that in *R. s. conatus* in heavily infected canefields.

OTHER WILD ANIMALS EXAMINED

Leptospires have been isolated from bats in Indonesia (Alston & Broom, 1958) and aquatic birds in Italy (Babudieri, 1958), and serological evidence of infection has been found in reptiles in Roumania (Combiesco *et al.* 1959) and Malaya (Gordon Smith, Turner, Harrison & Broom, 1961*a*), including a high incidence in the file snake (*Acrochordus javanicus*), which occurs also in Australia. A systematic survey of these groups was not attempted in the present study, but any individuals that were collected were examined. Sera were tested from nearly all, but dark-ground examinations and cultures from only about half.

Table 8. *Wild animals, other than marsupials and rodents, examined for leptospiral infections*

Species	No. exmd.	Species	No. exmd.
Monotremes		Birds (*cont.*)	
Ornithorhynchus anatinus (Shaw)	1	*Centropus phasianinus* (Latham)	4
Tachyglossus aculeatus (Shaw)	4	*Pitta versicolor* Swainson	1
Bats		*Cracticus quoyi* (Lesson & Garnot)	12
Pteropus conspicillatus Gould	33*	*Acridotheres tristis* Linnaeus	1
Pteropus alecto gouldii Peters	1	Reptiles	
Rhinolophus megaphyllus Gray	12	*Liasis childreni* Gray	1
Hipposideros bicolor (Temminck)	5	*Boiga fusca* (Gray)	3
Nyctinomus loriae Thomas	1	*Acrochordus granulatus* (Schneider)	2
Miniopterus schreibersi (Kuhl)	8	*Stegonotus plumbeus* (Macleay)	1
Miniopterus australis Tomes	5	*Natrix mairii* (Gray)	1
Nyctophilus bifax Thomas	2	*Oxyuranus scutellatus* Peters	1
Birds		*Denisonia* sp.	1
Rallus pectoralis Temminck	1	*Laticauda* sp.	1
Hypotaenidia philippensis (Linnaeus)	3	*Varanus tristis orientalis* Fry	8
Rallina tricolor Gray	2	*Tiliqua scincoides* (Shaw)	2
Amaurornis ruficrissus (Gould)	1	*Tiliqua gerrardii* (Gray)	2
Lobibyx sp.	1	*Egernia major* (Gray)	3
Nettapus pulchellus Gould	1	*Emydura latisternum* (Gray)	1
Accipiter fasciatus (Vigors & Horsfield)	1	*Crocodilus johnsoni* Krefft	1
Podargus strigoides (Latham)	1	Amphibians	
Halcyon macleayi Jardine & Selby	1	*Bufo marinus* (Linnaeus)	21

* Antibodies at 1:100 or more to *L. australis* in 6.

The species examined are listed in Table 8. Of the monotremes, the platypus (*O. anatinus*) is confined to rivers and burrows in their banks, and the echidna is terrestrial, itinerant, and lives on ants. The first two bats listed are fruit-bats or 'flying-foxes'; the others are all insectivorous, and some of them have a wide distribution outside Australia. The birds are a miscellaneous lot, some of which were caught in traps set for the small mammals. The reptiles include representatives of four families of snakes, two lizards, one freshwater tortoise, and one freshwater crocodile. The toads (*B. marinus*) were introduced into Queensland to control sugar-cane grubs, and are now extremely abundant.

No leptospires were isolated from any of these animals, and antibodies were detected only in the flying-foxes (*P. conspicillatus*).

INFECTIONS IN DOMESTIC MAMMALS

The coastal plain in North Queensland, in which nearly all the human cases of leptospirosis originated, and from which most of the wild animals in the survey were obtained, carries relatively few domestic stock. Dairy farming is, however, a major industry on the slopes to the west of this area, on the Atherton Tableland, and in the north of the Douglas shire (Fig. 1). Pigs are also raised on many of the farms. Beef cattle predominate in the drier areas of the Atherton Tableland and west to the Gulf of Carpentaria. Domestic dogs and cats are everywhere.

These animals are relevant to the present investigation in two ways: they may be primary reservoirs, as is well known for *L. canicola* in dogs in other parts of the world (Alston & Broom, 1958) and for *L. pomona* and *L. hyos* in pigs and cattle in southern Queensland (Johnson, 1950); or they may serve as incidental hosts, providing a possible link in a chain of infection from wild animals to man. Infections in cattle may also present a significant economic problem in Queensland (Sutherland, Simmons & Kenny, 1952), and an episode of this kind is described below.

Leptospirosis in a dairy herd

An outbreak of illness and abortions occurred in a dairy herd in the Palmerston section of the Johnstone shire, and veterinary officers made a provisional diagnosis of leptospirosis. Blood samples were taken from thirty-six cows, one calf, and one horse, and urine samples from two cows and the calf. At that time, six of the seven cows which had not been in calf when the outbreak began had been ill, sixteen of the others had aborted, and thirteen were still pregnant. Subsequently, six of the pregnant cows aborted and five calved (with the death of one calf, apparently from leptospirosis); the histories of the other two were not followed. Eighteen of the cows had antibodies to *L. pomona* at titres of 1:1000 or more, four had titres of 1:300 or 1:100, four 1:30, and two 1:10. Leptospires were observed in the urine of one cow which had an antibody titre of 1:3000. The calf had been very ill: leptospires were observed in its urine, and its serum had a titre of 1:30,000 to *L. pomona*.

The horse showed titres of 1:100 or more to the following serotypes: *L. icterohaemorrhagiae* 1:300, *L. australis* 1:300, *L. bratislava* 1:10,000, *L. pomona* 1:30,000, *L. grippotyphosa* 1:3000, *L. kremastos* 1:100, *L. mini* 1:100, *L. autumnalis* 1:3000. The significance of this result is not clear. One other horse, several dogs, and the pigs on the farm were not investigated.

Nine months later, blood was taken from ten of the cows previously tested, of which five showed negligible changes in antibody titre, four had gained antibody, and one remained negative. The antibody titre in the calf had fallen from 1:30,000 to 1:10,000. Of four animals then bled for the first time, one had been in the herd at the time of the outbreak, and had been ill; the other three had been introduced after the outbreak had subsided. All showed titres of 1:1000 or more against *L. pomona*. No sickness had been noticed in the introduced animals, and they may

have been immune when purchased, or have acquired inapparent infections on the farm.

The origin of the outbreak is uncertain. No illness had been noted in the pigs, but the drainage from the pig pen ran over land accessible to the cattle. The farmer had allowed cattle from another herd to graze on his land before the outbreak.

None of the five persons on the farm suffered from clinical leptospirosis at that time, and the owner's serum did not react with *L. pomona*. Nevertheless, as with the cane farm in Table 7, the experience indicates strikingly the weight of infection to which particular groups of rural workers may sometimes be exposed.

The other side of the epidemiological picture is illustrated by the fact that antibodies (alone, or in addition to antibodies against *L. pomona* and the serotypes which cross-react with it) were detected to *H. hyos* in two of the cows, to *L. celledoni* in six, and to the *hebdomadis* serogroup in nine. Two *H. chrysogaster*, 1 *U. caudimaculatus*, 18 *M. lutillus*, 12 *R. assimilis*, 1 *R. rattus*, and 5 *M. musculus* (all included in Table 2) were trapped on the farm in areas to which the cattle had access. Antibodies to *L. hyos* were found in 1 *H. chrysogaster* and 2 *R. assimilis*, 1 of which had leptospires in the kidney, and *L. celledoni* was isolated from 1 *R. assimilis*. No bandicoots were trapped, although the owner of the farm stated that they were numerous. The association between the domestic stock and the small mammals was close, and it seems likely that there had been some exchange of infections between them.

General survey

The results of agglutination–lysis tests on the sera of 253 domestic mammals are set out in Table 9. The discrepancy between the numbers of animals reacting and the numbers with titres to the individual serogroups is accounted for by multiple infections. Except for one dog, cultures were not made from animals in this series.

Cattle. Excluding the 41 dairy cattle discussed above, sera were tested from beef and dairy cattle from widely scattered areas in North Queensland, including other localities in Johnstone shire (16), Atherton Tableland (47), Daintree (35), Charters Towers (24), Ingham (12) and Hughenden (3). Sixty showed titres of 1:100 or more, *hyos* (29), *australis* (16), and *pomona* (12) being the commonest serogroups, although some of the titres to the *australis* serogroup were probably heterologous reactions from *pomona* infections. One animal showed titres of 1:1000 to both *pomona* and *hyos*. Reactions were also obtained with all other serogroups used except *hebdomadis*, which, however, had been recorded in the Palmerston cattle. Twenty-six of 35 sera from twelve Daintree dairy herds contained antibodies at titres of 1:100 or more, including 14 with antibodies to *hyos*, but none to *pomona*.

Pigs. Twenty out of forty domestic pigs and five out of fourteen wild pigs showed agglutinins at titres of 1:100 or more to five serogroups. Pig sera collected at Innisfail from Atherton Tableland animals showed evidence of infections with *L. pomona* and *L. hyos*. The Douglas shire animals (including one wild pig) had antibodies to *icterohaemorrhagiae*, *pyrogenes*, and *australis* serogroups.

Thus, *L. pomona* and *L. hyos* are present in cattle and pigs on the Atherton Tableland and adjoining areas carrying domestic stock. Sutherland *et al.* (1952)

Table 9. *Serological survey of domestic animals*

Species and no. tested	Animals reacting		Numbers reacting with serogroups									
	Numbers	At titres of	*icterohaemorrhagiae*	*canicola*	*pyrogenes*	*australis*	*pomona*	*grippotyphosa*	*hebdomadis*	*hyos*	*celledoni*	*autumnalis*
Cattle, 137	30	1:100	5	1	6	11	2	2	—	13	1	2
	18	1:300	—	—	—	4	3	—	—	12	—	—
	9	1:1000	—	—	—	1	5	—	—	4	—	—
	3	1:3000 or more	1	—	—	—	2	—	—	—	—	—
Pigs, 54	9	1:100	—	—	2	6	2	—	—	—	—	—
	4	1:300	2	—	2	—	—	—	—	—	—	—
	6	1:1000	2	—	—	—	3	—	—	1	—	—
	6	1:3000 or more	—	—	—	2	4	—	—	—	—	—
Sheep, 7	2	1:100	—	—	1	1	1	—	—	—	—	—
	1	1:300	—	—	—	—	1	—	—	—	—	—
Dogs, 34	11	1:100	—	—	3	3	1	—	4	—	—	—
	2	1:300	—	1	—	1	—	—	—	—	—	—
	2	1:1000	—	—	—	1	—	—	—	1	—	—
Cats, 21	—	1:100	—	—	—	—	—	—	—	—	—	—
	1	1:300	—	—	—	—	1	—	—	—	—	—

had reported *pomona* leptospirosis from all dairying areas of the state except the Atherton Tableland. The results suggest also that cattle and pigs may become infected with serotypes belonging to several other serogroups. The animals were not reported as ill, and these serotypes have not been associated with disease in stock in Australia, although antibodies to *icterohaemorrhagiae* and *grippotyphosa* serogroups in both pigs and cattle and to *hebdomadis* serogroup in cattle have been recorded (Wannan, 1955; Forbes, Keast, Wannan & Lawrence, 1955).

Sheep. The wet coastal belt does not carry sheep. The seven sera in Table 9 were collected at an Innisfail slaughter-house from sheep brought from Hughenden in the drier western part of North Queensland. Two reacted with *L. pomona*, at titres of 1:100 and 1:300, and one with *L. zanoni* at 1:100. The significance of these results is uncertain. Leptospiral infections in sheep have not been recorded from Queensland, although they have been reported from other Australian states (Wannan, 1955) and overseas countries (Alston & Broom, 1958).

Dogs. Gray (1940, 1942) has described canine leptospirosis in southern Queensland, but it is seldom recognized by veterinarians in the north. None of the dogs reported here showed signs of sickness, and all but one were contacts of human cases of leptospirosis. One had antibodies to the *canicola* serogroup, but was not associated with a human case. Only three of the fifteen dogs which reacted to a titre of 1:100 or more had antibodies to the serotype that was responsible for the human infection in the household: one of three belonging to a patient with a *medanensis* infection; one at the home of a child with a *zanoni* infection; and one of three at the home of another child with a *zanoni* infection (but one of the other dogs had antibodies at 1:1000 to *L. hyos*). A higher correlation would have been expected, if dogs had been reservoirs, or had formed a significant link in the chain of infection from the small ground mammals to man.

Cats. Cats are common domestic pets in the area. The 21 in Table 9 had been household pets in or near Innisfail, and the sources of the *pomona* infection in one of them can only be guessed. Domestic cats have become feral in the canefields, but this population has not been sampled.

REVIEW OF THE SEROTYPES

It will be convenient, at this point, to review briefly what is known of the status and hosts of the serotypes that occur in North Queensland, as a background to a discussion of their ecology. The findings to date, including, for completeness, those of Battey *et al.* (1964), are epitomized in Table 10.

Leptospira icterohaemorrhagiae

This serotype is present in southern Queensland, but it is not certain that it occurs in the north. Of ten infections in man recorded as *icterohaemorrhagiae* by Derrick (1957), only two were diagnosed from cultures, and absorption tests to exclude other members of the group were not being undertaken at that time. Moreover, the occupational diversity of the cases, including some in cane farmers and timber getters, and the restricted distribution of *R. norvegicus* in the area,

Table 10. *Summary of hosts of leptospiral serotypes in North Queensland*

Serogroups	Serotypes	Relative frequency in man (%)*	Probable maintaining hosts	Probable incidental hosts
Icterohaemorrhagiae	*?icterohaemorrhagiae*	3	?	*?Perameles nasuta*, *?Hydromys chrysogaster*, *?Rattus assimilis*, *?R. rattus*, ?cattle, ?pigs
Canicola	*canicola* *broomi* *bindjei*	4·5	*?Isoodon macrourus* *I. macrourus* *?Melomys* spp.†	*P. nasuta*, *Uromys caudimaculatus*†, *R. rattus*, dogs
Pyrogenes	*zanoni*	26	*R. rattus*, *R. norvegicus*, *Mus musculus*, *?R. s. conatus* (focal)	*P. nasuta*, *I. macrourus*, *H. chrysogaster*, *U. caudimaculatus*, *Melomys lutillus*, *M. cervinipes*†, *R. s. conatus*, *R. assimilis*, dogs
	robinsoni	6	*?U. caudimaculatus*, *?R. s. conatus*	?
Australis	*australis*	29	*R. s. conatus* (in canefields), *R. assimilis* (in rain forest)	*P. nasuta*, *I. macrourus*, *Hypsiprymnodon moschatus*, *H. chrysogaster*, *U. caudimaculatus*, *M. cervinipes*, *M. lutillus*, *R. rattus*, *M. musculus*, *Pteropus conspicillatus*, cattle, pigs, dogs
	bratislava	1	?	?
Pomona	*pomona*	2	Pigs, cattle, *R. assimilis* (in rain forest)	*P. nasuta*, *I. macrourus*, *U. caudimaculatus*†, *M. cervinipes*, *R. s. conatus*†, *R. rattus*, dogs, cats
Grippotyphosa	*grippotyphosa*	0·3	*R. s. conatus*†	*Thylogale stigmatica*, *P. nasuta*†, *I. macrourus*†, *U. caudimaculatus*†, *R. rattus*
Hebdomadis	*kremastos* *mini* *medanensis*	12 3 1	*P. nasuta*, *I. macrourus* *I. macrourus* *P. nasuta*†, *I. macrourus*†	*Trichosurus vulpecula*, *R. assimilis*, cattle, dogs
Hyos	*hyos*	6	Cattle, pigs, *?R. assimilis* (in rain forest)	*P. nasuta*, *I. macrourus*†, *H. chrysogaster*, *U. caudimaculatus*, dogs
Celledoni	*celledoni*	6	*?P. nasuta*, *?I. macrourus*, *?M. cervinipes*, *?R. assimilis*	*H. chrysogaster*†, *M. lutillus*, cattle

* From Derrick's (1957) analysis of 382 cases.
† From Battey *et al.* (1964).

indicate that the local epidemiology differs considerably from that of classical Weil's disease.

L. canicola

Only two infections in man have been identified since the serotype was reported from Queensland by Sinnamon *et al.* (1953); both were in the Mulgrave shire. There is no evident association with dogs, and the maintaining hosts are either bandicoots or not among the species investigated.

L. broomi

This has been the most frequent serotype in the *canicola* serogroup infections of man in the area, and it has been isolated also from *I. macrourus* and *R. rattus*. The high incidence of serogroup antibodies in the bandicoot would suggest that it is the maintaining host. However, the *canicola* serogroup cases studied by Derrick (1956) showed a significant preponderance ($P < 0{\cdot}05$) away from canefield occupations, so other reservoirs may also be involved.

L. bindjei

The occurrence of this serotype was not recognized before our work was completed, but it is now known to have produced several human infections in the Douglas shire, including one on the cane farm where it was isolated from *M. lutillus*. It has since been recovered also from *M. cervinipes* in the same district (Battey *et al.* 1964).

L. zanoni

Human infections with this common serotype were widespread in the area, with a significant predominance over other serotypes in the Johnstone shire, and an almost equal incidence in and away from canefields; it also caused urban leptospirosis in Innisfail. The introduced rodents are the maintaining hosts, but it has been recorded from many other species, although it appears to intrude but little into the rain forest.

L. robinsoni

Infections in man have been limited almost entirely to the southern half of the area, and were equally distributed in and away from canefields. It has been isolated from *U. caudimaculatus* and *R. s. conatus*, and may have contributed to the '*pyrogenes*' antibodies found in other species.

L. australis

This serotype is the cause of classical canefield fever in North Queensland. It is widespread, with a significantly high incidence in the Mulgrave 3 division, and a highly significant association both with canefields and with *R. s. conatus*. It is highly pathogenic to laboratory mice (Emanuel, 1959); but field experience has shown that it can produce benign infections in a variety of hosts, and *R. assimilis* appears to be able to maintain foci of infection in rain forest.

L. bratislava

This serotype was isolated from a cane-cutter and from cane trash at Babinda. It was named *L. esposito* by Smith & Brown (1955), but was subsequently found by Wolff & Bohlander (1961) to be serologically identical with *L. bratislava*. Owing to its cross-agglutination pattern, it can be identified only when isolated in culture, and its animal hosts are still unknown.

L. pomona

Pigs appear to be the primary hosts in southern Queensland, with a considerable overflow into cattle, in which it may cause serious clinical disease, and from both pigs and cattle into people who have contact with them. The same sequence occurs in North Queensland, but there is an appreciable amount of infection in the small ground mammals as well. A variant has been found in *R. assimilis*, and it is not yet clear how far it may contribute to infections in other species, including man.

L. grippotyphosa

This serotype was isolated from a cane-cutter in the Douglas shire (Smith & Brown, 1955), and a second infection has recently been identified serologically in a soil chemist who was working in Brisbane with samples of soil from the same district (Tonge & Smith, 1961). Antibodies were found in 3 *R. rattus* from the same area as the human cases and in one *Thylogale stigmatica* from near Innisfail, but it now seems probable that *R. s. conatus* is the maintaining host in the Douglas shire (Battey *et al.* 1964).

L. kremastos

Originally isolated from a dental nurse whose home was on a cane farm near Babinda, *L. kremastos* was subsequently found to be more prevalent in human infections in the Johnstone shire, and to be almost equally distributed in and away from canefields. The reservoirs are marsupials, and it has been isolated from both species of bandicoot.

L. mini

In contrast with *L. kremastos*, *L. mini* was first isolated from an Innisfail hospital worker, and then found to be more prevalent in Mulgrave 3 and significantly associated with canefields. It has been isolated from *I. macrourus*, but it is not known whether it may occur in other species.

L. medanensis

This serotype has been recorded so far only from five patients in the Douglas shire. It was not recovered from animals, and significant titres to it were not recorded from local bandicoots, but it has subsequently been isolated from both species (Battey *et al.* 1964).

Of the infections identified only as *hebdomadis* serogroup, the habitats of the animals suggest that those in *R. assimilis* in rain forest and in the Palmerston dairy

cattle were more likely to have been caused by *L. kremastos* than *L. mini*, but those in dogs on the more southern cane farms could have been due equally to either. The serum of the possum (*T. vulpecula*) from the Atherton Tableland (and of an unidentified marsupial from the same area, not included in Table 2) gave a significant titre only to *L. medanensis*, as did that of a dog belonging to a patient with *medanensis* leptospirosis.

L. hyos

The epidemiology of *hyos* infection in man is similar to that of *L. pomona*, occupational contact with cattle and pigs generally being an essential feature. However, some of the northern cases were in forest workers. There was no clear indication of infection in the thirteen wild pigs examined, but an appreciable incidence in *P. nasuta*, *U. caudimaculatus*, *R. assimilis*, and the water rat, *H. chrysogaster*. There is reason to believe that this serotype, like the variant of *L. pomona*, can be maintained in rain-forest mammals not in contact with cattle, and it may be significant that the strains isolated from these animals show distinguishing subserotype characteristics.

L. celledoni

This distinctive serotype was described by Smith *et al.* (1954), and it proved to be widespread in the area, twenty-four human infections being recorded, including the only case of urban leptospirosis in Cairns. It seems to be associated primarily with forest animals, but it has also been found in *I. macrourus* and *M. lutillus* in canefields, and it may infect cattle.

DISCUSSION

The concept of principal and incidental hosts (at least so far as man is concerned) has been inherent in the definition of zoonoses, but Audy (1958) gave it precision by introducing the term 'maintaining hosts', and drawing a clear distinction between maintaining and incidental hosts in the animal populations. We have inferred that there are particular species of mammals on which the populations of the leptospiral serotypes in North Queensland depend for survival, and the most important questions to answer, from both practical and theoretical points of view, are whether this is true, and, if it is, how these maintenance associations are to be recognized, and what conditions influence their stability and effectiveness.

It is a basic principle in any study of the interactions between hosts and parasites that no two host–parasite associations are precisely alike. Whether they differ so little as not to affect a particular inquiry, or so much as to be fundamental to it, can be determined only by observation. It follows that we should examine each of the associations separately, and it is a defect in the work reported here—as it is also in the Malayan studies of Gordon Smith *et al.* (1961*a*, *b*)—that that cannot be done with any real precision. The species of hosts can be stated precisely, but not those of the leptospires. There are two reasons for this. One is that, although the serotype is treated as a species for purposes of nomenclature, many serotypes have not yet been shown to be stable genetic entities. The other is that, as it is impractic-

able to identify serological reactions to infection beyond serogroup, the identity of an infecting serotype can often only be guessed. This introduces an unfortunate element of vagueness into a situation that is already inherently complex by reason of the numbers of serotypes and host species involved. These weaknesses must limit the precision, though not necessarily the nature, of the conclusions that can be drawn.

Gordon Smith *et al.* (1961*a*) defined the requirements for survival of populations of leptospires, and we may present them in a somewhat different way here, in order to define the ecological problems more clearly. They are:

(1) A host–parasite interaction that provides opportunities for spread of leptospires in the host population without significantly reducing its level and activity.

(2) An independent set of favourable properties of the hosts, including habits, habitats, longevities, ranges, and population densities.

(3) Another independent set of external factors favouring survival (and perhaps multiplication) of leptospires outside the bodies of the hosts.

We are concerned, in this paper, primarily with the first.

Babudieri (1958) has given a clear statement of the basic relationship between host and parasite that characterizes a maintenance association. After describing the course of an infection, from early leptospiraemia to later lodgement in the kidneys, and, *in some infections only*, development of dense colonies of leptospires in the convoluted tubules whence the organisms are shed into the urine, he goes on: '. . . a state of biological equilibrium is easily established between some serotypes of leptospires and some animal species. This equilibrium is either lacking or much more difficult to establish in other cases. Both components of this symbiotic association play a part in this phenomenon. In fact, it is observed that, while certain animals easily become carriers of some serotypes of *Leptospira*, they do not become so for others. This confirms anew the biological validity, discussed by some investigators, of the subdivision of *Leptospira* into serotypes; at the same time it demonstrates the fact that the distinction made initially between temporary shedders and persistent carriers, although unconventional, is one that rests on a solid if poorly explicable biological basis. Any animal susceptible to infection by leptospires may become a temporary shedder, but only those animal species that present a particular condition of biological sympathy for a determined serotype of *Leptospira* can become carriers.'

Our approach has been somewhat different, and it provides a link between Babudieri's account of what happens in the host and what may happen in the field. Emanuel (1959) inoculated laboratory mice with blood-clot from patients with acute leptospirosis, and found that the resulting infections formed a series which could be divided conveniently into three grades:

I. Severe, with obvious illness and high mortality, the survivors becoming chronic urinary carriers (*L. australis*, *L. grippotyphosa*).

II. Moderate to mild, with few clinical signs, usually ending in recovery and continued urinary excretion of leptospires (*L. icterohaemorrhagiae*, *L. broomi* (recorded as '*canicola*'), *L. zanoni*, *L. robinsoni*, *L. bratislava*, *L. hyos*).

III. Inapparent, with no clinical signs, often transient, but mice sometimes

becoming urinary carriers (*L. pomona*, *L. kremastos*, *L. mini*, *L. medanensis*, *L. celledoni*).

There was some difference between strains, but the general pattern was reasonably constant, and the following strains subsequently recovered from animals have been found to conform to it: 25 *L. australis*, 8 *L. zanoni*, 1 *L. robinsoni*, 2 *L. hyos*, 11 *L. ?pomona*, 1 *hebdomadis* serogroup and 2 *L. celledoni*, while 1 *L. bindjei* produced similar grade II infections to *L. broomi*.

We may, then, visualize the mouse as a 'good' host for the serotypes in grade II. Those in grade I might survive as local populations (but probably with marked epizootic fluctuations if they did), while those in grade III, although not harming the host, are unlikely to reach the exterior in sufficient numbers to ensure maintenance of the population.

It does not follow that infections by these serotypes would produce the same pattern of response in other species of mammals. Indeed, as Babudieri showed in his review, there is evidence that they do not. Man is a case in point, human infections in Queensland differing both in gradation of severity and types of reaction from those in the mouse. In the animals, *L. zanoni* produces moderate to mild infections in mice, but three strains from patients all produced severe reactions in guinea-pigs. A culture of *L. zanoni* obtained from a wild *M. musculus* was inoculated into four laboratory mice, one *Melomys lutillus*, one *M. cervinipes* and one *I. macrourus*. It produced inapparent infections in all, but urinary carriers only in the mice. On the other hand, *L. bindjei* produced urinary carriers in 2 *M. cervinipes*, 1 *M. lutillus*, and at least six of the eight laboratory mice into which it was inoculated. Similar results have been obtained in other parts of the world. Thus, Walch-Sorgdrager (1939) showed experimentally that *L. icterohaemorrhagiae* regularly produced persistent urinary infections in *R. norvegicus*, whereas *L. canicola* produced only transitory infections, with rare, brief excretion of leptospires. Conversely, Packchanian (1940) demonstrated that different species of rodents differed markedly in their susceptibility to experimental infection with *L. icterohaemorrhagiae*, while Stavitsky & Green (1945) and Neghme, Christen, Jarpa & Agosin (1951) found that different races of a host species differed in their susceptibility to infection with a single strain of *Leptospira*.

We would, then, expect the situation in the field to present a mosaic of host–parasite relationships, with the order of the serotypes in the graded sequences differing from species to species of the host range.

It would have been desirable to test this hypothesis by experimental infections in the wild species, but that has not been possible for two reasons. One is that most of the serotypes studied lose their adaptations to parasitic life so quickly when passaged in culture that it is necessary to work directly from the original hosts, which means blind inoculation and a continuous supply of experimental hosts. The other is that adequate numbers of native animals reared in the laboratory have not been available. It has, consequently, been necessary to seek for an indirect method of evaluation, and we have placed most reliance on the excretion index for this purpose, because it would be less influenced by external variables than the overall infection or excretion rates.

Sick animals are not likely to enter traps, so the method of collecting would be biased against detecting grade I infections. Survivors would have a high excretion index, but the overall infection rate would be low, unless there were an epizootic, and no indication of one has been seen, either in the general or mark–recapture observations. Moreover, although no deductions can be made about young animals, comparison of the grown animals in the mark–recapture experiments has revealed no regular loss of weight or condition (Harrison & Emanuel, 1960) and no reduction in the monthly survival rate (Emanuel & Harrison, 1961) of those with infections as compared with the uninfected animals in the populations.

On the other hand, a combination of high index with high rate may, with one exception to be noted below, be accepted as good evidence of a grade II infection. More simply, when an abundant species of animal has a notably higher excretion rate of a particular serotype than any other species, then it can usually be accepted as a maintaining host of that serotype in that area. Thus, *R. s. conatus* was clearly a maintaining host of *L. australis* in most canefields sampled, *R. norvegicus* of *L. zanoni* in Innisfail town, *M. musculus* of *L. zanoni* in the cane farm in Table 7, and *P. nasuta* of a member or members of the *hebdomadis* serogroup (Table 4). A high index with a low rate is more difficult to assess, because it might be an expression, either of survival from severe infections, or of grade II under conditions of reduced frequency of transmission. The latter is more likely, particularly when the combination is observed in different samples or over a period, and we would therefore add *L. zanoni* in *R. rattus*, *L. ?pomona* in *R. assimilis*, and possibly *L. hyos* in *R. assimilis* to the associations that are accepted as maintaining ones.

Grade III infections would be characterized in the field by low excretion indices accompanying infection rates which would vary according to the amount of overflow from the maintaining hosts. There are numerous examples of this sort of relationship in Table 4. There is one condition under which the excretion index would rise. Some animals with this grade of infection do excrete leptospires for a period. When there are dense populations of maintaining and incidental hosts at a time when the environment is particularly favourable for transmission, the proportion of recent infections in the sample would rise, and the numbers excreting leptospires would be expected to rise with it. Some of the *L. zanoni* infections in *R. s. conatus* and of *L. australis* in *R. rattus*, as well as of both serotypes in the bandicoots, may belong to this category, which it should be possible to recognize from collateral information collected with the sample. The epidemiological significance of incidental hosts has been discussed by Audy (1958), and we have no specific information to add from our own experience.

Gordon Smith *et al.* (1961*a*) adopted a similar ratio to assess the infections they studied, although they seemed to consider that it depended on characteristics of the host species, rather than of particular host–parasite associations, and they added the caution that it could be applied only when infection leaves recognizable serological evidence that it has occurred. They also considered that their serotypes could be arranged in a graded series of antigenic capacity in rats (apparently independently of species), from *L. icterohaemorrhagiae* at the top to *L. hyos*, which

produced no antibodies at the 1:200 level, at the bottom. We have analysed our records to test both possibilities, with the following results:

(1) Except for the *R. s. conatus–L. australis* association, the bandicoots produced antibodies at consistently higher titres than the rodents against three serogroups (*pyrogenes*, *australis*, *pomona*) for which comparison was possible, and equally high titres against the *canicola* and *hebdomadis* serogroups which they maintained.

(2) Titres to *L. australis* in *R. s. conatus* were higher than in any other association in rodents, including *L. australis* in other species.

(3) Titres to *L. zanoni* were higher in the introduced rodents than in the native species.

(4) *L. australis* gave a higher incidence of antibodies than *L. zanoni* in excreting rats, and consistently higher titres than *L. zanoni* in bandicoots, maintaining rodent hosts, and incidental rodent hosts.

(5) Of three bandicoots and sixteen rodents infected with *L. hyos* (including those in the mark–recapture experiments), five rodents were excreting, and four of these had antibody titres less than 1:100; in the whole series, nine had titres of 1:100, three of 1:300, and four of 1:1000 or more. *L. hyos* thus appears to stimulate antibody production less readily than other local serotypes (though better than the Malayan strain), but the differences were not statistically significant.

These findings do not invalidate use of the excretion index for comparison of the North Queensland records. They show, too, that both suggestions of the Malayan workers are partly correct for our data, though the differences recorded are probably no more than expressions of the varied specific adaptations between hosts and parasites that Babudieri (1958) and we have described.

The index might also be affected by independent variations in the duration of excretion and persistence of antibodies. Both were followed for considerable periods in some of the mark–recapture studies (Harrison & Emanuel, 1960), and some variations were observed, but they were not of a magnitude that would materially reduce the statistical usefulness of the index.

Three aspects of the problem remain to be considered. The first is the excretion index that would mark a limit below which an independent association could not be maintained. It is likely to be quite variable, depending on the sum of the ecological factors enumerated earlier, and it is not to be forgotten, too, that quite a number of parasites seem to be able to maintain an extraordinarily tenuous existence under circumstances that are, at present, a complete mystery. We have, very tentatively, selected an index of 0·05 (equivalent to 5% of infected animals becoming chronic urinary carriers) as about the lowest at which an association is *likely* to be a maintaining one under favourable conditions, and 0·10 as a possible limit in less favourable conditions. These levels would fit the suspected maintenance of *L. mini* by *I. macrourus*, but more data are needed to define them with any confidence.

The second point is what happens when two or more grade II hosts for a given serotype occur together. There is no reason why it should not happen in nature, and it seems likely that some of the situations studied by the Malayan workers, as

well as our own experience with *L. celledoni* in rain-forest animals, may be examples of it. Presumably, the collective population of hosts serves as a unit, their combined status being the resultant of their relative numbers, positions in the grade, and habits in the field. It might be difficult to distinguish this situation from one in which there is an unusually high excretion index in incidental hosts, so it would be essential to test the findings in different localities and at different times before drawing conclusions from them.

The third point is that situations that are stable in one country may have little resemblance to equally stable situations in another. Racial differences between leptospires and between their mammalian hosts have already been mentioned, the dynamics of the associations undoubtedly differ, too, and there are many examples of a serotype being supported by one host in one country and a different one in another, *L. bataviae* by *R. rattus diardi* in Indonesia and *R. argentiventer* in Malaya and the *hebdomadis* serogroup by rodents in Malaya and marsupials in Australia being two striking examples. Some differences may be more local, and we still lack an explanation of the apparent absence of infection with *L. zanoni* in a sample of 129 *R. rattus* from the Douglas and Mulgrave shires.

To sum up, the concept of a mosaic of host–parasite relationships, each parasite having its own limited number of potential maintaining hosts and a larger, more widely shared range of incidental hosts, is well supported by our data. We believe it to be fundamental in understanding the ecology of the leptospiral infections, because it provides an element of relative stability on which the more variable factors may act. It incidentally provides support for the view that the leptospires are primarily parasites, because it is difficult to imagine how organisms that were primarily saprophytic could have evolved such patterns.

A mosaic of ecological factors is imposed on the host–parasite mosaic, resulting in a combined mosaic of foci of infection, large and small, and producing the varied associations that we see in the field. Focality, indeed, has been a striking finding of all the epidemiological inquiries that have been made in North Queensland, and the mark–recapture studies were designed to elucidate the variables that produce it. The results obtained from them have been summarized by Harrison & Emanuel (1962), and will be presented in greater detail in another paper. It may then become possible to examine the problem of controlling the incidence of infection in man.

SUMMARY

Leptospirosis is a zoonosis that causes appreciable ill health and economic loss in North Queensland. Fourteen serotypes of *Leptospira* were known to infect man in the area, and information on their local distribution and seasonal and occupational incidence had been obtained. The next step in the investigation was to determine the reservoir hosts and estimate their significance.

Wild animals examined comprised 5 monotremes, 643 marsupials, 2355 rodents, 67 bats, 30 birds, 28 reptiles, and 21 toads. Evidence of leptospiral infection was obtained from 223 marsupials, 309 rodents, and 6 fruit-bats. Analysis indicated that the principal maintaining hosts were:

Peremeles nasuta—of *L. kremastos* (and possibly *L. mini*) in canefields and rain forest.

Isoodon macrourus—of *L. broomi*, *L. mini*, and *L. kremastos* in canefields.

Rattus rattus, *R. norvegicus* (focal), and *Mus musculus* (focal)—of *L. zanoni* in town and canefields.

Rattus sordidus conatus—of *L. australis* in canefields.

Rattus assimilis—of foci of *L. australis*, a variant of *L. pomona*, and probably of *L. hyos* in rain forests.

Random infections were also encountered, but the maintaining hosts of the other serotypes known to infect man in the area (*icterohaemorrhagiae*, *canicola*, *bindjei*, *robinsoni*, *bratislava*, *grippotyphosa*, *medanensis*, *celledoni*) were not determined.

Domestic animals examined included 137 cattle, 54 pigs, 7 sheep, 34 dogs, 31 cats, and a dairy herd in which there was an epizootic of *pomona* leptospirosis. Again, there was a wide scatter of infection, but, apart from the long-known association of *L. pomona* and *L. hyos* with cattle and pigs, there was no evidence of maintaining hosts in this series.

As indicated in the Introduction, the work reported here was part of a planned investigation into the epidemiology of leptospirosis. Some of the early material was collected by Dr M. J. Mackerras, Dr R. L. Doherty, Mrs D. G. Delamoir, and Miss C. J. Ross, who had previously been stationed at the Institute's Field Station, and we are indebted to Mr D. W. Lavers, Mr S. G. Knott, and Mr R. E. Dunham, of the Queensland Department of Agriculture and Stock, for a number of sera from domestic animals. Many of the mammals were identified by Dr W. A. McDougall, of the Queensland Department of Agriculture and Stock, Mr E. le G. Troughton, then of the Australian Museum, Sydney, Mr B. J. Marlow, then of the C.S.I.R.O. Wildlife Section, Canberra, and by Dr J. L. Harrison, Dr M. J. Mackerras, and Mr R. Domrow of the Institute. We are indebted also to Mrs M. Macgregor, Librarian of the Institute, for considerable help with the literature.

REFERENCES

Addamiano, L., Babudieri, B. & Smith, D. J. W. (1960). Zwei neue zur Gruppe *Leptospira canicola* gehörige Serotypen. *Zbl. Bakt.* (1 *Orig.*), **180**, 419–21.

Alexander, A. D., Evans, L. B., Toussaint, A. J., Marchwicki, R. H. & McCrumb, F. R., Jr. (1957). Leptospirosis in Malaya. II. Antigenic analysis of 110 leptospiral strains and other serologic studies. *Amer. J. trop. Med. Hyg.* **6**, 871–89.

Alexander, A. D. & Smith, D. J. W. (1962). *Leptospira robinsoni*, a new serotype of the pyrogenes serogroup. *Aust. J. exp. Biol. med. Sci.*, **40**, 81–4.

Alston, J. M. & Broom, J. C. (1958). *Leptospirosis in Man and Animals*. Edinburgh: Livingstone.

Audy, J. R. (1958). The localization of disease with special reference to the zoonoses. *Trans. R. Soc. trop. Med. Hyg.* **52**, 308–34.

Babudieri, B. (1958). Animal reservoirs of leptospires. *Ann. N.Y. Acad. Sci.* **70**, 393–413.

Babudieri, B. & Gaspardis, D. (1959). Ricerche sulla presenza ed il significata di anticorpi per le leptospire nei sieri di bovini. *Zooprofilassi*, No. 1, 11 pp.

Battey, Y. M., Smith, D. J. W. & Barrow, G. (1964). The epidemiology of leptospirosis in North Queensland. II. Further observations on the hosts in the Mossman district. *J. Hyg., Camb.*, **62**, 485.

Broom, J. C. & Smith, D. J. W. (1956). *Leptospira celledoni.* A new leptospiral serotype. *Lancet*, ii, 886–7.

Combiesco, D., Sturdza, N., Radu, I., Sefer, M. & Nicolesco, I. (1959). Recherches concernant les sources d'infection dans les leptospiroses. *Arch. roum. Path. exp.*, **18**, 361–72.

Cotter, T. J. P. (1935). Progress reports on investigations into Weil's disease, North Queensland. *Health, Canberra*, **13**, 4–8.

Cotter, T. J. P. & Sawers, W. C. (1934). A laboratory and epidemiological investigation of an outbreak of Weil's disease in northern Queensland. *Med. J. Aust.* **2**, 597–605.

Derrick, E. H. (1956). Leptospirosis in North Queensland: an epidemiological comparison between the various leptospiral serotypes. *Med. J. Aust.* **1**, 281–7.

Derrick, E. H. (1957). Leptospirosis in North Queensland. *Papua N.G. med. J.* **2**, 23–4.

Derrick, E. H., Gordon, D., Ross, C. J., Doherty, R. L., Sinnamon, C. N., Macdonald, V. M. & Kennedy, J. M. (1954). Epidemiological observations on leptospirosis in North Queensland. *Aust. Ann. Med.* **3**, 85–97.

Doherty, R. L., Emanuel, M. L. & Moore, N. (1956). An outbreak of leptospirosis in North Queensland. *Med. J. Aust.* **1**, 276–81.

Emanuel, M. L. (1959). The susceptibility of mice to North Queensland strains of leptospirae. *Aust. J. exp. Biol. med. Sci.* **37**, 17–29.

Emanuel, M. L. & Harrison, J. L. (1961). Innisfail Field Station. *Rep. Qd Inst. med. Res.* 1960–61, pp. 7–9.

Forbes, B. R. V., Keast, J. C., Wannan, J. S. & Lawrence, J. J. (1955). The occurrence of antibodies for the *Leptospira grippotyphosa* serogroup in bovine sera in New South Wales. *Aust. vet. J.* **31**, 69–75.

Gordon Smith, C. E., Turner, L. H., Harrison, J. L. & Broom, J. C. (1961*a*). Animal leptospirosis in Malaya. I. Methods, zoogeographical background, and broad analysis of results. *Bull. World Hlth Org.* **24**, 5–21.

Gordon Smith, C. E., Turner, L. H., Harrison, J. L. & Broom, J. C. (1961*b*). Animal leptospirosis in Malaya. II. Localities sampled. *Bull. World Hlth Org.* **24**, 23–34.

Gray, D. F. (1940). Canine leptospiral jaundice in Queensland. *Aust. vet. J.* **16**, 200–3.

Gray, D. F. (1942). Canine leptospiral jaundice in Queensland. II. Aetiology, serology and epidemiology. *Aust. vet. J.* **18**, 2–13.

Harrison, J. L. (1962). Mammals of Innisfail. I. Species and distribution. *Aust. J. Zool.* **10**, 45–83.

Harrison, J. L. & Emanuel, M. L. (1960). Innisfail Field Station. *Rep. Qd Inst. med. Res.* 1959–60, pp. 16–21.

Harrison, J. L. & Emanuel, M. L. (1962). Host-parasite association of small mammals and leptospires in tropical Queensland. *Proceedings of the First Regional Symposium on Scientific Knowledge of Tropical Parasites, Singapore*, 5–9 November 1962, pp. 154–66.

Johnson, D. W. (1950). The Australian leptospiroses. *Med. J. Aust.* **2**, 724–31.

Lumley, G. (1937). Leptospirosis in Queensland: A serological investigation leading to the discovery of distinct serological groups of leptospirae causing leptospirosis as it occurs in northern Queensland, with some other related observations. *Med. J. Aust.* **1**, 654–64.

Morrissey, G. C. (1934). The occurrence of leptospirosis (Weil's disease) in Australia. *Med. J. Aust.* **2**, 496–7.

Neghme, A., Christen, R., Jarpa, A. & Agosin, M. (1951). Estudios sobre immuno-biología de las enfermedades parasitarias. II. Susceptibilidad de cepas puras de ratones a la leptospirosis experimental. *Bol. parasit. chil.* **6**, 4–5. (Abstracted in *Bull. Hyg.* **26**, 897.)

Packchanian, A. (1940). Susceptibility and resistance of certain species of American deer mice, genus *Peromyscus*, and other rodents to *Leptospira icterohaemorrhagiae*. *Pub. Hlth Rep., Wash.*, **55**, 1389–1402.

Sawers, W. C. (1938). Some aspects of the leptospirosis problem in Australia. *Med. J. Aust.* **1**, 1089–97.

Schebitz, H. & Dedié, K. (1955). Zur Bewertung von Leptospirentitern im Pferdserum. *Zbl. vet. Med.* **2**, 522–43.

Sinnamon, C. N., Pask, V. M., Smith, D. J. W., Brown, H. E. & Tonge, J. I. (1953). Canicola fever in Australia. *Med. J. Aust.* **1**, 887–90.

Smith, D. J. W. & Brown, H. E. (1955). Two additional serotypes of leptospirae from North Queensland. *Aust. Ann. Med.* **4**, 287–90.

SMITH, D. J. W., BROWN, H. E., TONGE, J. I., SINNAMON, C. N., MACDONALD, V. M., ROSS, C. J. & DOHERTY, R. L. (1954). The serological classification of 89 strains of leptospirae from North Queensland, including five serotypes new to Australia. *Aust. Ann. Med.* **3**, 98–105.

SMITH, D. J. W. & DOHERTY, R. L. (1956). Double infection with two serotypes of leptospirae: A case report. *Med. J. Aust.* **2**, 643–5.

STAVITSKY, A. B. & GREEN, R. G. (1945). Susceptibility of the young white mouse (*Mus musculus*) to experimental leptospirosis. *Science*, **102**, 352–3.

SUTHERLAND, A. K., SIMMONS, G. C. & KENNY, G. C. (1952). Leptospirosis in cattle. *Qd agric. J.* **75**, 225–32.

TONGE, J. I. & SMITH, D. J. W. (1961). Leptospirosis acquired from soil. *Med. J. Aust.* **2**, 711–12.

WALCH-SORGDRAGER, B. (1939). Leptospirosis. *Bull. Hlth Org. L.o.N.* **8**, 143–386.

WANNAN, J. S. (1955). Leptospiral serotypes in Australasia. *Aust. vet. J.* **31**, 297–301.

WOLFF, J. W. & BOHLANDER, H. J. (1961). The serological classification of *Leptospira esposito*. *Trop. geogr. Med.* **13**, 175–9.