

CHAPTER 8

GENERAL DISCUSSION - TOWARDS A STRATEGIC APPROACH TO WILDLIFE DISEASE CONTROL

INTRODUCTION

Individual parts of this overall study have been discussed at the end of each chapter, and this final chapter will deal primarily with putting the work in context in relation to epidemiological understanding of wildlife diseases, and on evaluating the effectiveness of the research strategy which has been pursued in order to understand tuberculosis in possums, and to identify improved control methods.

WILDLIFE RESERVOIRS OF DISEASE

The importance of wild animals as reservoirs for diseases in domestic livestock was first recognized in the latter half of the 19th century. This relationship was particularly evident in Africa where outbreaks of rinderpest, African swine fever and foot and mouth disease in the domestic animal population were blamed on transmission from wild animals (Fowler 1985). Wildlife reservoirs of disease can complicate the control of diseases in domestic animals to such an extent that it may seem impossible to achieve the goal of eradication or control of the disease. Methods which have proven their effectiveness in the control of diseases in domestic animals such as test-and-slaughter policies cannot be applied in most situations to wildlife populations. Effective control requires an understanding of the epidemiology of a disease, including its infection dynamics within domestic as well as wildlife populations. Studies of the epidemiology of diseases in wildlife species are quite challenging because individual animals are more difficult to follow up, wildlife populations are more subject to variation in environmental factors (vegetation, topography and climate) than domestic populations and most often the veterinary and ecological knowledge about the species involved is quite limited. Schnurrenberger *et al* (1987) stress that special thought must be given to the interrelationship of wild and domestic populations if they coexist in an area subjected to a disease control or eradication effort. One of the major questions which will have to be answered in such cases is whether or not any of the susceptible wildlife species is likely to be a maintenance or reservoir host of infection - in which case control efforts must take this fully into account- or whether the wildlife are merely spillover or dead-end hosts - in which case control efforts can focus almost entirely on the domestic animal population..

Schwabe *et al* (1977) define a reservoir host to be any species in which an infectious agent multiplies or develops and upon which it depends as a species for survival in nature. Other major veterinary epidemiology texts by Thrusfield (1986) and by Martin *et al* (1987) use similar definitions. Fowler (1985) lists a number of characteristics which are found in a reservoir host. First, the reservoir host must be able to maintain the disease in the absence of infection in other species. Second, the host must be able to shed the organism to allow infection of other species. Third, transfer of the organism from the wild host to a domestic animal must be demonstrated. This is usually quite difficult to demonstrate under field conditions. Fourth, reservoir hosts need not show overt signs of disease. Fifth, the reservoir host is usually not seriously affected by the organism. A reservoir host has to be distinguished from a spillover host (infected incidentally but infection is not self-sustaining in the species

without regular transfer from other species) and also from a dead-end host (infected incidentally, but cannot transfer infection to any other animals). If a species is a true spillover host for a particular organism, it would not be necessary to control the disease in the spillover host, because it would disappear or become epidemiologically insignificant once the disease has been controlled in the species which is a reservoir host. A spillover host provides an indicator for the presence of a particular disease in an area, but should not be the main target of a disease control effort.

Rinderpest, rabies and African swine fever are three animal diseases which represent a spectrum with regard to the degree of involvement of reservoirs of infection in wild animal species in producing disease in domestic animals. It was therefore considered appropriate to compare these three diseases with tuberculosis, because of the insights which this might offer.

Rinderpest

Rinderpest is an ancient plague which regularly devastated the cattle and buffalo populations of Asia and Europe and occasionally wrought havoc in North Africa. Currently the disease is endemic in most African countries north of the equator, in the Middle East, Pakistan and in India. It causes substantial losses in these countries unless vaccination is practised. Rinderpest is caused by a Morbillivirus and probably affects all cloven-hoofed animals (Scott 1990).

In Africa there has been a lot of discussion about the importance of wild animal reservoir species in the epidemiology of rinderpest. It was for example suggested that large concentrations of wild animals, as in the Serengeti region of East Africa, could act as "long-term reservoirs" of the rinderpest virus, in the absence of the disease among cattle. This was based on finding specific antibodies against the virus in these wild species. Clearly this cannot be considered sufficient evidence for a species to be considered a reservoir host (Pastoret *et al* 1988). As a consequence of the possibility that wild animals could be a reservoir for rinderpest virus some authorities of national parks and game reserves minimise or delay reports of mortality in rinderpest-susceptible species in order to avoid subsequent investigations (Plowright 1988). Plowright (1982) used the disappearance of rinderpest from the Serengeti region in Tanzania to suggest that game animals are not acting as reservoir hosts for the virus. He concluded that in the Serengeti region, at the periphery of which cattle-game contacts are frequent, a feed-back mechanism of cattle-to-game and game-to-cattle transmission had been necessary to maintain the disease continuously during the decades up to 1962-63, after which it disappeared. By that time vaccination campaigns had been under way in East Africa during the previous ten years. Plowright writes that it is likely that after 1963 local outbreaks have still occurred, but tended to die out rapidly in the presence of high densities of susceptible animals in relatively small areas. From the evidence in the literature it appears that in East Africa game animals are acting as spillover hosts for the rinderpest virus and the reservoir host is likely to be the domestic cattle population.

Rabies

Rabies is a viral infection of mammals which is transmitted in the saliva of rabid animals and usually manifested by a fatal encephalomyelitis. It is caused by a Lyssavirus and it occurs world-wide, except in Antarctica, Australasia and a number of islands and small countries. In addition to maintenance through dog infection (which was the primary problem but has been controlled in many countries by vaccination), rabies has been found to circulate in wild species - mainly in foxes, skunks, raccoons, mongooses and bats. Domestic animals as well as humans are at risk of contracting the disease due to exposure to rabid wild animals.

In large parts of Asia, Africa and Latin America the rabies virus still circulates principally within the dog population - accounting for 95% or more of all diagnosed rabies cases. In contrast, in Western Europe and North America dog populations do no longer constitute a major reservoir of rabies infection. This has been achieved through stray dog control and widespread vaccination. Yet, the disease still has not been eradicated, due to the presence of smaller but nonetheless significant reservoirs of infection in wildlife species. It is widely accepted that notably foxes, mongooses, skunks and raccoons represent major maintenance hosts for the rabies virus. This problem has proven to be a much greater challenge than control of the disease in domestic animal populations. In the past, efforts have been made to control or eradicate rabies in wildlife reservoirs using techniques such as population reduction and more recently vaccination against infection with the rabies virus. It is now widely accepted that a thorough understanding of the epidemiology of rabies infection is required in order to make progress towards the ultimate goal of rabies eradication.

MacDonald (1980) describes the epidemiology of rabies in Europe as relatively simple because the fox is the single most important maintenance host in wildlife. The general view is that in Western Europe rabies can be eradicated, once the disease is controlled in foxes. Kaplan *et al* (1986) warn that this may not be the case as there are other biting animals such as the raccoon dog which could replace the fox as a maintenance host of infection.

Fox Rabies

The epidemiology of fox rabies has been extensively studied. It has been concluded that the social behaviour of foxes and the social structure of fox populations is of crucial importance for a bite-transmitted disease such as rabies. Social behaviour follows an annual cycle determined by reproduction and mortality. Adult foxes seasonally share a home range as male-female pairs with their young of the year. They occupy home ranges which in some areas are exclusive and in others overlap. Juvenile foxes may leave the family territory at the age of about six months and disperse over quite a large area. Young female foxes may stay in or near the home territory, whereas most males emigrate. Rabies incidence and spread in Europe reaches its minimum during May or June, immediately after the whelping season, when the population has reached its maximum. Then incidence increases steadily over the second half of the year until it reaches a maximum in February or March of the following year, and diminishes thereafter. Considering the relative brevity of the rabies incubation period (around 1 month), it

is relatively easy to identify social behaviour which may be associated with this seasonal incidence pattern. During the mating season wandering increases and contacts multiply, thus favouring transmission of the virus during fights among males and later, biting of females. The increase in incidence during the second half of the year is attributed to the dispersion of young foxes. During this time of the year the fox population is artificially elevated by newly independent young who are trying to find a territory. The majority of rabies cases during the second half of the year involves young male foxes which are more precocious and disperse over greater distances than young females (Toma and Andral 1977). During the clinical stages of the disease rabid foxes can show three different types of behaviour. If they develop the less common furious form (11% of artificially infected foxes in captivity), they wander around and attack any object encountered. They might show increasing paralysis and become very docile. MacDonald and Voigt (1985) concluded that given the available information it appears that rabid foxes in general behave like healthy ones (i.e. have the same movement patterns and frequency of social encounters) yet become more aggressive.

In Europe, cattle almost exclusively contract rabies from foxes. Cattle are exposed to infection between April and November when they are kept on pasture. The incidence of bovine rabies parallels the pattern of fox rabies incidence with a lag of 2 to 3 months. Given that they are only exposed until November, incidence diminishes as expected after December (Toma and Andral 1977).

Fox rabies in Europe shows a cyclic temporal pattern. During an epidemic, rabies reduces the fox population by about 50%, with an additional proportion destroyed by population control conducted by humans in response to the increased risk of human exposure. In the year following the epidemic, rabies incidence is very low. But the sharp reduction in fox population density improves survival and increases reproduction. In the absence of any population control over the next 2 to 3 years the population can increase to the levels it had prior to the occurrence of the epidemic and the conditions are favourable for an increase in rabies incidence (Toma and Andral 1977).

In Europe the fox rabies front advanced at the relatively slow rate of about 30 to 60km/year suggesting that spread of infection is generally of nearest neighbour type (Ball 1985). Occasionally the rabies front appears to “jump” ahead due to infected juvenile animals which dispersed in autumn into the rabies-free area and set up a local epidemic. The shape of the front is influenced by geographical obstacles such as mountain ranges and streams. Rivers running parallel to the rabies front can represent temporary barrier, but can in fact accelerate spread when running parallel to the general axis of the progression (Toma and Andral 1977).

Field experience and simulation modelling exercises suggest, that if in Western Europe a fox population has been reduced below a certain level (by rabies itself and/or fox control), rabies disappears in foxes and in all other terrestrial mammal species (except in bats; Wandeler *et al* 1993). Despite this situation only few fox population control campaigns have been

successful. This can be partly explained by the fox's resilience to persecution and its high reproductive potential in connection with high carrying capacities of rural and suburban habitats (Wandeler 1988). Denmark for example succeeded in creating an artificial barrier of low fox density in South Jutland, which protected the rest of the peninsula. But over the years the country has experienced a number of invasions by rabid foxes which were very expensive and difficult to control. MacDonald (1980) questions the successfulness of the Danish rabies control programme because at the same time the disease also disappeared in the German state of Schleswig-Holstein bordering on Denmark where no control had been conducted. Macdonald and Voigt (1985) conclude that in Europe culling of foxes had little effect on the speed at which the rabies epidemic advanced. To prove the success of a control campaign such as that for fox rabies is complicated by the cyclical occurrence of rabies. The disease can be completely absent for periods of up to 5 years. The use of vaccination shows signs of producing more promising results than population reduction. Belgium has for example reported that since a large-scale vaccination campaign was conducted in a 2200km² region in 1989-1990 no livestock rabies has been reported. MacDonald (1980) writes that there are only two outbreaks of rabies in Europe which can claim to have been completely eradicated by fox control, one in Dijon in 1923 and the other in Corsica in 1943.

Rabies in Other Species

In North America there are four wildlife reservoir species for rabies - namely foxes, raccoons, skunks and bats. In contrast to Western Europe population reduction has not been encouraging at all as a method for controlling rabies in wild animals (MacInnes 1988). Smith and Baer (1988) report that in most geographical areas of Canada and the U.S.A. a number of different wildlife species may be involved in the epidemiology of rabies, but most often there is a predominance of cases in a single host species. For example in 1986 75% of 3565 skunk rabies cases occurred in a large area extending from southern Alberta, Saskatchewan and Manitoba in Canada across to the central United States to the Rio Grande River. Ninety-nine percent of 1609 rabid raccoons reported occurred in the southeastern and mid-Atlantic United States. Of 1915 fox rabies cases reported in 1986, 90% occurred in southern Ontario, Quebec and northern New York. Within any of these areas the cases which occur in other animals are regarded as "spill-over" infections from the major reservoir animal(s). Monoclonal antibodies have been used to identify antigenically different variants of rabies virus. This information was then utilized to analyze the geographical occurrence of rabies in different animal species. In Texas where skunk rabies predominates a cluster of rabies in gray foxes is observed every year. Virus isolates from these foxes were easily distinguishable from the skunk isolates. It was concluded that there may be an independent cycle of infection within the fox population. Under such circumstances it would not be possible to eradicate rabies by eliminating skunk rabies. Monoclonal antibody analysis of isolates was also used to investigate the role of bats in the epidemiology of rabies in North America. It was found that none of the reaction patterns that characterize bat rabies was represented in the patterns of viruses isolated from the major

terrestrial rabies enzootics. Parker (1975) conducted a detailed study of rabies in skunks. He identified some aspects of skunk behaviour which contributed significantly to the spread of rabies virus infection within a skunk population. One factor is that skunks are using communal dens with a single dominant male in the den and the other is that young unattached males tend to wander around during the late winter months. Parker suggests that this contributes to the peak of cases in the second quarter of the year, since the breeding season is late February and March, and a subsequent incubation period of 30-60 days would coincide with the peaks observed. He also mentioned the possibility of perinatal transmission.

Carey (1985) discusses multispecies rabies in the eastern United States. He concludes that there are three manifestations of multispecies rabies in this region. First, rabies virus is maintained concurrently, but independently, in bats and in one common terrestrial carnivore such as the striped skunk, gray fox, red fox or raccoon. Second, during epidemics in terrestrial carnivores, up to 20 other species may be infected which can be considered "spill-over" hosts. Third, the major terrestrial carnivore species involved may change over time. Rabies is known to occur in epidemic cycles. It is not understood whether it reaches an endemic state at low prevalence between epidemics or if it disappears from the maintenance host population and after a period of time is reintroduced from an inapparent reservoir such as mustelids, bats or felids. Carey also suggested that fox rabies persists in areas of rugged topography with a mixture of farms and forest. Such an environment results in patchiness of fox population density and may reduce depletion of the number of susceptible foxes by producing more rapid spread of the epidemic, leaving islands of unaffected foxes.

Voigt and Earle (1983) conducted a study on the interaction between coyotes and red foxes in the Canadian province of Ontario. They found that foxes avoid raising pups in areas where coyotes traditionally travel and raise pups. Voigt and Earle suggest that foxes may have vacated certain areas due to the presence of coyotes. And this may explain why in some areas of Ontario no rabies has been reported during a 10-20 year period, whereas southern Ontario has the highest incidence of wildlife rabies in North America.

Bat rabies occurs in a number of countries of the world. Bats live at extremely high densities which provide favourable conditions for transmission through aerosol or biting. Epidemiologists have concluded that rabies in insectivorous and fructivorous bats exists as an endemic largely independent from the cycle in terrestrial animals, and that rabies in bats cannot be considered an important reservoir for infection of terrestrial wildlife populations (Smith and Baer 1988). Brass (1993) quotes a number of studies in support of this theory. In most of these studies it was found that in regions where rabies was endemic in local bat populations, no rabies infection had been detected in terrestrial species. In contrast Tinline (1988) writes that it may well be possible that bats may cause epidemics in other species. They migrate over long distances and could set up new reservoirs of infection or introduce new strains into an area. These views may not be totally inconsistent, since bat rabies may be a separate cycle under

normal circumstances, with spillover on rare occasions - which are of note when they precipitate an epidemic in a particular region.

In Latin America the epidemiological situation is different again, and dog rabies still accounts for over 50% of reported animal rabies. Vampire bats are believed to be a significant reservoir for rabies infection in some of these countries, with certain areas being particularly at risk. The importance of this source can only be indirectly estimated through the considerable incidence of bovine rabies, which is considered to be mainly caused by transmission from vampire bats.

Mongoose are a principal maintenance host of rabies infection in the Caribbean. On the island of Grenada, rabies has been continuously monitored since the late sixties. During this time a number of control campaigns of the mongoose population have been conducted, which led to temporary reductions in mongoose-transmitted rabies. A poisoning campaign in 1973 destroyed about one to two thirds of the mongoose population. Unfortunately in most areas the population recovered within 6 to 9 months. Everard and Everard (1985) conducted surveys for the presence of serum-neutralizing antibodies in the mongoose population of Grenada which led them to the conclusion that these animals actually developed immunity to rabies which in turn caused the temporary localized disappearance of rabies. They describe the occurrence of a cycle of high antibody/ low rabies and low antibody / high rabies cycle, because after the dispersal or death of the immune individuals in the population, more susceptibles are available to promote resurgence of the disease. As the immune population builds up, the cycle is repeated. At the end of a cycle up to 60% of individuals may show antibodies. Everard and Everard conclude that poisoning campaigns for the control of mongoose populations may defeat the purpose as it also removes immune animals. They recommend the use of vaccination soon after the peak of an epidemic has been reached, when the proportion of immune mongooses is at its maximum and it should be possible to eradicate rabies completely.

In Africa knowledge about the involvement of wildlife species in the epidemiology of rabies infection is limited, with the exception of South Africa. Blancou (1988) suggests that the dog may be the major reservoir of rabies infection in most of Africa as the disease disappears in countries where canine rabies has been controlled. Given the experience from other continents Blancou suggests that this may be a result of inadequate reporting in some of the African countries. The importance of the interaction between a particular type of habitat and a wild animal species can be illustrated using the example of the yellow mongoose. In the grassveld, an area from 1200 to about 1800 above sea level, rabies is considered to be endemic. In this region the yellow mongoose is the most important host of rabies and spread of infection to any other species is not common. This area is well defined in terms of vegetation and altitude. Further north, in the sub-tropical and tropical climate zones, the vegetation changes to predominantly thorn bushes and trees, the bushveld. In this region rabies occurs typically in epidemics, and the species involved are mainly feral dogs and black-backed jackals. The limits

of the area where rabies epidemics occur is less clearly defined than the grassveld where endemic disease is found. There is also a larger number of vector species involved and spread from canid hosts to for example farm animals is much more common in the bushveld than in the grassveld (Kaplan *et al* 1986). Blancou (1988) writes that between 1977 and 1988 an independent cycle of rabies had established in the kudu antelope in Namibia.

The above examples show that there are major differences in the epidemiology of rabies between geographical regions. It is interesting to note that in some areas of the U.S.A. it appears that foxes are “spill-over” hosts of infection rather than maintenance hosts, which they generally are in regions where rabies is present. It is likely that fox densities are too low in order to maintain endemic disease levels. The possibility of having several independent cycles of rabies infection in different wildlife in a single area complicates any control attempts to a great extent.

African Swine Fever

African swine fever provides one of the classic examples of a disease where wildlife was the primary reservoir but initial spillover of infection into domestic animals later produced a new epidemiological situation with different maintenance hosts. This acute contagious viral disease emerged as a disease of domestic pigs in East Africa at the beginning of this century, where it probably had previously been endemic only in wild species of the Suidae family. In 1957 the disease occurred in Portugal through importation of infected pork and it spread despite control efforts into Spain, Italy and France (Losos 1985). It is now considered to be endemic in Sardinia.

African swine fever occurs in many countries of the African continent where warthogs, bushpigs and forest hogs act as reservoir hosts of the virus. Typically the disease is only locally endemic in the wild pig population. Wild pigs do not develop clinical disease, but occasionally are the source of infection with virulent strains for domestic pigs which suffer high morbidity and mortality. The disease is transmitted between warthogs primarily by a argasid tick which live in the same burrows as the pigs. The virus can be maintained in the argasid tick for long periods in the absence of fresh infection from pigs, so that they can act as a temporary reservoir. Young warthogs, which have virus circulating in the blood, are the main source of infection for ticks. In older pigs the virus is mainly found in lymphatic tissue. In Africa the endemic infection is maintained through infection cycling between ticks and the warthog population.

Transmission from warthogs to domestic pigs was a relatively rare event, and was thought to result mainly from transmission through the tick vector. Double fencing to exclude wild Suidae and their ticks from access to domestic pigs was a very successful control method, and the disease did not occur commonly in commercial piggeries. Chronic cases of the disease and asymptomatic carriers of infection were unusual in domestic pigs.

Until 1957 it was thought that African swine fever was one of the specifically African diseases. In that year it appeared in Portugal and in 1960 in Spain where it caused the death of thousands of pigs. In both situations contaminated pork was found to be responsible for the initial outbreaks. Local argasid tick species became involved in the cycle (Liess 1988; Blood and Radostits 1989; Sanchez-Vizcaino 1992). Later it spread into Italy.

Over the next few years a new situation developed in Africa as well as in affected European countries with regard to the pathology of African swine fever. A less acute form of the disease emerged, with more frequent chronic cases and asymptomatic carriers. Within an infected herd, virus spreads rapidly via direct and indirect transmission. Asymptomatic carrier pigs and contaminated meat both became important sources of infection, and the ticks were no longer essential for maintenance of the disease. Outbreaks have occurred in a number of countries, notably in the Americas, due to consumption by pigs of pig meat scraps in garbage, mainly from airline food. Wild European pigs are susceptible to infection and are the maintenance host for African swine fever on Sardinia. In contrast, wild pigs became infected in some other European countries, but infection either disappeared spontaneously or it proved possible to control it.

BOVINE TUBERCULOSIS

Mycobacterium bovis has one of the broadest host ranges of all known pathogens (Grange and Collins 1987), with a complex epidemiological pattern which involves interaction of infection among human beings, domestic animals and wild animals. However the epidemiological patterns and the importance of various species in transmission of the disease appears to have varied over time, and among different countries. For example, bovine tuberculosis in man has declined from an important syndrome to unimportance over the last century and does not appear to have had a resurgence in parallel to that of *Mycobacterium tuberculosis* in man, bovine tuberculosis is important in badgers in the United Kingdom and the Republic of Ireland but not in other parts of Europe, and the disease is widespread in the possum in New Zealand but not in its native Australia. While the explanation for these recorded differences is in some cases well-known but in others is unclear, the differences result broadly from issues of host susceptibility, from the existence and scale of different transmission pathways, and from behavioural factors in various species which determine whether a specific potential transmission pathway can be expressed.

The susceptibility of a particular species to infection with *Mycobacterium bovis* is a major factor determining its importance in the epidemiology of bovine tuberculosis. Schliesser (1985) produced a susceptibility ranking for a number of domestic and laboratory animals. He suggests that chimpanzees, rhesus monkeys, rabbits and guinea pigs are highly susceptible to infection with *Mycobacterium bovis*. Cattle, sheep, goats, pigs, cats are susceptible, horses, dogs and mice are not very susceptible and rats and poultry are not susceptible to infection. Schliesser based this information on data from the literature as well as his own experience. He

mentions that a comparison of susceptibility between species is quite difficult as it depends on the doses which were used in the individual experiments. In terms of a genetic influence on susceptibility of a particular species, Schliesser came to the conclusion that this may have an effect on the pathogenesis of tuberculosis in the species, but is unlikely to influence initial susceptibility to infection.

There is only limited information available regarding the susceptibility of wild animals to infection with *Mycobacterium bovis*. This is further complicated by the fact that many wild animal species are likely to show a different immune response in captivity compared with the response they would show in the wild. This is particularly evident in possums where Corner and Presidente (1980, 1981) found that these animals in captivity succumbed very quickly to experimental infection with *Mycobacterium bovis*. Buddle *et al* (1992) used lymphocyte stimulation using T-cell mitogen as an indicator of the effect of stress on cellular immunity and estimated that a minimum of 4 weeks of cage adaptation after capture was required before possums could be used for experimental studies, to avoid producing atypical fulminating disease in stressed possums.

Reliable estimates on the prevalence of bovine tuberculosis in wild animal species is scarce due to the fact that these species are not normally included in national bovine tuberculosis surveillance programs. In general it has been considered that the occurrence of *Mycobacterium bovis* in wild animals was dependent on the presence of infection in domestic animal species. Schliesser (1985) considered that in Europe as long as the disease remained in domestic animals, sporadic cases were reported in wild animal species such as deer, elk, chamois, pigs, foxes, hares, beaver and hedgehogs. He mentions that in zoo animals bovine tuberculosis occurs to a significant extent even in countries where the disease has been eradicated from domestic animals. He adds that the available data on infection in zoo animals is likely to be an underestimate of the true bovine tuberculosis prevalence as the majority of communal and private zoos, wildlife parks and pet shops would not report occurrences of the disease.

It is of note that the susceptibility of the three major wildlife reservoirs which have been of concern in tuberculosis eradication programs in recent years - possums, badgers and deer - was established, as for numerous other species, some years before evidence began to accumulate that they could act as maintenance hosts. It is uncertain for badgers and deer whether the recognition of an emerging problem signified increased occurrence or improved detection, but in the case of the possum the evidence seems unequivocal that a new reservoir host had been established.

The second major factor influencing whether a wildlife host is of practical importance in tuberculosis control is the effectiveness of transmission pathways within the species, and between the potential maintenance host and domestic stock. In this regard, there are a number of similarities between badgers and possums, which are the two species studied in most detail.

Both species can develop lymph node abscesses which discharge through the skin and hence provide a mechanism of transmission between animals of the same species, and a source for contamination of the environment. They both develop tuberculous lesions predominantly in the lung which is likely to result in excretion of bacteria via the respiratory route. There is evidence in the case of both the badger and the possum that 'pseudo-vertical' transmission from an infected mother to her young may be of considerable importance for maintenance of disease within an area, as well as for spread of infection between populations. After young animals become independent of their mothers some of them will disperse in order to establish a new home range. A proportion of these animals may be infected with *Mycobacterium bovis* and would therefore be able to set up new disease foci.

The issue of how possums and badgers transmit infection to domestic stock is still currently being resolved, since it has so far proved impossible to answer this question directly, and indirect evidence has been used to infer how transmission takes place. Early evidence on badgers gave emphasis to aerosol transmission, but later reports have emphasized the fact that excretion in urine is common, and hence have suggested pasture contamination as the main route. In possums the reverse trend has occurred, with early papers focussing on transmission through contaminated pasture, and later work favouring aerosol transmission as the central method. With regard to badgers, the published evidence shows that urinary excretion occurs commonly, but it does not show unequivocally that this is how transmission commonly occurs, rather than by aerosol. It may be that in both species aerosol transmission is the pathway of greatest practical importance (Morris *et al* 1994).

Therefore the transmission mechanisms exist in possums and badgers for them to maintain infection in an endemic state within the species, and to transmit infection to the important domestic animal host species.

There is also now good evidence (Paterson 1993) that the third component of a successful maintenance host occurs in the possum, in that behaviour of both possums and domestic stock facilitate transmission of infection. Terminally tuberculous possums show changed behaviour in that they wander around during the day, stay closer to feeding areas such as pasture and do not avoid cattle as actively as healthy animals. They are likely to attract the attention of and be investigated by domestic animals, which will provide excellent opportunities for transmission of infection. There is circumstantial evidence that a comparable behavioural pattern facilitating exposure may occur in the badger as well as in the possum, although this has not been demonstrated under controlled conditions.

The aspect in which there is an important difference between badgers and possums is related to the social behaviour patterns of the two species, and why they can both act as reservoir hosts when they have such different social structures. Badgers are social animals and live in social groups which occupy exclusive territories. Each territory contains a number of underground burrows termed setts, the main one in a territory being very extensive. Within a

social group they will share setts, and sleep in close contact with each other. A badger sett provides good conditions both for survival of organisms and for aerosol transmission, which would greatly facilitate dissemination of tuberculosis within a social group.

In contrast possums are not considered to be true social animals, nor do they occupy exclusive territories. Close contact sufficient for direct animal-to-animal transmission would be likely to occur under much more limited circumstances - mainly between mother and young during the rearing period, between adult males and females during courting and mating, between adults (males in particular) during competitive and agonistic behaviour, and during simultaneous den sharing by two or more animals (excluding mother-joeey sharing, which is stated above).

However possums occur at much higher population density per square kilometer than badgers, and it would seem reasonable that at possum densities transmission mechanisms which have a relatively low probability of success per encounter are effective because of the high number of other animals to which each possum is potentially exposed, whereas in badgers the number of contacts is far smaller but because of the degree of social interaction transmission is as successful as in the possum.

Thus it would appear in retrospect that the necessary factors were present to allow both possums and badgers to act as successful wildlife reservoirs for tuberculosis, and might perhaps on some future occasion allow other species to convert from occasional hosts to reservoir hosts if circumstances are right. In this connection it would be very informative to understand exactly how the possum became endemically infected with tuberculosis in New Zealand but did not in Australia. At this stage it is possible to put forward reasoned conjectures, but there is insufficient solid evidence to formulate a firm hypothesis.

EPIDEMIOLOGICAL COMPARISONS OF TUBERCULOSIS AND OTHER WILDLIFE DISEASES

The three diseases chosen for comparison with tuberculosis were deliberately selected to cover a spectrum of the epidemiological patterns and trends found in diseases which involve wildlife, not because they are all epidemiologically similar to tuberculosis. All three are viral rather than bacterial diseases and the disease process is generally more acute than for tuberculosis, but nevertheless useful comparisons can be made. Since some essential pieces of epidemiological information required for the design of tuberculosis control programs which would be effective in the presence of a wildlife reservoir are not yet available, there is merit in looking for parallels with other diseases which might offer clues to the best chances of making progress.

Of the three diseases, the one which shows closest parallels to tuberculosis is rabies, because despite the fundamental differences in the nature of the diseases there are ecological factors which produce intriguing similarities in the epidemiology of the two diseases. Firstly, in the form of interest here, tuberculosis is maintained principally in a single reservoir species (possum or badger), with infection in the domestic host species being largely of the spillover

type. There tend also to be low levels of what appears to be spillover of infection into predator and scavenger species in particular, though producing sometimes higher prevalence in these species than in the reservoir species, due to the concentration effects of such animals consuming large numbers of the reservoir species. Rabies shows the interesting feature of typically having one primary maintenance host in most areas, but with lower incidence in one or more other hosts - which might become important as a residual source of infection if a control program is limited to the principal maintenance host. In some cases there is a second apparently independent cycle of infection being maintained in another species. The issue of whether any wildlife hosts other than the possum can genuinely act as maintenance hosts either in small foci or nationally remains controversial but a very open question. It is unlikely to be decided for some years, but requires continuing evaluation. Some new evidence is expected to arise from later stages of the longitudinal study described in this thesis. To date the only wildlife species in New Zealand that has been shown to be a true maintenance host is the possum, and the same would appear to be true with regard to the badger in the United Kingdom and Ireland. In New Zealand feral deer may also qualify as maintenance hosts in that they appear to maintain infection within the species without the need for cross-infection from other species, and may perhaps infect possums in previously clear areas on occasions. Although arguments have been presented that other species are also acting as maintenance hosts, there is little firm evidence that this is true, and it seems likely that if other species do fill this role it is at most in isolated areas with special epidemiological features. So as for rabies, in the case of bovine tuberculosis in the United Kingdom, Ireland and New Zealand there is typically only a single important maintenance host, although many more species can be shown to be infected.

Temporal trends also show intriguing similarities between tuberculosis and fox rabies. In both cases the annual maximum falls in spring, but there is no apparent common ecological reason for this and it is probably a chance similarity. Both badger tuberculosis and rabies show multi-year cycles in incidence, which probably does result from common factors related to the generation length of the host species and the number of susceptibles in the population at particular times. It is not yet clear whether there is any long-term cyclicality in possum tuberculosis, but the short generation length of possums makes major multi-year cyclic patterns less likely to be important. There has been no published evidence of specific weather influences on rabies of the kind provisionally identified in the longitudinal study for possum tuberculosis. This suggests that rabies transmission and pathogenesis are little influenced by short term weather patterns because there is not a stress component to the determinants of the rate of pathogenesis, whereas in possum tuberculosis the evidence from the longitudinal study suggests that pathogenesis is variable in length and shortened by weather or nutritional stress.

Fox rabies and tuberculosis in badgers and possums show spatial similarities with regard to the spread of infection to new populations, which in all three species is a result of dispersal of juveniles approaching maturity. In all three cases this is critical to control success,

and is a major factor inhibiting development of improved control policies for tuberculosis, whereas the use of oral vaccination of fox populations has made a major difference to the success of control by substantially reducing outward spread of infection through dispersers. The argument of Carey (1985) that rabies in the eastern US persisted best in rugged terrain with a mixture of farms and forest has its parallel in possum tuberculosis with the bush-pasture margin as the area of prime importance for maintenance and transmission of tuberculosis. Spatial patchiness is a notable feature of both diseases, although because of the differences between the diseases in the lengths of both incubation period and clinical period, plus the different susceptibility to environmental influences, patchiness in tuberculosis is a much more pronounced and locally persistent feature of the disease.

There are few “models” which can be used as a guide in thinking about the likely effectiveness of control policies for possum tuberculosis, and it would appear that fox rabies comes closest to providing such a model, despite the obvious differences. If it does represent a useful parallel example, then it certainly emphasizes the difficulty of achieving effective control by a policy which relies solely on large scale periodic population reduction, and the accumulating evidence in New Zealand suggests that additional control procedures will be necessary to make greater progress in control. The notable success of wildlife vaccination as a control strategy in fox rabies supports the importance of pursuing this control option in wildlife tuberculosis as a long term control option, and the computer simulation model should in future be enlightening in evaluating this strategy.

African swine fever and rinderpest clearly do not have such strong epidemiological similarities to tuberculosis as rabies, because as shown in the consideration of these diseases the determinants of spatial and temporal patterns of disease transmission between wildlife and domestic hosts are quite different, so control policies need to be different as well. Thus consideration of wildlife disease epidemiology must take fully into account the ecology of the wildlife host(s) and the opportunities for behavioural interaction between the wildlife and at-risk domestic stock.

The main insights which these two comparisons offer relate to the flows of infection between wildlife and domestic stock. In rinderpest early concern about wildlife reservoirs of infection has given way to a dominant view that in this particular case domestic stock are the reservoirs and wildlife are the spillover hosts. This has greatly simplified regional control of the disease. African swine fever offers salutary lessons of a disease which was initially easy to limit to the wildlife reservoir species, but which for various reasons established a new and quite different transmission cycle which made it totally independent of the wildlife mammalian reservoirs and allowed it to spread to new countries and cause outbreaks. These have been persistent and difficult to control in the presence of a suitable tick vector, but easier to control in countries where there was no tick vector. Possum tuberculosis is a very clear example of a disease establishing in an entirely new maintenance host, which happens to be a very successful coloniser of diverse habitats, being probably the most geographically widespread wild animal in

both Australia and New Zealand, and one of the most numerous animals in both countries. Both population control and habitat management are therefore not easy to achieve for the possum as ways of controlling tuberculosis, and much more subtle combined control strategies will be necessary. There also remains a risk that with infection occurring widely in the possum, that one or more of the current spillover hosts could in future become a maintenance host. It can be conjectured that feral deer might be the original maintenance wildlife hosts in New Zealand, and that the possum was a spillover host which for ecological and epidemiological reasons became a very successful maintenance host. If this were true, then the sparseness of feral deer in Australia would help explain why the possum did not become a reservoir species there. For New Zealand the implications would be the importance of preventing any further species from becoming maintenance hosts, and the possible need to see feral deer tuberculosis control as an essential adjunct to possum tuberculosis control.

Parallels can be drawn with the resurgence of human tuberculosis over the last decade. Until then tuberculosis control had been one of the success stories of human medicine. Since the middle of this century the disease had been largely confined to populations living in underprivileged conditions in industrialized as well as developing countries. It could be argued that the advent of diseases such as AIDS in combination with specific behavioural patterns and environmental conditions has created an epidemiological situation which facilitates maintenance of *Mycobacterium tuberculosis* infection in specific subgroups of the population.

Relatively few of the diseases which involve transmission flows between domestic and wildlife hosts have been investigated in sufficient epidemiological detail to enable detailed comparisons to be made. The three examples chosen here for comparison were selected both because they are among the best understood, and because they represent a spectrum from rabies (which offers some of the best insights into the problems which must be faced in controlling possum tuberculosis), through African swine fever (which has parallels mainly with regard to the risks of new epidemiological patterns emerging) to rinderpest (which is at the opposite end of the epidemiological spectrum, in that wildlife are spillover hosts, and control in the wildlife is far less important than control in domestic stock). This comparison shows where tuberculosis fits in the spectrum of diseases shared between wildlife and domestic stock, and although analogies must be treated with considerable caution, the comparison does help to identify epidemiological issues which should be borne in mind in thinking about control policies.

EPIDEMIOLOGICAL STUDY METHODS FOR DISEASES IN WILDLIFE

Diseases in wildlife provide a major challenge to modern veterinary medicine. It should be noted that on a world-wide scale diseases which involve wildlife reservoir species such as rabies, bovine tuberculosis and trypanosomiasis are amongst those animal diseases which have provided modern veterinary medicine with the most difficult challenges when attempting to achieve control or eradication. This is an area where a sound epidemiological approach is

required in order to make progress. Over the last 20 to 30 years the methodology used in veterinary epidemiology has become very advanced and has been used successfully in dealing with diseases in domestic animals. Very sophisticated and successful animal disease surveillance systems have been developed for the control of diseases in domestic animals. It is now clear that these methods cannot be applied to wild animal populations without considerable modification and refinement. With minor modifications, the reduction of the number of animals has been the major method used in the control of diseases in wild animal populations. In some circumstances this can change the balance sufficiently to bring a disease under control, especially during the peak of an epidemic - when other factors also favour a reduction in incidence. However for endemic diseases the ecological forces which exert strongest influence tend to be those favouring rebuilding of the host population and maintenance of the disease within the rebuilding population, which contains a high proportion of susceptibles. Thus the more endemically stable a disease is in the wildlife population (and clearly tuberculosis in possums fits this situation), the less effective large area population control is likely to be in producing a stable low prevalence of the disease. Refinements can be introduced which improve the effectiveness of population control by differentially reducing infected populations, dispersing animals and other high risk groups. The example of fox rabies also shows the major benefits of reducing the proportion of susceptibles in the population through wildlife vaccination.

Epidemiological studies of diseases in wild animal populations require a different methodology than studies of diseases in domestic animals largely because the information which can be collected on the population as a whole as well as on individuals is subject to a number of limitations. These include difficulties in accessibility, identification, examination and follow-up of individuals. It can be very difficult to make observations on the interaction between individuals in the population as well as between them and other wild and domestic species. For these reasons it is very challenging to conduct longitudinal or cohort studies which are among the most powerful tools which can be used by epidemiologists in wild animal populations. The data collected in most epidemiological studies of wild animal populations will be subject to sampling error or measurement bias to a larger extent than in the case of the study of human or domestic animal populations. Often there are many unknown risk factors as well as confounding factors which may influence the results, but cannot be measured and controlled for in the analysis. For these reasons, despite their major limitations in providing an understanding of the dynamics of the disease, cross-sectional studies have been widely used to study the epidemiology of diseases in wild animals - primarily because they are relatively easy to conduct. The pathogenesis of diseases in wildlife is usually studied under experimental conditions, which means the findings will often misrepresent the course of disease in the wild. Epidemiologically sound observational studies are therefore necessary to obtain information on the pathogenesis of the disease under uncontrolled conditions. Ecologists are used to dealing

with the problems involved with studying wildlife populations. Some of those methods are likely to be applicable in veterinary wildlife epidemiology.

EVALUATION OF THE STUDY METHODS ADOPTED FOR TUBERCULOSIS

Longitudinal Study of Bovine Tuberculosis in Possums

The longitudinal study approach is one of the most powerful techniques which can be used in wildlife disease epidemiology. It is possible to estimate the incidence of disease, and the temporal relationship between hypothesized causes and their effects can be tested. The problems involved with this methodology include difficulties in the follow-up of individual animals. They are also very costly and labour intensive. One of the major difficulties in the study reported in this thesis was related to the diagnostic methods, which were poorly sensitive for detecting possums infected with *Mycobacterium bovis* and only moderately sensitive for detecting clinically diseased possums.

The results of the data analysis for the longitudinal study showed that prevalence and incidence levels of clinical disease in a possum population with endemic infection vary significantly between seasons and between years. Adverse environmental conditions such as cold and rainy weather, limited availability of food resources and suitable den sites resulted in an increase of the incidence of clinical disease. These factors also limit survival of clinically diseased possums. A possum which develops clinical disease under rather favourable environmental conditions may be able to survive for extended periods. If it is exposed to rain, wind and low environmental temperature, it would not be able to survive for more than maybe 1 or 2 months from the onset of clinical disease. Therefore, during cold and rainy weather there could be many clinically diseased and infectious animals in the populations for a relatively short duration. Under more favourable conditions only a few clinical cases would be present, but they could survive for extended periods. It appears from the longitudinal study that this variation in survival is an important feature of the dynamics of endemic infection in possum populations. Prior to this study information from experimental studies suggested that the period between the onset of clinical disease is very short (O'Hara *et al* 1976, Corner and Presidente 1980, 1981). The pathology of the disease could not be studied in detail, but many animals with clinical disease had lymph node abscesses which discharged through the skin. Most tuberculous possums had lesions in the lung tissue and /or peripheral lymph nodes.

The data collected during the study provided basic information on the importance of the different potential disease transmission paths. The evidence suggests that transmission from infected adult female possums to their offspring is one of the major factors in the epidemiology of the disease. Pseudo-vertical transmission could be responsible to a significant extent for maintenance of the disease within a possum population as well as for the spread of infection to other populations. Young infected animals can remain subclinically infected for an extended period, the length of which remains quite uncertain. It can commonly be a number of months, and may in some cases be as long as 1 to 2 years - or even longer. Infected animals appear

likely to develop clinical disease under conditions of stress induced by the environment, feed shortages or through behavioural stresses such as those occurring at mating or dispersal. Once they are clinically diseased these possums can infect other animals, including both domestic stock and other wild animals as well as possums. Especially during the mating season a clinically diseased adult male possum can readily transmit infection to susceptible adult female animals in association with the courting and mating process, as well as to susceptible adult males during agonistic behaviour. From the data of the longitudinal study it appears that these patterns of social interaction comprise the main transmission mechanisms in the epidemiology of *Mycobacterium bovis* in possums. Indirect transmission - on pasture during feeding, during sequential sharing of den sites and through contamination of sites used for marking - probably occurs, but on the evidence from the longitudinal study seems to be of much more limited importance than direct methods of transmission.

The longitudinal study also provided information on transmission of infection to other domestic and wild animals. The two restriction-endonuclease strains of *Mycobacterium bovis* which dominated in possums were also found in cattle and wild pigs, and a minor strain was found in a ferret. It is unlikely that during the study infection was transmitted from any of these species to possums. In this scenario cattle, wild pigs and ferrets are more likely to be spillover hosts. Evidence from the study also suggests that terminally ill and possibly dead tuberculous possums are important sources of infection for spillover hosts including cattle.

Cross-sectional Study of Bovine Tuberculosis in Possums

Cross-sectional studies can be used to obtain baseline epidemiological information, but they can only provide a static picture of the disease situation. They can be relatively easily conducted in a wildlife situation. The Hauhungaroa study had been conducted at relatively low intensity over too large an area, which resulted in a loss of detail of information at the farm-level. Also the different areas which were included in the study were quite heterogeneous in that some of them had been subject to possum population control, the habitat was different and the possum populations were sampled at different seasons, without regard for possible seasonal variation in prevalence. Especially the last factor complicated the analysis considerably in that the possum populations were compared at different stages of their yearly biological cycle. The diagnostic method for identification of tuberculous possums was poorly sensitive because laboratory confirmation was not based on culture, which is the most sensitive method available in routine diagnosis.

The results of the cross-sectional study suggest that on a larger geographical scale about 2% of possums have clinical tuberculosis. Prevalence in local clusters can be up to 20%. Tuberculosis prevalence was higher in adult possums than in immature possums. In immature possums males were more likely to show clinical disease than females, which was not the case for adults. Breeding female animals were more likely to have tuberculous lesions than adult females without a pouch young. A large proportion of possums with clinical disease had lesions in the respiratory tract and/or peripheral lymph nodes. The presence of localized

tuberculous lesions did not appear to affect condition to a significant extent. The study also allowed a statistical examination of the relationship between possum tuberculosis prevalence and cattle tuberculosis incidence. The correlation between the two measures was poor. This result should be interpreted with care, because it was not exactly known when and how long cattle had been kept in paddocks adjacent to the areas which had been sampled for the presence of tuberculous possums. Also, the data collected did not allow possum tuberculosis prevalence to be estimated for a particular farm, because in most cases only a small area had been sampled. What can be said is that cattle from farms adjacent to possum populations with endemic tuberculosis were more likely to have reactors than cattle from properties where no tuberculosis was found in the associated possum sample.

The longitudinal and the cross-sectional study complement each other. The longitudinal study provides detailed information about the epidemiology of *Mycobacterium bovis* infection in a possum population. It is possible to obtain information about the infection dynamics and transmission patterns. On the other hand the cross-sectional study provides an impression of the epidemiology of the disease at a larger geographical scale.

Both studies come to similar conclusions on points which they could both measure - such as that the disease is clustered in space and that the predominant sites for tuberculous lesions include the respiratory tract and peripheral lymphnodes. The results of the longitudinal study did not confirm that immature male possums were more likely to show clinical lesions than immature females. A meaningful conclusion cannot be drawn from this finding in the longitudinal study because only few immature animals with clinical tuberculosis were identified.

Case-control Study of Tuberculosis Breakdowns in Cattle Herds

A case-control study approach is typically used when studying rare diseases. The number of breakdowns in cattle herds due to tuberculosis infection is still a relatively rare occurrence and in a case-control study it was possible to include the maximum number of case herds in order to achieve meaningful results in the analysis. Matching of cases and controls allows the control of confounding factors. In this study the presence of infection in local possum populations and the type of farm enterprise were considered major confounding factors. Therefore whereas a typical case-control study has all controls chosen on the same basis, in this case two differently chosen controls were used for each case. Each case herd was matched with one control herd of any enterprise type from the surrounding region (random control) and a second nearby control farm with the same type of enterprise (matched control). The matched case-control approach should allow the analysis to minimise the confounding effects of the factors which have been used for matching. Controlling for the risk of infection from local possum populations by selecting controls from the vicinity of case herds may not have been a very effective way of matching because tuberculosis infection in possum populations is extremely clustered in space and the risk of infection for local herds may vary considerably between neighbouring herds. However without extending the study to include examination of possum populations on candidate farms, this was the best matching which could be attempted.

The study provided some insight into the importance of transmission between and within species, for both possums and cattle. The results suggest that in the Waikato area possum to cattle transmission may be of importance for herds which are located closer to the tuberculosis endemic area. In these herds it is more likely that infection is present in the local possum population. But it is also possible that these farmers were more likely to have bought animals from the tuberculosis endemic area which may have been infected with *Mycobacterium bovis*. It appears that cattle-to-cattle transmission by contact between neighbouring herds (contiguous spread) is unimportant under the management conditions in the Waikato area. No inferences can be drawn regarding the importance of domestic and wild deer as no suitable data was available which could have been included into the analysis.

The study had been conducted because there had been a number of breakdowns in the area and it had not been possible to explain them by demonstrating the presence of tuberculous possums on the farm, since extensive cross-sectional studies had failed to detect any tuberculous possums, despite a strong suspicion that they were present in the area. (Subsequently infected possums were found in the area). The results of this study suggest that exposure to infection in local possum populations is not the dominant cause of breakdowns in these herds and that the occurrence of clusters of outbreaks does not demonstrate that they were caused by infection in local possum populations. Farmers who had a breakdown were more likely to be among those who followed herd management practices considered to put farms at risk of buying in infection in purchased stock. This finding and the fact that many farmers had only limited knowledge about the disease suggests that there is a need for an educational effort and incentives for farmers to change their purchase policies in order to minimise the risk of introducing infection. The case-control approach used here provided a method of provisionally distinguishing the relative importance of the two major transmission hypotheses fairly quickly and inexpensively - something which would have been much more costly to do by any alternative investigational approach.

RESEARCH TECHNIQUES

It was possible to apply a number of research techniques to the analysis of the data collected in the various studies included in this project which have not previously been used widely for epidemiological investigations. Based on the type of data they can be broadly grouped into techniques which can be used in the analysis of difficult longitudinal data, data including a spatial component and/or temporal component, and multivariate analysis techniques.

Analysis of Difficult Longitudinal Data

Data collected during a longitudinal study of diseases in wildlife populations is characterized by losses to follow-up and by new recruitments. Losses to follow-up can represent deaths as well as emigration. New recruitments can be animals which were born locally or can be animals which immigrated into the study area. In ecology, capture-mark-recapture techniques are used to estimate parameters describing the dynamics of such populations. The main techniques

which were used in this analysis include a set of models developed by Otis *et al* (1978) and the Jolly-Seber model (1982). Both techniques allow estimation both of population size for each point in time when the population was sampled and of survival between sequential visits during data collection. Capture-mark-recapture techniques are finding wider use in epidemiology, and have been used recently to estimate population at risk for sexually transmitted diseases (Rubin *et al* 1992). In the longitudinal study reported here they were essential to reduce bias in estimating the population at risk and entry/removal rates.

Analysis of Data including a Temporal Component

Data with a true temporal component is collected typically during a follow-up study and possibly during a retrospective study. The variable measured is the time elapsed until occurrence of a particular event of interest. This type of data may be complicated by four factors: loss of individuals during follow-up (right censoring), individuals which do not develop the outcome of interest during the period of the study (right censoring), new recruitments during follow-up (left censoring or staggered entry) and change of attributes of an individual during follow-up (time-dependent variables). Survival analysis (or event-history analysis, as it is called in the social sciences) provides a methodology which can take account of these factors. Cox's proportional hazards regression model adds a powerful multivariate analysis tool to survival analysis. These techniques have proved valuable both in the longitudinal and the modelling study.

Analysis of Data including a Spatial Component

Proximity is one of the major factors in the epidemiology of contagious disease processes. Over the last 5 years the storage and management of spatial information in a computer has become more practical with the advent of software termed geographic information systems (GIS). It is now possible to use these systems to provide data which can be used in epidemiological analyses. The main areas of application include the preparation of descriptive maps of disease occurrence and of relevant features in the environment such as topography and vegetation, as well as the quantitative description of spatial relationships between features. The descriptive mapping component of such systems was used in the longitudinal study to display and compare the overlap of the areas which were used by possums infected with individual strains of *Mycobacterium bovis*. Distances to features of potential importance such as herds with tuberculosis infection or the distance to the tuberculosis endemic area in the case-control study were used as potential risk factors in the multivariate analysis of the particular study. In the longitudinal study analyses were conducted to identify risk factors which differentiate den sites used by possums with tuberculosis and den sites only recorded as used by non-tuberculous possums. These risk factors included locational characteristics such as slope and height above sea level, which were retrieved using spatial database management techniques such as overlay operations from the relevant information layers.

Analysis of Data Including Both a Temporal and Spatial Component

Clustering of disease occurrence in time and space is typical for an infectious disease process. The management of joint temporal and spatial data is extremely complex. It is an area which is still under development. At the moment methods such as Mantel's time-space regression method can be used to investigate such relationships. The general view appears to be that this technique is not very sensitive. In this analysis the use of different transformations of the time and space variables resulted in inconsistent results. The results of the analysis were quite difficult to interpret. The use of time-space interaction analysis gave hints of transmission associations which were helpful in formulating an overall hypothesis of disease transmission, but it is considered that at this stage of development of the techniques they must be used very cautiously as purely an exploratory tool.

Analysis of Multivariate Data

A number of multivariate analysis techniques were used in the different studies included in this project. Correspondence analysis and multidimensional scaling both allow the visual description of relationships between variables. Correspondence analysis is only applicable to categorical data. It places each category as a point in a type of scatterplot or 'map'. The relative positions of the category points in this map indicate certain levels of similarity or association between categories. Multidimensional scaling is used for the analysis of preference data such as used in the case-control study to describe the interviewee's self concept. Both techniques are only useful for descriptive analysis and the interpretation of the distances between points and the meaning of the axes in the map largely depends on the judgment of the reader.

In veterinary epidemiology multivariate data is commonly analysed using a two step technique. In a univariate analysis the data is first screened for statistically significant relationships between the dependent variable and each potential risk factor. This step is then followed by a multivariate analysis which includes the variables which were statistically significantly associated with the outcome variable in the univariate analysis. The multivariate analysis can be conducted in a number of ways. One option is a stepwise regression approach which results in a final regression model purely based on statistical grounds. These models do not provide a comprehensive view of the causal web they are trying to describe. Path analysis is a technique for multivariate data analysis which allows the researcher to combine biological understanding of the problem under consideration with the power of statistical techniques. Both path analysis techniques which were used in the case-control study, standard regression path analysis and structural equation modelling, involve the development of a hypothetical path model by the researcher which is then tested using statistical methods. Both path analysis approaches are very useful for exploring the causal structure of the data. Compared with standard regression path analysis, the structural equation model is more powerful and has more options. Its advantages are that it provides a quantitative estimate of overall fit of the path model, latent variables can be included, the model does not have to be recursive and direct as well as indirect effects can be quantified. Its disadvantages include the complexity of the

modelling process compared with regression path analysis, the methodology for dealing with categorical variables is not yet fully developed and sample sizes have to be quite large to estimate stable and meaningful path models. Classification tree analysis is another method which can be used in the multivariate data analysis step. This statistical technique is used to develop an hierarchical representation of the data space using a binary tree. This "decision tree" can be readily interpreted and integrated into a decision framework. Classification tree analysis produces the best results when large sample sizes are available. It is of limited use for causal analysis.

COMPUTER SIMULATION MODELLING AS A TOOL IN DISEASE CONTROL

Computer simulation modelling provides an essential tool for investigating the epidemiology of a disease and for comparing potential disease control strategies. As pointed out by Martin *et al* (1987), in addition to its other uses the modelling exercise brings to light important deficiencies in the available body of data. Consequently a model can be used to demonstrate the importance of the missing data and to direct data collection efforts (Hurd and Kaneene 1993). A simulation model which provides an adequate representation of the epidemiology of a disease can be used to test the effect of different control strategies. Anderson (1991) emphasizes that the transmission dynamics of the infectious agent in question and its distribution and abundance within the host population, which can both be represented in simulation models, have a major influence on the intensity and frequency of control intervention required in order to halt transmission. The effectiveness of new methods of disease control can be assessed in order to decide which of a number of options should be considered for medium- or long-term research efforts.

One of the objectives of this epidemiological project was to develop a computer simulation model describing the epidemiology of endemic bovine tuberculosis infection in a local possum population. A systems analysis approach targeted at generating knowledge about the system under study was adopted in order to provide the information required for the development of the simulation model. At the beginning of the study a general overview of the system was obtained by conducting a literature review, and by communication with people working in the subject area. This general information was then used to design and implement the observational field studies as the next stage in the system analysis. These sources of information generated the knowledge which was used to develop the simulation model. The model is the synthesis of the information available through the two first stages of the system analysis. Once the model is developed a feedback of information to the two lower levels can occur, resulting in design of studies to test hypotheses generated by the model or formulation of hypotheses which can be tested directly by the simulation model. A number of studies have already commenced as a result of data requirements identified as necessary for the model (e.g. survival of *Mycobacterium bovis* in the environment, and behavioural interaction between possums and domestic livestock).

As explained above, the knowledge about the system currently implemented in the model is based on a detailed literature review and analysis of data from a longitudinal and a cross-sectional study of *Mycobacterium bovis* infection in possum populations. In combination, both field studies provided an epidemiological framework for generating hypotheses about important factors and their interaction in the dynamics of tuberculosis infection in wild possum populations. The development of the simulation model required qualitative knowledge about these factors as well as parameter estimates describing quantitative relationships between factors. The longitudinal study provided this information. This epidemiological knowledge was used to design a Monte-Carlo simulation model as described by Morris (1976). The model adequately represents the epidemiology of bovine tuberculosis infection in local possum populations. Some of the quantitative estimates of the model parameters may have to be refined as soon as new information from other field studies becomes available. Currently there is no independent field data available, against which the model could be validated. A number of options for disease control have been tested using the model, but these results have to be interpreted keeping in mind the early stage of model development which has so far been reached and uncertainty about some of the parameter estimates. The model will be developed further in order to simulate the impact of control at a larger geographical scale and eventually will form part of a planned decision support system for tuberculosis control in New Zealand.

Simulation and Disease Control in Animal Populations

A major objective of developing a disease simulation model is to evaluate the effect of different control options on the pattern of disease within a population. A number of simulation models have been developed for the epidemiology of diseases in wildlife. Results from a wide range of models on epidemiology of rabies in fox populations have been published. These include a small number of stochastic models using a modelling approach similar to that used for the development of the current model. A model developed by Preston (1973) was used to investigate the possible interactions in a rabies controlled population. It included a spatial component, but did not allow extensive evaluations of disease control strategies. Voigt *et al* (1985) developed a stochastic simulation model of rabies (Ontario Rabies Model) which simulates the spatial and social behaviour of red foxes. It was specifically designed in order to assess control strategies and evaluate tactics of oral vaccination. This model has been used extensively for planning rabies control operations in the Canadian province of Ontario (MacInnes *et al* 1988). Experiments with the Ontario Rabies model have improved the understanding of temporal and spatial patterns of rabies incidence (Tinline 1988). Smith and Harris (1989) used the Ontario Rabies Model to investigate the potential pattern of spread of rabies in Britain from a single source of infection. They came to the conclusion that they had to develop a new model, because the Ontario Rabies Model could not be applied to the detailed simulations required in the British situation. Smith and Harris (1991) reviewed 18 of the available rabies models and noted that most of the models were designed to reproduce various factors involved in the spread of rabies, rather than attempt to examine possible means of

controlling the disease. Based on this experience they developed a discrete time simulation model with monthly steps modelling the spatial and social behaviour of individual foxes. This model uses numeric maps of the simulation area defining topographical barriers and locations of fox families. It is also possible to define the areas where fox control will occur. In their paper the authors concluded that their model provides a valuable tool in planning rabies control operations and helps to unite the bodies of theoretical and practical knowledge on rabies spread.

Various tuberculosis models have also been developed. A deterministic simulation model of the epidemiology of bovine tuberculosis infection in badger populations has been developed by Anderson and Trehwella (1985). Barlow (1991a) developed a deterministic simulation model with stochastic components for bovine tuberculosis in New Zealand possum populations. He used this model to evaluate a number of possible control measures (Barlow 1991b).

Strategies for Pest Management

An assessment of methods for control of possum populations requires some background information on the theory and practice of pest management. Putman (1989) reviewed approaches which have been taken to pest management. He writes that a population reduction (short of extermination of the species which causes the damage) is bound to have only short-lived success. Therefore it must be sustained and possibly repeated year after year. He adds that artificial population reduction may release the density-dependent brake on population growth resulting in increased reproduction and reduced natural mortality. Putman writes that even if we cannot prove density-dependent effects on recruitment and survival, local reduction of the species in one area is rapidly compensated by immigration from outside. In many situations control of a pest problem by direct reduction of population size may be the only realistic option available and may prove effective, if carried out intelligently and with full understanding of the underlying dynamics of the pest population.

Fischman (1985) points out that for control of rabies in wildlife, classic population reduction methods including trapping, poisoning, gassing and hunting have proven expensive, objectionable, often not effective and, even where effective, require periodic repetition. He advocates the use of chemosterilants as offering an attractive alternative which would also be much more acceptable to the general public. Fischman suggests that if a wildlife population is experiencing a rabies epidemic, a bait should be delivered containing an oral vaccine to halt the epidemic and an antifertility compound lowering the fertility at the next breeding season. The antifertility compound would have to be used repeatedly to keep the population at a predetermined level.

Conway (1976) described the following major techniques for vertebrate pest control: culling (use of chemical compounds, trapping or shooting to kill or remove pests), biological control (use of natural enemies, either by augmenting those present or by introducing new species), habitat manipulation (use of agricultural or other practices to change habitat available

to pests), exclusion (use of fences or other barriers to prevent entry) and plant or animal resistance (breeding of animals and crop plants for resistance to pests). More recently fertility control has been taken into consideration as a more humane method of population reduction (Bomford 1990).

Pest Control and Animal Population Dynamics

In order to make a decision on the optimal control strategy it is necessary to understand the mechanisms which determine the population dynamics of the species to be controlled. This includes knowledge about the following characteristics of a population: its increase and decrease, its fertility, mortality, dispersal and immigration rates. The effect of perturbations on the population dynamics has to be understood. It has been demonstrated that conventional control by increasing mortality results in increased reproduction and survival in the remaining animals, and possibly increased immigration and decreased dispersal. The population would therefore recover relatively quickly to its original density.

MacArthur and Wilson (1967) and Pianka (1970) defined the r - K continuum to categorize species according to the bionomic strategies reflected in their population dynamics. r - strategists are opportunists, selected for maximum food intake by the exploitation of their ephemeral habitats. Southwood (1977) writes that in contrast K -strategists maintain a steady population at or near the carrying capacity of the habitat; they are in equilibrium with their resources, whose renewal they do not adversely affect. Recruitment, migration and mortality rates are low, and if their numbers are reduced to low levels, they become extinct. On the other hand the population dynamics of r -strategists are characterized by 'boom and bust' as Southwood writes. This strategy is dominated by large-scale migration, and new populations are continually developing from a handful of colonisers. Southwood adds that a species may be able to move along the r - K continuum to a limited extent, if the ecosystem it lives in changes. He defines in addition to r - and K - pests the class of intermediate pests who are generally held at a level lower than carrying capacity of their habitat by the presence of natural enemies. These animals will achieve pest status if they are introduced into new areas free of their natural enemies (for example the gypsy moth after introduction to North America). Southwood writes that r -pests are always so numerous that they may in certain places destroy their habitat (he cites the example of smallpox virus and *Bacillus anthracis*). They typically show a high migratory tendency, essential for their movement from one 'dying habitat' to a new one. K -pests usually are relatively large organisms such as the African elephant which began to damage its habitat after man confined the species to game parks. The African trypanosomes and their vectors, tsetse flies, are considered to be typical examples of K -pests. Pianka (1983) emphasizes that no organism is completely r - or K -selected. The r - K selection should be thought of as a continuum and an organism's position along it can only be determined in a particular environment at a given point in time. The concept of r and K selection has been criticized by a number of authors as too simplistic and too inadequate to explain variation in life-history traits in nature (Fleming 1979).

Southwood (1977) postulates that for r -pests a powerful control strategy is to decrease immigration. Population levels of these species are always fluctuating. It is the nature of r -strategists that after reduction control the population will quickly 'bounce back'. Southwood *et al* (1974) write that r -strategists have very efficient mate finding tactics at all densities, and therefore have a very low extinction point. Stenseth (1981) developed a conceptual framework for formulating pest control strategies on the basis of knowledge about the pest's demography, its habitat, and the control strategies available. He considers the extremes in the r - K spectrum of population selection. Stenseth concludes that if there are methods for reducing immigration into empty patches by almost 100%, regardless of the population demography of the pest most of the available resources should be spent on reducing immigration. If there is no particularly effective method for reducing immigration available, different methods should be used for r - and K - selected species in the same environment and habitat. With r -selected species most economic resources should be devoted mainly towards reducing reproduction rather than increasing mortality. For K - selected species Stenseth recommends directing most resources towards reducing dispersal but some for increasing mortality. In particular reduction in reproduction is likely to be inefficient for controlling K - strategists. Whereas Southwood (1977) recommended the use of pesticides for controlling r -selected species, Stenseth suggests that because most pests are r -selected this may explain why the extensive application of pesticides has proven inefficient in protecting the world's food supply against destruction.

Tyndale-Biscoe (1979) classifies the brushtail possum as an r -selected species, as it is extremely adaptable and occurs in a wide range of habitats. How (1978) reviewed the population strategies of four species of Australian possums and he considered *Trichosurus vulpecula* Kerr to be closer to the r - end of the r - K continuum than the other three species. Given the above theories about population strategies it would appear that for possum population control population reduction clearly is unlikely to be fully effective alone.

Modelling of Infectious Diseases

Models of infectious diseases are frequently used to test the effectiveness of disease control strategies in animals and humans. Most of these models are based on sets of abstract mathematical equations. In order to understand the problems involved with developing and using such *mathematical* models it is necessary to describe their main components.

Mathematical models depend to a large extent on an estimate of the basic reproductive rate of disease \mathbf{R}_0 which is defined as the average number of secondary infections attributable to a single infectious case introduced into a fully susceptible population. This principle is one of the most important concepts in mathematical epidemiology (Anderson 1991). Under the theoretical 'mass action' assumption \mathbf{R}_0 is a function of population size or density. The goal of mathematical modelling is to estimate a threshold population density for disease maintenance (Fine *et al* 1982). Given this figure it is possible to calculate minimum host densities required for the disease to remain endemic within a population. Using this approach Anderson *et al* (1981) estimated threshold values for maintenance of rabies infection within fox populations.

Voigt *et al* (1985) found that in Canada rabies was persisting at fox population levels lower than the ones where it disappeared in Europe. MacDonald and Voigt (1985) point out that the contact rate is the most fundamental ingredient of a rabies model. It is not a constant, but a complex function of the social organisation and density of the vectors, and thus the frequency of meetings amongst them. They write that in the stochastic world of complex animal populations, measurement of contact rate is notoriously difficult; the frequency of meeting between individuals (and hence the potential contact rate of disease) is a reflection of their population density, social organization and their ecology. MacDonald and Voigt add that it is known that the spatial organization of vectors influences the local rate of contact, the reintroduction and the long-range dispersal of vectors. Mollison (1987) emphasizes that the major problem in developing a mathematical model for disease dynamics lies in estimating the value of R_0 even in undisturbed populations or worse in predicting its new value under some control strategy. He writes that in the case of rabies in foxes because of a social disruption caused by culling part of the population, R_0 may not change in proportion to population density. In a more detailed discussion of the implications of control on model parameters Mollison (1985) points out that on the one hand, by reducing fox density competition for available food will be reduced and therefore contact will be less common. But on the other hand, as mentioned above, families will be broken up by the culling. This social disturbance will result in more contacts. He adds that with diseases such as measles and whooping cough where the population density is not affected by the disease the reproductive rate can be extremely low and the effects of heterogeneous mixing gain importance. Mollison (1986) writes that estimating R_0 resists any general quantitative approach. He states that species do not have an absolute value of R_0 but a value relative to a particular habitat or ecosystem.

Anderson (1985) developed models for both rabies in foxes and for bovine tuberculosis in badgers. He writes that both diseases show very different epidemiological patterns within their respective host populations. Rabies is an epidemic disease exhibiting large fluctuations in prevalence from year to year, with an overall relatively low number of rabid animals. In contrast, bovine tuberculosis in badgers appears to be stable, with the prevalence remaining relatively constant through time and a high standing crop of infected animals. Anderson suggests that the characteristics of the epidemiology of rabies in foxes would suggest that control should be relatively easy by taking advantage of the instability of infection within host populations. But this can only be achieved given an intense and sustained control effort in areas of good fox habitat to maintain fox populations at a sufficiently low level of abundance to eradicate disease. Ideally control should be applied between epidemic intervals. In the case of bovine tuberculosis in badger populations Anderson suggests that eradication of infection may require the eradication of the host species. Summarizing, he writes that rabies control in fox populations requires less intensive control effort at one point in time, but sustained effort at frequent intervals. On the other hand bovine tuberculosis in badgers needs intense control effort to substantially reduce host abundance, applied at relatively infrequent intervals. The

discussion earlier in this chapter of this same issue suggests that although there are differences between rabies and tuberculosis in wildlife, there are also important epidemiological similarities and that these two diseases merit continuing comparison since investigation of each can provide useful insights into control options for the other.

For rabies in foxes, Tinline (1988) used the 'Ontario Rabies Model' and came up with some factors which he suggests may affect persistence of infection within southern Ontario. He firstly hypothesizes the existence of 'rabies units' which form distinct spatial clusters and temporal patterns of infection. He suggests that there must be a balance between good and poor habitat to maintain spread but slow it down sufficiently to allow populations to rebuild behind epidemics. The conclusion was that by concentrating control on particular units with good habitat it may be possible to break the chain of infection. He called this spatial heterogeneity at a macroscale, which he thinks could well be the primary mechanism causing persistence of rabies in southern Ontario. Tinline postulates a threshold size for rabies to persist in certain areas. On a macro scale he found that using the 'Ontario Rabies Model' he could only simulate an endemic situation if the area being modelled was at least 4000km². Other factors which Tinline investigated using the 'Ontario Rabies Model' included the interaction between reproduction and mortality, the incubation period and the interaction between contact rate and population density. Obviously the epidemiology of fox rabies is distinct from the epidemiology of bovine tuberculosis in possums, but this example shows how a stochastic simulation model can be used to improve the understanding of the epidemiology of a disease in wildlife populations to a significant extent.

Modelling and Planning for the Control of Bovine Tuberculosis in New Zealand

Norton (1988) advocates a decision analysis / systems analysis approach to vertebrate pest management. The method includes two key concepts. A decision model describing the four major factors affecting decision making - the pest problem itself, the control options available, the decision maker's perception of the problem and his or her objectives. Each of these factors has to be considered during the actual decision making process. The second key concept concerns development pathways. Norton sees the direction of the agricultural and pest management development process as influenced by factors such as economic forces, agricultural policy, technological as well as previous development.

In New Zealand the decision process for implementing control of tuberculosis in possum populations was mainly based on the finding that once a local possum population with endemic tuberculosis infection had been reduced, the incidence of infection in cattle grazing in habitat overlapping with possum habitat dropped significantly. It was noted that a few years after possum population control, cattle reactor numbers increased again. It was therefore concluded that repeated culling of possums was a suitable method for controlling the disease locally. The other dimension of the problem was that infection was also spreading between local possum populations. It was realized that it was very difficult if not impossible to contain the disease within local possum populations. Given the cost of these possum control operations the

Ministry of Agriculture and Fisheries decided that the primary objective of tuberculosis disease control was the containment of *Mycobacterium bovis* within endemic areas. From a population theorists' point of view, both the rapid recovery of population numbers and the fact that possums are a good vehicle for spreading a disease spatially are characteristics which are probably fairly typical of *r*-selected species.

Given the above situation, mathematical models were developed to provide information for guiding and justifying disease control management decisions at a national level. Although various models have been developed to represent aspects of possum tuberculosis in New Zealand, the one which comes by far the closest to representing the field control problem and the only one which has been used in practical policy formulation is the model developed by Barlow (1991a, 1991b). This model has evolved substantially over a period of years, and has proved very informative as a guide to development of control policies. The first version was used to lay out a strategy for local possum control and for preventing spread of infection from endemic areas. For local possum control Barlow writes that the results of simulation analyses suggest that widespread poisoning operations, or single intensive control operations followed by 'maintenance control' are highly effective and offer the only means for rapidly reducing tuberculous possum density. He suggests that sterilization as a control measure is less effective than culling, and vaccination would be least promising. It was also concluded that as long as diseased possums can immigrate, it is impossible to eradicate the disease. But Barlow suggests that the disease will not spread if the population is maintained below the threshold for disease persistence. This is a conclusion which has also been used extensively to justify destruction of foxes for rabies control. Yet control of fox populations through population reduction has not been successful. Barlow also states that immigration of susceptibles has relatively little effect on the disease recovery rate following control. Stochastic effects at a small scale may have a large impact on the outcome of a control operation. Therefore it seems appropriate to express the results of comparisons of control options using a simulation model in terms of probabilities of achieving control/eradication, as well as expected prevalence, which hides the stochastic variability in the system. This would give disease control managers critical information which can be used in the decision process. This type of information is difficult to provide using deterministic mathematical models, but can be easily produced by a Monte-Carlo simulation model.

There is some evidence from a major field study of post-control population and tuberculosis dynamics which suggests that a control strategy combining an initial population reduction of at least 70% with regular maintenance control as recommended by Barlow cannot maintain low possum numbers for long enough to prevent recovery of tuberculosis disease levels in some areas (Hickling 1991). In this particular study tuberculosis prevalence dropped from pre-control levels of 2.3% (N=830) to 1.2% (N=489) for aggregated data from the post-control surveys. This reduction was not statistically significant ($p=0.17$). Tuberculous possums were generally found in the same locations as during the pre-control survey. In a different type

of habitat with a population free from tuberculosis infection it was found that after a control operation which produced an 88% successful population reduction, the local possum population appears to require at least 8 years to reach pre-control levels. This agrees with predictions of Barlow's mathematical model (Brockie *et al* 1991).

Barlow (submitted) developed a variation of the basic model which allows the simulation of spread of bovine TB between local possum populations. One of the major conclusions from this model was that a 3km 'buffer' zone with reduced possum density surrounding an endemic area would reduce the number of possums dispersing into the non-endemic area by effectively moving the source of possums 3 km further back. Knowledge about dispersal behaviour of possums with regard to distance and the role of local density as a factor influencing direction is still very limited. But it has been suggested by Cowan and Rhodes (1988) that areas of reduced population density while not necessarily exhibiting a 'vacuum effect' may in fact promote improved survival of infected dispersing juveniles.

One of the major conclusions from Barlow's model was that 'spot control' of locations in non-endemic areas, where cattle test positive for tuberculosis, in combination with a low possum population density buffer zone prevents spread of the disease. Cattle reacting to the tuberculin test would be used as indicators for the presence of tuberculous possums. The assumption behind this strategy is that the probability for transmission from possums to cattle is more than 0.10. Yet, the validity of this assumption has not been adequately verified. Before these conclusions are used as the basis for policy decisions, the sensitivity of the model using a range of transmission probabilities from possums to cattle should be tested. If it were that spatial spread of *Mycobacterium bovis* infection within possum populations does progress faster than local cattle populations can pick up the disease from infected possums, then relying on "spot-control" would be very dangerous. At present the answer to this question is simply not available. It may also be that spot control offers benefits, but for reasons which differ substantially from those which underlie the model analysis. For the case of fox rabies Bacon (1981) points out the dangers associated with the delay in detection of rabid foxes which may lead to misjudging the progress of the 'infection wave front'. Mollison (1986) notes that occasional long distance dispersers have a disproportionate importance for spread and therefore for attempting control.

Past experience with the maintenance of infection within endemic areas and the spread of bovine tuberculosis infection in New Zealand has shown that the conclusions which can be drawn based on mathematical models are limited. Development of a disease simulation model requires converting a biological system into an abstraction of the same system which can be implemented on a computer. Commonly the mathematical researchers who develop the model do not have direct field experience with the disease problem. In such cases, validity of the model structure can only be ensured if model logic and mechanisms can be explained to non-mathematicians, and evaluated in detail for biological validity. With mathematical models it is often attempted to represent a complex biological system with an abstract set of mathematical

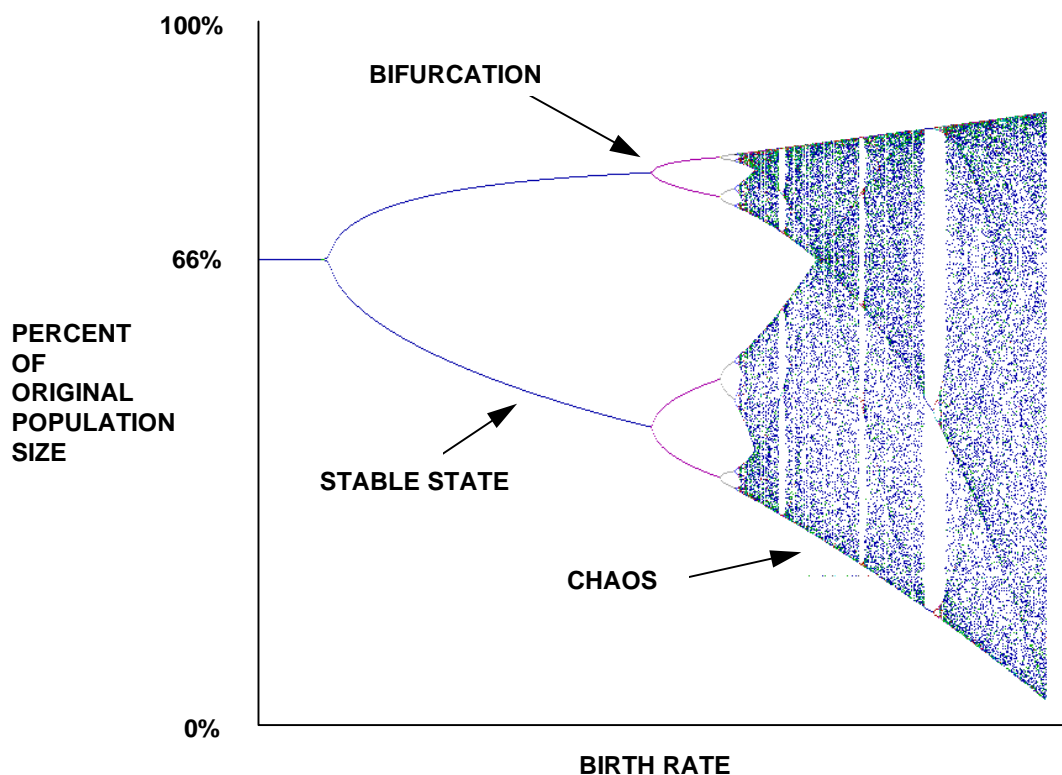
equations, which non-mathematicians find very difficult to understand. Some of the problems with false predictions produced by Barlow's earlier models probably are attributable to these difficulties which are inherent to mathematical modelling process. Also, when Barlow developed his first models, the understanding of the epidemiology of tuberculosis infection in possum populations was much more limited than it is today. There is thus a need to ensure that any models used for policy formulation adequately represent biological reality, and that both developers and users understand each other very fully. Even more important, when technical knowledge of a disease is fairly limited it is only possible to build models which reflect that level of knowledge, and to evaluate control options in broad terms. As knowledge grows, policy decisions can become more precise and the models used to support such decisions must reflect the increased level of understanding.

The model described in this thesis does not yet allow valid comparisons of disease control strategies. The simulation results so far have shown the importance of sensitivity analysis in combination with model verification/validation. Yet, the structure of the model does appear to represent the current understanding of the epidemiology of tuberculosis infection in possum populations adequately. Using sensitivity analysis it was possible to clearly identify the areas which need improved estimates.

Simulation Models of Infectious Diseases in Epidemiological Research

As has been pointed out by Bradley (1982), the conclusion both from reviewing the literature and from experience with developing this simulation model is that for real progress to be made, both the mathematical modeller and the epidemiologist must have "mud on their boots". It is one of the dangers with using models in an operational setting that they are expected to produce accurate predictions of what is going to happen in the future. Nonlinear modelers have shifted their emphasis away from prediction towards varying different variables in order to learn about a system's critical points and its resistance to change (Briggs and Peat 1989). Prigogine and Stengers (1984) point out that individual events can easily affect the direction of the behaviour of a system at a macroscale. They refer to bifurcation regions such as the ones produced by logistic growth equations where the behaviour of one individual can upset the global state of the system and produce unpredictable behaviour (see figure 109). Assuming average behaviour for individuals who are part of a system as is done in mathematical modelling disregards the importance of variation in effects at a microscale. Monte-Carlo models such as the one developed for this thesis are sensitive to stochastic events at a small scale. By using the results from repeated runs they can provide the decision makers with success probabilities for particular disease control strategies. Such models have the disadvantage that they are more complex and development takes longer than for mathematical models. Using both types of approaches to model a single system would combine the strengths of the approaches. Monte-Carlo techniques could be used to represent smaller-scale and mathematical modelling to represent large-scale effects.

Figure 109: Map of bifurcating attractors and underlying structure of chaos for a non-linear birth rate equation



In conclusion, the development of this model has provided valuable insights into the epidemiology of *Mycobacterium bovis* infection in possum populations. The structure of the model allows its mechanisms to be explained and discussed with its potential users without requiring advanced mathematical training. The model provides an example of an integrated approach where an epidemiological field study was used as the basis for model development. The model is unique by incorporating a strong geographical component. This allows it to be used for analysing the epidemiology of bovine tuberculosis infection in possum populations in different geographical locations of the country. Such models however require more resources than mathematical models since their structure is more complex, and this particular model will continue to evolve for some considerable time, taking advantage of new data emerging from current epidemiological studies. The objective of this stage of development was to formulate an initial model structure, not to complete development and validation of a full model.

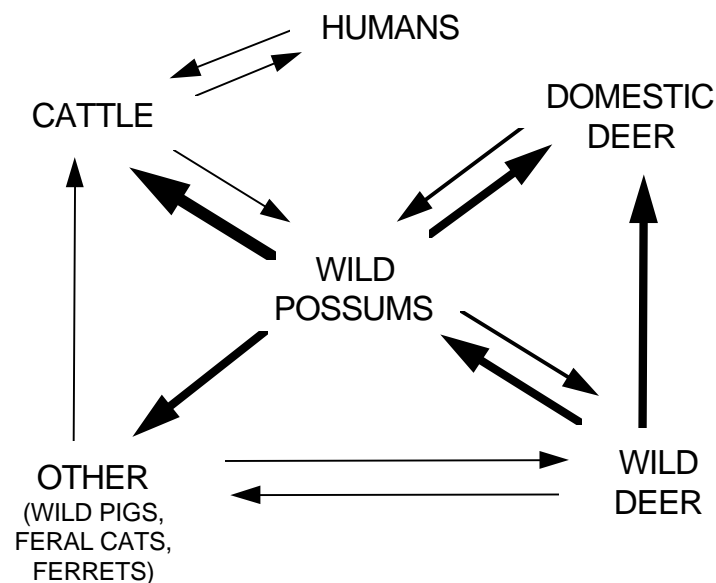
BOVINE TUBERCULOSIS DISEASE CONTROL

The success of disease control strategies depends to a significant extent on an adequate understanding of the epidemiology of the disease. The wider the host spectrum of an infectious organism the more difficult it is to gain sufficient insight into its epidemiology. If the major hosts include wild animal species as maintenance hosts, the problem becomes even more

challenging. In such a situation control of the disease in domestic animals, which is the primary objective of most disease control programs, cannot be achieved without controlling disease in wild animal reservoir species. Most industrialised countries which successfully eradicated diseases in domestic animals did not have densities of wild animals large enough to become a significant reservoir of infection, with limited exceptions such as badgers in the south west of England and Ireland. In many other countries, where there is a potential for wild animals being a significant reservoir of infection, economic and social reasons may have prevented disease control programs from reaching the stage where domestic animals become a spillover host of infection in wild animals, or where this becomes recognized as a residual problem, when direct transmission within the domestic population declines to a very low level. This is what occurred with tuberculosis in New Zealand. In yet other cases, such as the example of rinderpest cited earlier, it is the domestic stock which are the reservoir and the wildlife merely spillover hosts.

The epidemiology of *Mycobacterium bovis* infection in New Zealand is very complex and it now appears that over the last three decades the bovine tuberculosis disease control program has been successful in changing the cattle population from a maintenance host to a spillover host of infection from wild animal reservoir species. Figure 110 shows a systems diagram representing the importance of the different species involved in the epidemiology of bovine tuberculosis in New Zealand and transmission between species. Current understanding strongly suggests that the brushtailed possum is the major reservoir species for the disease in New Zealand. Cattle, feral pigs, feral and wild carnivores can be considered spillover hosts. The importance of domestic and wild deer is not yet clear, and they may either be spillover hosts or low level maintenance hosts. Both feral and domestic deer could conceivably provide a source of infection for possums. This may not be a frequent event, but could be the cause for the development of new, isolated disease foci within the country.

Figure 110: Species involved in the epidemiology of *Mycobacterium bovis* in New Zealand (arrows representing direction and importance of transmission between species)



The evidence suggests that the control of bovine tuberculosis in domestic cattle can be achieved using the standard test-and-slaughter policy based on the tuberculin skin test. For domestic deer the national disease control scheme employs a skin test as well as serological tests to diagnose the disease. Both, domestic cattle and deer are part of an intensive disease surveillance scheme which allows continuous monitoring of the effectiveness of the program. Although there clearly are cases of infected animals which are not detected by the test and legal or illegal movements of infected animals, this is unlikely to be a major cause for continuing occurrence of the disease in some areas of the country. The situation is different for disease control in wild animal populations, especially the brushtailed possum. The standard technique for controlling bovine tuberculosis in possum populations is currently based on population reduction through culling. This method has shown its effectiveness through a reduction of numbers of cattle reacting to the tuberculin test following possum population control operations. However, large scale possum population control operations can only be seen as a short-term measure because they are unlikely to achieve eradication and have to be repeated periodically. Also, there is some evidence which suggests that possum population reduction may not be sufficiently effective in limiting the spread of infection between possum populations or maintaining an insignificant level of disease in the local possum population. Hence, the tools for disease control which are currently available will reduce cattle reactor numbers within areas of endemic bovine tuberculosis infection, but at substantial cost and they are unlikely to fully prevent expansion of these endemic areas. It is therefore necessary to develop new methods of control which are more effective, less costly and also more acceptable to the public and New Zealand's overseas trading partners.

Martin *et al* (1987) provide a list of the activities which can be used in preventing, controlling or eradicating disease. They include slaughter, quarantine, reduction of contact, chemical use, modification of host resistance, environment and/or management control, education and finally biological control. The areas of modification of host resistance and biological control could be applicable for the control of bovine tuberculosis in possums. Both would require a long-term research effort before they could be applied in the field. In the short- to mid-term domestic cattle and deer will have to be protected against transmission of infection from local possum populations by possum population reduction through culling, with the addition of new techniques involving education of farmers leading to management changes on individual farms and possibly to environmental control.

Strategies Available in the Short-Term

There are a number of improvements to the current disease control scheme which can be made immediately. These new strategies can be targeted at changes in regional policies used by disease control authorities and at initiatives to be taken by individual farmers on their property. Regional control policies currently implemented by the Ministry of Agriculture and Fisheries do not sufficiently take account of the epidemiology of bovine tuberculosis infection in possum populations. In the light of the new findings in the different studies of this project it appears that the effectiveness of possum cull operations in reducing possum tuberculosis depends significantly on the timing, the frequency and the geographical selection of target areas for such operations. Given the seasonality of overall mortality, incidence of clinical tuberculosis and of hypothesized major transmission mechanisms (mating and pseudo-vertical transmission), in any given geographical location it could be argued in principle that possum cull operations are likely to be most effective during times when possums are ranging over large areas (as they are more likely to encounter a poison bait) and when their populations are at peak densities. This would be the case during the summer/autumn period. Also it would appear that in terms of bovine tuberculosis epidemiology it would be an advantage to reduce population density during the times of mating when social interaction between animals is at a maximum. The time of March/April would satisfy both criteria. Any control operation conducted during winter (which is currently the case) would have the disadvantage that adult, pregnant females are more likely to avoid poison baits. Also, during winter time there will be more clinically diseased possums in the population and if these animals are late in the disease process they may be less likely to take a poison bait. The issue of whether the carcasses of such animals act as a source of infection for cattle or deer if they die on open pasture is also not fully resolved. In winter, contaminated carcasses will take longer to decompose and bacteria will be able to survive longer than during summer.

There is a lot of scope for initiatives to be taken by farmers. Given sufficient background information on the epidemiology of the disease farmers may well know which are the areas on their property where contact with tuberculous possums is most likely to occur. It should be possible to devise management strategies adapted to the conditions of individual farms which

minimise contact between domestic animals and infected possums populations, and this can be combined with local possum population control operations targeted at the problem locations within the farm. Farmers may be prepared to adjust their domestic stock management and clear vegetation which provides den sites for possums. They could conduct local, very targeted possum kill operations using bait stations which remain permanently in the problem areas and are used during specific times of the year in consultation with epidemiological advisers from the Ministry of Agriculture and Fisheries.

Strategies Available in the Medium-Term

In the medium term it may be possible to develop a vaccination strategy for possums, domestic cattle and deer. At the moment the BCG vaccine which has been used extensively in the control of human tuberculosis is being evaluated by a number of research groups for its usefulness to control bovine tuberculosis in any of the above target species. The outcome of such evaluations depends on the degree of protection which is required in these species. In domestic animals the efficacy of the vaccine would have to be considerably higher than in wild animals in order to be epidemiologically more effective than a test-and-slaughter strategy. Typically in cattle herds, reactor numbers are low which is indicative of the status of a spillover host, and a vaccine would have to be extremely effective to prevent this from happening. The test-and-slaughter strategy performs quite adequately in minimising lateral spread within herds. But in possums, tuberculosis prevalence is comparatively high and the objective of a vaccine would be to break the cycle of infection within the population -which may not require a very high protection level. Therefore, for this and other reasons it would seem most appropriate that any vaccine strategy should have the possum population as its main target.

Tuberculosis vaccination of domestic animals would require acceptance in the international community and is likely to produce responses to the tuberculin test in vaccinated but uninfected animals. Vaccination of possums could be used in combination with population reduction methods. Currently efforts are made in selected parts of New Zealand to prevent the spatial spread of infection by placing "buffers" between 3 to 5 km wide of low possum population density around the fringes of tuberculosis endemic areas. A vaccination buffer may be more effective and would not influence immigration of possums.

Large-scale oral vaccination against rabies has had considerable success in preventing the spread of rabies infection in fox populations (Brochier *et al* 1991, Winkler and Bögel 1992). In relation to the large-scale vaccine trial which had been conducted in Belgium (Brochier *et al* 1991), Anderson (1992) describes a number of problems which have to be addressed before this strategy can be widely adopted. First, mass immunization of wildlife is very costly. Second, Anderson raises some questions regarding the interpretation of the trials which have been conducted to evaluate the effects of vaccination. The design of the Belgian trial did not include a comparable control area in which vaccine baits were not distributed. It is known that rabies incidence is typically cyclical in fox populations, with an inter-epidemic period of 3-6 years depending on fox density. If the trial happened to be carried out during a period of low

incidence, the results would be quite misleading. Anderson also points out that the level of vaccination coverage achieved in the Belgian trial was in accord with the level required to block transmission in a fox population of a given density according to epidemiological theory. He concludes that above all the development of a method for disease control not involving slaughter of wildlife would be only too welcome in the conservation-conscious climate of the 1990s.

Strategies Available in the Long-Term

There are a number of control options which may provide the answer to the bovine tuberculosis problem in New Zealand but would require a medium- to long-term research effort. Vaccination could be one of these methods, either using BCG or development of a new and superior vaccine. A major disadvantage of vaccination of possums is that it does not help solving the immense problems these animals are causing to native flora and fauna. Biological population control through introduction of natural enemies would provide a solution to both problems, bovine tuberculosis and environmental damage. The requirements for a suitable biological control agent are that it has to be host specific and it should pose no threat to the native flora and fauna of the country of introduction, particularly to its endangered species (Waage and Greathead 1988). Biological control of rabbits through introduction of the Myxomavirus causing myxomatosis was initially considered successful in Australia, but changes in the virus and other factors have led to a rebuilding of rabbit populations, and myxomatosis is no longer effective as a control method. In New Zealand this method was recently rejected as an option for rabbit control.

Another method for population control which has received a lot of publicity is fertility control. Bomford (1990) conducted a major review on the role of fertility control in wildlife management. She came to the conclusion that currently no method for effective fertility control in wildlife management is available. Bomford provides the following list of research directions which show promise of pay-off in the future: the development of delivery techniques for drugs that control fertility, the development of drugs which cause permanent, humane, non-toxic sterility in both sexes of target wildlife species; investigation into fertility control to prevent recovery growth of populations reduced by other means; and investigation into the development of genetically engineered viruses to spread sterility-inducing agents through pest populations. She suggests that methods which require continuous or repeated oral dosing over extended periods, which involve surgical implantation and which only affect males show little or no promise for the regulation of abundant or widespread wildlife.

In evaluating any of the above methods it will be necessary to assess their effects not just on the individual in terms of its susceptibility to infection or its reproductive capacity. It will be at least as important to test the effect of the technique on the population dynamics of the target wildlife species. Hone (1992) assessed the potential of fertility control on population dynamics using mathematical methods. He came to the conclusion that effectiveness declines if a reduction in fecundity increases survivorship and that fertility control of pests may be most

effective in a pest population previously reduced in abundance. Caughley *et al* (1992) point out that a knowledge of the social structure and mating system of the host species is important before an attempt is made to suppress female fertility.

It appears that a long-term research effort should be directed towards a method of fertility control in possums. There are a number of options which will have to be evaluated. They include the use of chemosterilants causing permanent or temporary sterility in either sex, or alter the fertility of offspring produced. Another method which has been discussed is immuno-contraception.

DIRECTIONS FOR FUTURE RESEARCH

The studies which are part of this project provide an indication for future directions of research. The results of the longitudinal study suggest that more information is required on the behavioural interaction between possums and between possums and domestic animals. Possum tuberculosis epidemiology can be divided in two main research areas, the dynamics of infection within a local population and the spatial dynamics of infection between local populations.

The longitudinal study has focused on the epidemiology of bovine tuberculosis in population with endemic infection. The results of the study provide an overview of the epidemiology and suggest a number of areas which require further research. More detailed knowledge on the social interaction between possums during the mating period is required. Some of the questions which would have to be answered include how long do male and female mating partners stay together, do they form a consort relationship and is there significant aggressive interaction between males during mating time. Currently no information about interaction in relation to den sites and surrounding areas is available.

The cross-sectional study provided a crude impression of the dynamics of *Mycobacterium bovis* on a larger scale. It did only provide limited data on the spatial dynamics of infection between local populations. The influence of habitat on a larger scale on infection dynamics will have to be investigated. The importance of patch dynamics on the epidemiology of bovine tuberculosis in possums should be assessed. Rather than looking at the effect of individual local habitat patches, their locational relationship within a mosaic of habitat patches - their sizes, shapes, arrangement and connectedness - may be an important factor in the spatial dynamics of *Mycobacterium bovis* infection. In the case of rabies in fox populations it has been found that the heterogeneity of patchiness influenced the persistence of rabies (Tinline 1988).

The modelling approach provides a powerful tool for the longer term both for optimising use of existing control methods, and for evaluating the expected benefits of new ones. Some of the control options outlined above require careful evaluation within the model, to determine whether they perform as well in the model where feedback loops and other interactions can be represented, as they might be expected to do from general principles.

SYNTHESIS

This project provides an example for an epidemiological approach to studying the epidemiology of an infectious disease which is endemic in a wildlife population. Data collection and analysis techniques which are used by animal ecologists have been applied for both, the cross-sectional and longitudinal field study. The complexity of the data which had to be analysed required the use of a wide range of statistical analysis techniques including survival analysis, path analysis and time-series analysis. The spatial aspects of disease occurrence were investigated using geographical information systems and a range of statistical techniques for spatial analysis.

The results from this study provide a basis for further investigations into the epidemiology of *Mycobacterium bovis* in New Zealand. The study concentrated mainly on the dynamics of tuberculosis infection within feral populations of Australian brushtail possums. Current understanding suggests that this species is of principal importance in the epidemiology of *Mycobacterium bovis* in New Zealand. Further investigations will be required to determine the importance of other wildlife species such as deer, pigs and small carnivores, and to evaluate the expected benefits of potential new control options both in the simulation model and in the field.