Chapter 9: Visions for future research in wildlife epidemiology[1]

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"There are epidemics of opinion as well as of disease" Sir B. Brodie 1856

In recent years modern parasite ecology has experienced an epidemic of ideas. We now examine where we think the focus of work will concentrate over the next five years

9.1 Introduction

Prediction is a notoriously difficult task in any field of biology. The editors of this volume decided wisely to avoid this task, instead appointing as seers the doughty individuals who write below. Their prognostications are diverse, but share two common characteristics.

The first is a call for more *integration*, both conceptually (§ 9.4) and across disciplines (§ 9.2). In particular, there is a clear message that the field should seek cross discipline collaboration between field, theoretical and laboratory-based epidemiologists and specialists in other fields (mathematicians, human geneticists, immunologists, etc). This is true for both fundamental research problems (for example, the measurement of immunocompetence; § 9.3) and the applied questions (§ 9.6). In particular, in the modelling section the authors stress the need for a 'meeting of minds' between empiricists and theoreticians (§ 9.2, see also § 9.6.2). The former often needs a better appreciation of the power and – especially – the limitations of theoretical approaches. The latter frequently need to understand more completely the importance of empirical data and expertise, the difficulties of collecting data, and the key skill of balancing mathematical elegance and tractability with biological realism and relevance.

The second – though not always explicit – characteristic of the following sections is how much progress there *has* been in many areas since the 1993 Isaac Newton meeting and the book that arose from that meeting (Grenfell and Dobson 1995). Inevitably, given the uncertainties of research, this is truer for some areas than others. For example there is currently much theoretical interest in modelling complex pathogen strain dynamics, as well as fitting epidemiological models to data, and in understanding the stochastic persistence of pathogen metapopulations. As emphasized in the modelling section, the most fruitful ground for theoretical developments is frequently provided by the coevolution of models and data, as witnessed by the theoretical developments that have built on the current explosion of immunoepidemiological data (§ 9.3 and § 9.5). A particular focus for future immunoepidemiological work should be the collection of detailed spatio-temporal data for infections more immunologically complex than measles. This is currently in progress for many human and wildlife infections (Mills *et al*. 1999). However, such studies need to be part of a long-term research investment, since many important epidemiological questions can only be addressed with data over several decades (Grenfell and Dobson, 1995).

In a particularly clear vision of evolutionary aspects of the subject (§ 9.4), the authors begin in a more pessimistic vein, proposing that none of the questions that Read *et al*. (1995) raised in Grenfell and Dobson

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(1995) have been answered. However, as the section proceeds, their enthusiasm gets the better of them and they anticipate a time when parasite ecology might have as complete a degree of conceptual unification as behavioural ecology. This is arguably too negative a perspective. As the authors note, many problems in host-parasite evolutionary biology arise at the interface of evolution, population genetics and population dynamics and a relatively simple evolutionary 'toolkit' is not available to solve most of these problems.

However, the corollary here is that host-parasite interactions provide unique perspectives on a central question in biology – how do the ecological and evolutionary time scales interact? Again, collaboration across disciplines is of great importance in tackling this major problem. The Trento meeting provided the ideal setting to initiate, and in some cases rekindle, such collaborations.

9.2 Perspectives on modelling epidemiological systems

Since the seminal works of Anderson and May (1991) there has been a significant body of work applying mathematical models to study wildlife epidemiological systems. Progress has been achieved both in the areas of model construction and model validation. The theoretical interest in wildlife epidemiology is reflected in the fact that that over 300 papers have been published in these areas over the last five years. There are two broad areas where further advances are both urgently required and likely to be biologically exciting: that of incorporating stochastic variation (§ 5.3.3) and in a move towards complexity (§ 5.3.4).

9.2.1 Statistical developments

There are two main areas of challenge for numerical epidemiology and ecology over the next five years and these are united by their common requirement of ecologically realistic stochastic models. The first challenge arises from the realisation that real parasite populations are governed by a combination of deterministic and stochastic forces. The key regulatory, density dependent deterministic non-linear processes include such processes as mass action transmission (Grenfell and Dobson 1995, § 5.3), competition/interference (Rohani *et al.* 1998) and parasite induced host mortality. At the same time, important stochastic forces arise from birth and death processes (Bartlett 1956), environmental stochasticity (Rand and Wilson 1991) and spatial or temporal heterogeneities. This duality is now widely recognized across ecology (Turchin 1995), yet elucidating the relative influence of the deterministic versus the stochastic forces has become a major challenge. Finkenstädt and Grenfell's (2000) study of pre-vaccination dynamics of measles in England and Wales suggest that the large amplitude fluctuations in numbers of infected people in big cities may be astonishingly *non-stochastic* – many apparent irregularities are due to birth-rate variations. The challenge of understanding the noise-to-signal issue is heightened by the intricate interaction between non-linear regulatory processes and demographic (e.g. Wilson and Hassell 1997) or environmental stochasticity (Rand and Wilson 1991). Rand and Wilson (1991), for instance, demonstrated that the deterministically stable annual dynamics from an SEIR model (§ 5.2) could interact with stochastic forcing to produce chaotic stochasticity (also called chaotic transience). Drepper *et al*. (1994) suggested that such complex transience might also govern the dynamics of measles in New York. These and related phenomena, that arise from the interaction between regulatory and stochastic forces, are becoming the focus of a range of ecological investigations (McCann and Yodiz 1994, Cushing *et al.* 1998, Higgins *et al*. 1997). Understanding the dynamics of real populations, which are invariably affected by process-inherent stochasticity, will provide an exciting and rewarding playground for ecological theoreticians. As an example, recent studies in the statistical and physical literature indicates that *even* the classical (in a non-linear dynamics sense) notions of chaos and sensitivity-to-initial-conditions may require separate interpretation and methodology once dynamics are stochastic (Paladin *et al*. 1995, Yao and Tong 1995, Loreto *et al*. 1996, Tong 1996).

A second area that is currently exciting both for statisticians and empiricists is the tightening link between models and data. With recent developments in statistics, and in particular computer intensive resampling procedures (Efron and Tibshirani 1993, Gilks *et al*. 1996) the tools are now available to fit complex mechanistic models to data. The last few years have seen some exciting efforts to bridge the gap between theory and data. Begon *et al*. (1998) have used time series data to shed light on basic assumptions of mixing and mass-action (§ 5.3). Similarly, Ellner *et al*. (1998) and Finkenstädt and Grenfell (2000) have developed statistical methods to directly link models for microparasite transmission dynamics to mechanistic (or semi-mechanistic) models. These efforts parallel recent work in other areas of population ecology (see, for example, Dennis *et al*. 1995, Costantino *et al*. 1997, Kendall *et al*. 1999). However, the quality of data, and of the biological insight, which underlies its collection, is still the key to successful

developments in this area. Carefully designed experimental epidemiology and the need to grasp the nettle and undertake brave experiments at the right scale is a particular priority here (e.g. Hudson *et al*.1998, §3.5).

A particularly interesting avenue of research has been the development of discrete state-space models (i.e. modelling population size rather than population density) that now start to address some of the challenges laid down by Bartlett (1956). The advent of Markov Chain Monte Carlo methods naturally lends themselves to statistical estimation. They have the dual role that they can be seen as theoretical models for real populations (having a finite number of individuals, affected by demographic and environmental stochasticity) and at the same time as statistical models for abundance data. Abundance models (as a complement to density models) bear a promise of aiding our understanding of the dynamics of small populations with respect to extinction, critical community sizes and recurrent epidemics (§ 5.2, § 5.4, §8.3).

A key decisions arising in the model construction process itself is whether the formulations have to be stochastic to capture the essential features of the ecological system or whether the statistical aspects can be approximated by deterministic (Holt and Pickering 1985) or a pseudo-stochastic formalism (Anderson and May 1978). Moment closure methods offer a promising way forward here. Nevertheless, even these become very complex when we start to consider the full heterogeneities of real parasite population dynamics, especially in the macroparasites (Grenfell *et al.* 1995, Smith 1999 and Chapter 2). Keeping in mind that deterministic formulations can be obtained by taking the expectation (or the skeleton; *senu* Tong 1990) of stochastic models, we expect an exciting and pluralistic research programme over the next few years that will focus on the mechanistic link between classical (deterministic) theory, stochastic models and the sound data.

9.2.2 Complexity in multi-strain and multi-host systems

The dynamics of systems with many host species and/or multiple pathogen strains has probably been the most exciting area for development over the last 5 years.

Multiple pathogen strain interactions is currently a major area of focus, both empirically (§ 9.3, § 9.5) and theoretically. For microparasites, the main problem has been the curse of dimensionality – as we increase the number of strains considered (n), the maximum number of uninfected compartments we have to consider goes up by 2^n (Andreasen *et al.* 1997, Gupta *et al.* 1996). There is currently a major effort to address this problem - particularly for malaria and influenza, though the implications for wildlife infections are also profound. The main strategy involves efforts to decrease the dimensionality of the problem, by coevolving mathematical tractability and biological realism in terms of how we 'label' the immunological or susceptibility status of individual hosts (Andreasen *et al.* 1997, Ferguson *et al.* 1999, Gupta *et al.* 1996, Gupta 1996).

There are equally interesting problems for interaction of macroparasite strains and species (Chapter 4), with the added complexity of modelling the stochastic dynamics of parasite intensity (Grenfell 1995, Smith 1999).

Multiple host species. The other side of the problem of multiple strains is the structuring of the parasites and strains between host species. Much effort has been focussed on models of apparent competition, mediated by parasites (Holt and Pickering 1985, Begon and Bowers 1995, Hudson and Greenman 1998). Again, dimensionality is a problem leading to analytical intractability of models. Since it is only in exceptional cases that globally valid analytic solutions are possible, most studies have been restricted to a local analysis about the point equilibria of the model. However with higher dimensional models being developed, algebraic limits are being encountered even for local analysis (Holt and Pickering 1985, Begon and Bowers 1995). This has forced researchers to investigate model structure by exhaustive numeric search.

At the 1993 Isaac Newton Conference Begon and Bowers (1995), set out the difficulties in analysing multi-host, multi-pathogen models, thereby providing a challenge to modellers to develop more powerful tools of analysis that could enable at least a local analysis about the system equilibria to be completed. There has been some response to this challenge with the further exploitation of bifurcation methods to address some of these difficult problems. Details are provided elsewhere (Hudson and Greenman 1998,

Greenman and Hudson 2000, §3.8). These new techniques also have relevance to stochastic moment equation systems and the reaction-diffusion equation describing spatially heterogeneous systems. Their development raises the hope that there may be other areas of under-developed mathematical theory that could be of use in unravelling the complex behaviour of existing models.

While workers have tended to undertake stability analysis of system equilibria, some important insights can be obtained by examining transient behaviour in the system and in particular the speed of convergence onto the equilibrium state. This is certainly the case when one is looking, for example, at pest control or the effects of a vaccination programme. Ideally one would like to have explicit analytic expressions for the dominant eigenvalue at the equilibrium in question but even if this were available it would not provide information about the system trajectory far from equilibrium. The unavailability of tractable eigenvalue formulae and the difficulty of analysing non-local behaviour are further challenges to the mathematician to develop more sophisticated analytic tools.

Perhaps we are being too gloomy about the difficulties involved in looking at local and transient behaviour. Even for complex systems one can still derive important results about its subsystems, as evidenced by the work on R_0 threshold conditions (Roberts and Heesterbeek 1995, Heesterbeek and Roberts 1995, Diekmann and Heesterbeek 2000, § 3.2, §5.3). In some cases one can also make reasonable assumptions that simplify the structure sufficiently to allow extraction of analytic threshold conditions (Chapter 4, Dobson 1985). The work of Gupta *et al.* (1994) also shows that multipathogen models can be solved analytically when vector transmission is involved.

In summary, the next 5 years promise to be exciting ones for wildlife disease modelling; in particular, mathematical and statistical analysis are essential tools for understanding the increasing flood of genetic and automatically recorded data on population and spatial heterogeneities in hosts and pathogens. There has been much progress in these areas; however, we all agree that we still seek a meeting of minds between empiricists and theoreticians.

9.3 Challenges for immuno-epidemiology

This book reflects a number of recurring themes, which fall under the broad heading of immuno-epidemiology. In this section we begin by considering the status and future prospects of this area and the section ends with an immunological perspective on the much-discussed issue of measuring immuno-competence.

There are historically two branches of parasite biology concerning the immune response: immuno-epidemiology and parasite immunology. The former was essentially about the role of diagnostic markers of infection and the information that can be gleaned from biological specimens about a host's infection status, i.e. immunology informing epidemiology. Although the term has been coined for macroparasites in particular, it is little different in aims and practice from, for example, using immunological markers to assess the progression of an individual between HIV infection and AIDS. Parasite immunology concerns the role and mechanisms of the immunological response to parasites within individual hosts. This approach has largely ignored epidemiology - experiments to understand the immunology can be made considerably simpler by removing the epidemiology. Consequently, a divide has grown up, with little epidemiological or ecological information being used to inform immunological approaches: immunology being "proper" biology you understand.

It is a major challenge to build and analyse a model starting from an immune response within a host through to an epidemiological description of spread between hosts. There are very few examples of such an analysis, but those that are available show that having both aspects in a model can lead to substantial changes in the dynamics compared to a model disregarding immune response (e.g. Roberts and Heesterbeek, 1998). The situation allies most strongly to interactions in which immunity is not lifelong and absolute (as in measles) but requires ‘boosting’ by subsequent infection (as for many helminth parasites). The point is that the infectious output of an infected individual is a result of the battle with the immune system, but this output determines how much infection gets back into other hosts and with that also the level of challenge of the immune system. This presents not only a modelling challenge, but also a challenge in getting the right tools to analyse these models and, equally importantly, in being able to carry out the right sort of experiments. This is certainly an area where experimenters and theoreticians should collaborate

on animal infections, since there, in contrast to human epidemiology, the necessary experiments could actually be carried out.

To some extent, both epidemiology and immunology are given impetus by having to explain infections that are not as simple as measles. This infection is characterized by a short infectious period during which an effective immunological response develops and clears the infection (if the host does not die). Antibody markers of current infection are well known and form the basis for diagnostics and those for past infection remain for life. In the case of infections that persist, for example hepatitis-B virus, or those in which reinfection is common, for example rotavirus and macroparasites, the basic questions related to the measles paradigm are what are the mechanisms permitting persistence or reinfection (parasite immunology) and what markers of infection can be used to determine the recent or current state of the host (immuno-epidemiology)?

A criticism of the parasite immunologist's approach is that more often than not, the immunological response is studied without the context within which infection normally occurs. For example, single challenge infections are still commonly used rather than continual or trickle exposure. Hosts are raised and kept in environments of varying degrees of sterility although usually hosts are subject to frequent challenges by a variety of parasites. Hosts are frequently super-optimally nourished although all but a minority of human and domesticated hosts are sub-optimally nourished. Hosts are usually born to mothers that kept in the same clean, ideal environment although hosts are usually born to mothers with the same conditions as above. Social conditions with experiments usually restrict interactions between individuals despite the frequently observed relationships between stress, social status and infection/disease. The list could continue.

This criticism of parasite immunology is built on the ecological assumption that the immune response has developed and performs within a context, and that this context is ignored more often than not. Ecology has to inform immunology. Thus, for example, parasite immunologists might regard pregnancy as an abnormal state, whereas, for the great majority of host species (all if we ignore humans in post-war established market economies), sexually reproductive females spend the majority of their lives pregnant. Any results obtained from never pregnant, sexually mature female hosts must be of questionable relevance.

9.3.1 Questions in immuno-epidemiology

A persisting theme of this book is the importance of multiple-host and multiple-parasite systems, both systems where one parasite can infect many different hosts and those where one host is infected with many different parasites. Within the broad field of immuno-epidemiology the second set of host-parasite systems shines out as a beacon marking interesting questions where new theoretical models and new methodologies will allow significant progress. By investigating the rules determining the susceptibility and infectiousness of multiply-infected hosts, immuno-epidemiology may be able to create an important bridge between within-host and between-host parasite population dynamics (Gupta *et al.* 1996). Immuno-epidemiology should ideally encompass both immunology and epidemiology – what are the consequences of each to the other? What needs to be considered – immunological interactions?

What types of interactions take place between parasites sharing a single host? Competition for resources, apparent competition to escape from immune responses (innate and specific), genetic changes through mutation, recombination and sex. The list is long, but already a number of such interactions are well documented - sometimes well quantified. So far most of what has been done concerns monotypic infections or mixed infections with closely related parasites - for example competition between different strains of HIV for CD4 cells (McLean and Nowak 1992), and between different strains of malaria for erythrocytes (Gravenor *et al.* 1995). But some work concerns interactions between unrelated pathogens - for example the impact of live influenza vaccine upon the within-host dynamics of an established HIV virus population (Staprans *et al.* 1995). Generally, of course, hosts provide an environment for parasites within which the whole range of ecological interactions might be expected – from competition to symbiosis (White *et al.* 1998).

Buried within the question of apparent competition between parasites to avoid shared immune responses is the huge question of constraints upon the immune response. Immune responses are tightly regulated, but what is the impact of that regulation upon the outcome of the host-parasite interaction?

What should we consider - ecological interactions? Once it is accepted that immunological responses have developed and evolved to increase the overall fitness of the host, a whole variety of questions follow. Perhaps the biggest consequence is that immunity becomes just another factor to be traded off against processes such as growth and reproduction (Box 2.8). It has been recognized for some time that the neuroendocrine system regulates immunity and that there are complex relationships with, for example, circadian rhythms.

There is increasing ecological evidence that immunocompetence is regulated by sex hormones such as testosterone and often-clear gender differences in morbidity and mortality rates, with males usually doing worse than females (Boxes 2.7, 2.6 and 2.9). Is this a result of male mate competition indicating that growth is a better fitness investment for males than females resulting in reduced investment in immunity? Is the apparent reduction in immunocompetence of pregnant females a consequence of investment of resources in offspring rather than immunity? Is there an ecological explanation here somewhere for the greater incidence of autoimmune disease in women than men? Do individual hosts vary the proportion of investment in immunity by age (and reproductive status)? Is there a trade off between reproductive potential and immunity? Many of these questions might be addressed by careful comparative studies and development of life-history models that include infectious disease explicitly as a morbidity/mortality cause.

In multiple parasite infections of a single host (the norm), do hosts differentiate between parasites in determining the level of immune response to each? For example, it would make ecological sense to react quickly to a multiplying virus that would kill if uncontrolled, which in the context of constraints may imply reduction of immunity directed against parasites that are less dangerous. This would imply mechanisms (within the immune system) that are capable of detecting changes in abundance and spatial distribution of parasites within the host, and determining morbid consequences of each parasite population.

One of the continually reoccurring questions is how long lived hosts can survive in a world of rapidly evolving pathogens. Part of the explanation must lie in the ability of immune system to rapidly respond to new antigens, providing a mechanism that changes on a time-scale equivalent to that of the microparasites. However, there should also be a role for the dissemination of information from mother to offspring. It has been recognized for a long time that offspring derive protective immunoglobulins (either trans-placental or trans-mammary in all mammal species) against viral infections. Could not other mechanisms exist to pass information about the pathogenicity of other infections? The neonate receives "immunological wisdom" from its mother is in a much better position to juggle resource allocation between growth and immunity than one that is ignorant.

9.3.2 Estimating an immuno-epidemiological response

Molecular biology provides us with tools to count numbers of parasites and numbers of responding immune cells within infected hosts. In a number of cases such tools have been used to measure not only number of parasites but also their rate of change under specific interventions. In this way it is becoming possible to define *in vivo* demographic rates for parasites and responding cells (Haase 1999). The subject is in its infancy and so far most progress has been made in the field of understanding the pathogenesis of HIV. However the methods used are of wide application and presage the long-overdue return of an ecological approach to immunology and microbiology (Anderson 1998).

Can we measure immuno-competence? As part of an ongoing effort to explain the benefits of brightly coloured plumage in some birds, behavioural ecologists have hypothesized that such ornamentation may play a role in signalling immunocompetence to potential mates (Saino *et al.* 1997a,b, Sorci *et al.* 1997). A spin-off of these intriguing studies has been that we are forced to address the general question: what is immunocompetence? It is clear that the ability to withstand parasitic insults is an important component of fitness. Even in the apparently clean and ordered world inhabited by humans of the industrialized West, people with severely impaired immunity have greatly shortened expectation of life imposed by their burden of opportunistic infections (Grant and Feinberg 1996). Indeed, the rise of the epidemic of HIV infection has opened up a window into the array of infections that healthy people routinely withstand or suppress. What, then, is immunocompetence? A sensible definition is that it is the component of fitness that can be attributed to having an immune response (Box 2.8). This in turn is the difference between benefits in terms of the ability to withstand parasitic insult and costs in terms of energy expenditure and immune induced pathology.

Instantly, under this definition, one must ask "but what parasitic insult"? Existing studies have recognized the difficulty of this question and have neatly circumvented it by making very broad measures of the magnitude of the immune response: the size of a lesion induced by injecting a non-specific T cell growth stimulator or the protein content of a plasma sample as reflected in its colour (Sorci *et al.* 1997, Norris & Evans 2000). However, none of the measurements used to date have been shown to correlate with the ability to withstand a relevant parasitic insult. Existing studies may not be measuring relevant quantities. There are many technologies available for counting the magnitude of specific responses, some old (e.g. Elisas for measuring circulating specific antibody concentrations) some very new (e.g. tetramer assays for counting specific T cells, Altman *et al.* 1996). How numbers of specific cells relates to the net within host growth rate of parasites (and hence to immune protection) is an important outstanding question in the immunology of infectious disease. The recognition by ecologists of the role of immuno-competence in an individual's fitness once again highlights the need for a greater understanding of this relationship.

If specific rather than non-specific responses are to be used as surrogate for immuno-competence the question "specific for what?" must be addressed. Here there might be an interesting lesson to learn from the experience of the AIDS epidemic. The presence of severely immuno-suppressed people in a community acts as an indicator of the infections circulating in that community and controlled by healthy, immuno-competent people. Opportunistic infections are the parasitic insult that a healthy person's immuno-competence allows them to withstand. Might it not be possible to use immuno-suppressed individuals reintroduced into the relevant sites as indicators of the relevant parasitic insult in a given environment? To be more specific, newly hatched birds would be taken and the relevant organs surgically removed to severely suppress their immune response. When returned to the nest, any infection developing in the immune suppressed chicks would be very strong candidates to use as a challenge to test the immuno-competence of their nest-mates.

9.4. Evolutionary epidemiology - visions of an exciting future

In 1993, the evolution and genetics working group at the Newton meeting was asked to produce a list of promising avenues for research over the subsequent five years (Read *et al.* 1995). Five years later, that list remains an exciting vision of the future (see also Lively and Apanius' chapter in the same volume); so far as we can see, none of the issues have yet been resolved, and there has only been progress on the minority of them. Rather than re-hash the issues there, and rather than simply extending that already long list, we want to discuss a more general vision. We urge readers interested in the case for studying issues such as coevolutionary dynamics, genetics in epidemiology, intra-generation (somatic, immune) versus intergeneration evolution, virulence, resistance and local coadaptation to look at the report from the Newton Institute and to section § 9.5 in this chapter.

A striking feature of host-parasite interactions is overwhelming diversity. Consider for example the enormous variation in life cycles of parasites, of invasion routes and strategies for survival with hosts, and the staggering variation in key life history variables such as age to maturity and daily fecundity even in groups as morphologically similar as nematodes (Skorping *et al.* 1991). Variation in host responses to infection is equally striking. The worm burden that triggers inflammatory responses, for instance, depends on worm species, host species, the organ system involved, and host factors such as age, sex, condition and reproductive status. Much of this sort of diversity is catalogued in often excruciating detail in many textbooks, and further description of this variation is proceeding apace. To us, a critical challenge is to make sense of it all.

Consider how textbook writers guide students through this enormous diversity of natural history facts. Perhaps the most common way to do it is to arrange things taxonomically – indeed phylogeny remains our best way of cataloguing this diversity (Smyth 1994). Others, such as immunology textbooks, are lists of mechanisms, again sometime arranged by parasite taxa (Wakelin 1996). Some arrange things by the problems parasites (or hosts) have to solve, but do little more than list the range of solutions they use (e.g. Matthews 1998). Clearly, these authors are not interested in a general synthesis and for their purposes the details are sufficient in themselves. This does not, however, obviate the general issue: there is no synthesis of this diversity, which would allow predictions about which of these solutions, arises and when. Parenthetically, our best way to predict what might be happening for any given host or parasite is from knowledge of what its phylogenetic relatives do – a phenomenological approach that hardly explains why they do it.

If an aim is to generate principles general enough to be applied to a range of different systems, but also of sufficient realism to provide workable predictions about the real world, classic parasitology has not been very successful. An alternative approach, which seems to us to offer the most exciting possibility is a functional one, aimed at determining how natural selection acts on host and parasite traits, and the constraints on the outcome of selection. This approach has allowed the NeoDarwinian synthesis to successfully make sense of the hugely disparate facts collected by traditional natural historians. The adaptationist programme, particularly in the formal guise of optimality models, has been particularly successful in explaining variation – both qualitative and quantitative – in previously inexplicable phenomena such as the diversity of foraging behaviour and breeding systems of free-living organisms. These sort of cost-benefit analyses ought to be able to explain, for example, differences in virulence or why some parasites exploit two hosts, not one. At least one recent textbook summarizes attempts to date to employ a functional approach (Poulin 1998a). Notably, the textbook is thin but it does point to an exciting future.

Understanding how different selective pressures will affect the evolution of important parasite traits is not only of academic interest, but is urgently needed from a practical point of view. Efforts to control parasites, for example by drugs, inevitably impose new selection pressures. Our current ignorance of the outcome of these large-scaled evolutionary experiments is alarming. Recent papers have suggested that under given conditions drug-use could lead to the evolution of more virulent strains within nematodes (Skorping and Read 1998) and bacteria (Wilkinson 1998). Other outcomes, depending on how the drugs are applied, could be more chronic diseases, changes in parasite habitats or adaptations to new hosts.

Given the success of evolutionary theory in explaining variation in free-living organisms, how would we apply the same line of reasoning to the often considerably more complicated world of host-parasite interactions? Take host specificity as an example. Why can a species like the digenean *Cryptocotyle lingua* use dozens of different host species within widely different orders and even classes, while others, like *Schistosoma haematobium*, are restricted to just one species? Generally, as complexity increases so realism of epidemiological models declines the more host species a parasite can exploit. An extreme generalist that can successfully use a diverse range of hosts must be a true nightmare to the practical epidemiologist. This suggests an obvious advantage of being a generalist: the risk of extinction should decline with the number of hosts. Fortunately, most parasites tend to have a restricted number of potential host species, suggesting that there are costs of exploiting several hosts. But we have few guiding principles, so determining host specificity, requires substantial, and often undirected, field work. Clearly, if we could make realistic assumptions on host specificity based on some general ecological information about the parasite, epidemiology would be easier. It might also be possible to predict when parasites might jump to new host species.

Comparative analysis may be one way to proceed. Using parasites where host specificity has been well studied, and assuming that problems with confounders such as sample size and study effort can be overcome, this method could for example tell us if specificity is related to location within the host, to geographical gradients, to species diversity within host taxa or to life cycle complexity. Such correlative studies could help us discover the broad patterns, but if we want to understand how different selective forces affect the number of hosts a parasite can use, we need to identify any tradeoffs between the costs and benefits of specificity.

In epidemiological terms, the good thing (from the parasite's point of view) of using several hosts is that the effective host population density is increased. This means that the contact rate between transmission stages and hosts should be higher for a generalist than for a specialist, all other things being equal. The cost of using many hosts could be several: locating, infecting, growing and reproducing within one particular host, may require different adaptations than dealing with another host. If the main problem is to grow and reproduce, we may assume that the average fecundity of a generalist is lower than that of a specialist. We should therefore expect a tradeoff between the parasites' contact rate with hosts and fecundity. This will act as a constraint on the number of hosts a parasite should use. Although the shape of the tradeoff curve is likely to vary between species, this framework suggests that the degree of specificity of a parasite depends on the relative importance of fecundity versus contact rates. What kinds of parasites would we expect to sacrifice high fecundity in order to increase contact rates? Monoxenous parasites in hosts of low densities, or parasites in highly fluctuating host populations are possible candidates and we encourage the reader to think of others.

Parasites also show considerable variations in how many sites within the hosts' body they can exploit. For example, *Ascaris*-species can be found both in the liver, in the lungs and in the intestinal system during their development, while *Trichuris spp*. are restricted to the large intestine. Comparative studies may be useful to test hypothesis for such variation (Read and Skorping 1995). An interesting question related to both site and host specificity is the following: what is the easiest for parasites to adapt to - new sites within a host species, or the same kind of sites within new hosts? Phylogenetic analysis, studying radiations of taxa in relations to habitats and hosts may solve this issue.

The study of the evolutionary ecology of free-living organisms has progressed by a combination of theoretical modelling, field observations, elegant experiments that manipulate causal variables, and comparative (cross-species) studies. It seems like that the same approach will bear substantial fruit in the study of host-parasite interactions. In this context we see the principle problems as follows. Many traits of interest lend themselves to relatively straightforward optimality models. For example, the optimal level of host responsiveness to a given challenge is likely to be a simple consequence of the microeconomics of the fitness consequences of responding or not. But frequently the value of these traits in a population will have population dynamic consequences, which will in turn affect the optimal value of the traits. Continuing the example, the optimal level of responsiveness is likely to depend on the probability of reinfection; which will be in part determined by levels of host responsiveness in the population. Optimality modelling for traits with population dynamic consequences is not insurmountable (see van Baalen and Sabelis 1995 in the context of virulence), but how often general answers will emerge is unclear.

Cross-species associations between trait values and ecology have played a key part in evolutionary ecology (Harvey and Pagel 1991); arguably the majority of our knowledge of adaptation is based on comparative studies. Such studies rely on the existence of substantial cross-species variation, which is uncorrelated with phylogeny. This is frequently not the case for major parasite taxa: variation in life cycles, migration routes, intra-host habitats etc. is great within mammalian nematodes, for example, but the variation often lies between higher taxa rather than within them, substantially reducing the ability to statistically separate different factors. Whether this is an artefactual consequence of convergence in life histories and the morphological characters on which the phylogenies are based is unclear; modern molecular techniques may reveal more frequent transitions between character states than are revealed by morphological phylogenies (e.g. Blaxter *et al*. 1998).

The primary aim of both comparative studies and optimality models is to generate hypothesis that must, when possible, be tested experimentally. Experimental studies in evolutionary ecology have frequently involved selection experiments or experimental evolution (e.g. Verhulst *et al.* 1999). These are not always a panacea – they assume the genetic variation found in the laboratory is typical of that available to natural selection. In any case, such experiments will be technically more difficult where at least two species are involved. Another experimental route successfully employed in studies of free-living organisms is to look at trait polymorphism within species. This can involve comparisons of genetically distinct morphs, though arguably more profitable have been studies of adaptive phenotypic responses to environmental variation. For example, some of the best evidence for adaptive sex ratio theory is that parasitoid wasps adjust brood sex ratio under conditions predicted by theory (Herre 1985). In some special circumstances, this may be possible with parasites (eg. Read *et al.* 1995, Gemmill *et al*. 1997), but these may prove to be exceptional. But for many traits of interest – like number of hosts per life cycle – there is frequently no polymorphism within species. Quite why that is so is an extremely interesting question in its own right, but if it is generally true closes off an approach that has proved highly successful elsewhere.

Surmounting (or side-stepping) these difficulties should allow us to explain the diversity of traits such a life cycle, age to maturity, immuno-responsiveness, mechanisms of resistance, virulence, migration routes, intra-host habitats, mechanisms of immune evasion and so on. It would be very exciting if in ten years time it was possible to teach the natural history of host-parasites in the way in which the huge number of previously disparate facts concerning animal behaviour are synthesized by behavioural ecologists. Indeed, the benefits of general principles can be seen in ecological epidemiology (see the rest of this volume), where the same framework can be successfully employed in many different contexts, allowing working scientists (and students) to recognise generalities in the usually overwhelming sea of biological details.

9.5 Challenges from a genetic perspective

There are a number of different approaches that could be taken to consider the genetic inter-relationships between a pathogen and its host. Several of these have already been examined in detail in the previous two sections. In this section we examine a series of thoughts and questions that focus on the context of genetic aspects of wildlife diseases, specifically the evolution of mutation rate, the role of the multiple histocompatibility complex (MHC) and genetic polymorpohisms. Many of these arose from reading the inspiring chapters by Lively and Apanius (1995) and by Read *et al*. (1995) from the Newton Institute meeting (Grenfell and Dobson 1995). The approach taken is one that is intentionally meant to 'adventurous' and 'provocative'. Since I am a relative novice to this field, please ascribe to my ignorance any factual blunders and attempts to make age-old concepts appear novel!

9.5.1 Evolution of mutation rate

Every organism faces a barrage of ever-changing challenges over evolutionary time. On the one hand, pathogens and predators tend to evolve to overcome methods of defence, whilst on the other, hosts and prey evolve to resist attack. Consequently, it is almost a truism to state that any organism, which is unable to adapt, will sooner or later go extinct. Equally, there can be too much novelty. Any organism that mutates too fast runs the risk of causing enough disruption of function to outweigh the benefits brought by adaptability. Somewhere in between these limits exists an optimal rate of change, sufficient to allow adaptation but not enough to cause disruption.

Different organisms are likely to vary greatly in their optimal mutation rates. High mutation rates will be favoured in species that produce large numbers of energetically cheap offspring, each of which has a small probability of surviving and species where rapid adaptation is at a premium. Low mutation rates will be associated with species that invest heavily in small offspring and which live in relatively stable environments. Parasites will tend to fall within the former class, with their constant need to overcome host defences.

Although it is clear that species do vary in their underlying mutation rates, intriguing questions remain to be answered such as: how fast do mutation rates evolve? An optimal system might involve mechanisms for producing mixed offspring, some of which are like their parents whilst others show great diversity. Have such mechanisms evolved? Equally, an optimal genome might be one that is partitioned into regions with different intrinsic mutation rates. To some extent, this has already happened, in the form of, for example, the mammalian MHC genes. However, what about subtler effects, perhaps a general tendency for genes associated with infection and resisting the immune response to lie in regions with higher mutation rates? To study these important effects, there is a need to develop improved technologies for mutation detection. PCR and point mutation detection systems offer some hope. An exciting possibility should effective technology emerge would be, at the risk of some circularity, to turn the question on its head. If critical regions associated with pathogenicity are indeed more variable, methods capable of identifying regions with enhanced mutation pressure will identify candidate genes that are likely to be important in pathogen - host interactions.

9.5.2 How important is the MHC?

It is a widespread belief that high MHC diversity is useful for resisting disease, and hence that species which have lost diversity through, for example, a genetic bottleneck, will be at risk over evolutionary time. However, this argument is called into question by the observation that many diseases are highly species-specific. A pattern composed primarily of one-to-one interactions appears more consistent with hosts being generally well-protected against most infections. Extant diseases in the main probably represent continuations from rare, ancient events, in which a pathogen found and exploited a weakness in new host. Persistence then continued through round after round of an evolutionary arms race in which the host strove to eliminate its weakness and the parasite strove to counter each innovation and so maintain its access. The MHC seems unlikely to play a prominent role in such arms races because most MHC diversity is not inherited but is generated anew each generation. Instead, the MHC may play very much a second line of defence, against pathogens and parasites that have already gained access to the body. Further analysis of long-term studies that show MHC-parasite resistance correlations are essential to teasing out the role of the MHC (Paterson *et al*. 1998).

In this light, an interesting future line of research would be to put more effort into genetic control of parasites. What are the key phenotypic and genetic differences that cause changes in virulence when parasites and pathogens are introduced to naïve populations? What is the relative importance of each component of the infection pathway, from behaviours that influence host-pathogen encounter rates, through nasal biochemistry and architecture, feeding behaviour and preferences, entry into to establishment and persistence within the gut, blood or other tissue? Recent studies on St Kilda sheep indicate the powerful potential links between detection of parasite genes in natural populations and modern, genomically-based animal breeding (Smith *et al*. 1999).

9.5.3 Using genetic polymorphisms as tools.

The more we understand about how different pathogen sequences evolve, the more we can reconstruct historical and current patterns of transmission. For example, modelling transmission rates between alternative hosts in multi-host systems, inferring strain-specific virulences (should they exist) and mapping evolutionary timescales onto the patterns of spread within epidemics (e.g. comparing a phylogenetic tree of morbillivirus isolates from the harbour seal epizootic with spatio-temporal patterns of disease incidence). However, this requires knowing how each marker evolves, and generalisations from mammalian studies may not hold. Examples of actual and potential problems include:

1. Whole classes of vertebrate marker systems which are either absent or not useful in micro-organisms (microsatellites and minisatellites are the most important genetic markers in vertebrate studies but these are rare and often not polymorphic in micro-organisms).

2. Unusual modes of inheritance such as the biparental inheritance of mitochondrial DNA in molluscs (may also be true for some nematodes), micro and macro nuclei in Protozoa and alternation between sexual and asexual forms.

3. Base composition biases. Species vary greatly in their patterns of nucleotide base utilisation and some of these difference may well be functional, for example involved with infecting hosts with different body temperatures or living in guts regions with different pHs. Any shift in ecology could cause a radical change in the rate and nature of base substitutions that might be informative (if recognized) or confusing (if ignored).

4. It is easy to forget that the very large effective population sizes of some pathogens can almost negate the power of neutral genetic drift to cause changes in allele frequencies over the sorts of time-scale we can study. Conversely, while the use of neutral markers may be of limited use in some systems, most differences can be attributed to natural selection.

9.6 The applied and veterinary perspective

Applied problems in wildlife epidemiology generally involve issues that can be broadly categorized as “quality of life” (usually relating to conservation or animal welfare) or ”wealth creation” (perhaps more accurately “wealth threat”). Both sets of issues may be involved in a particular problem.

Quality of life issues will arise if a wildlife disease can cause a dramatic decrease in its host population. Examples of such diseases are the phocine distemper virus, which caused up to 60% mortality in some European seal populations (Harwood 1998, Swinton *et al*. 1998, Box 3.3 and 5.2), and the calicivirus responsible for European Brown Hare Syndrome (Box 5.7). Often the management options, which are available to combat these diseases, have financial implications beyond those of the control operation itself, as may well be the case with Brown Hare Syndrome. If they do, then wealth creation issues will also arise.

Wealth creation issues are created by diseases whose hosts include both wildlife species and domesticated animals. In extreme cases, such as swine fever (Box 5.6), the potential effect of the disease on domestic animals is so great that there is a national or international programme to eradicate the disease. If the proposed management solution to such a problem involves large scale culling of the wildlife host, this will be unacceptable to some sections of society and immediately raises quality of life issues.

We believe that problems, which combine quality of life and wealth creation issues, will become more and more important in wildlife epidemiology over the next decade. The problems faced by scientists who have to provide advice to managers and politicians in these complex situations constitute a major topic in themselves. Here, we consider only the scientific issues involved in investigating them.

9.6.1 Identifying objectives and priorities for management

It is important that all of the stakeholders in a particular problem are brought together at an early stage so that their expectations of what management intervention might achieve can be identified. Appropriate stakeholders include not only those with a technical interest in the problem (wildlife epidemiologists, veterinarians and managers) and those who will be effected economically, but also those who are concerned about the ethical implications of different management options, and those who are involved in the legal and political aspects of the problem.

An essential first step in such discussions is a critical review of the available information on the problem. An agreed and authenticated dataset should be established which can then be used as the basis for further calculations and mathematical modelling. Ideally, a set of objectives for management that is likely to be achievable should be agreed, and priorities assigned to each objective. However, since the different stakeholders will probably have rather different priorities and objectives, this may not be possible. Nevertheless, it should be possible to agree on a core set of objectives and to identify the constraints which the interests of the different parties are likely to impose on other objectives.

9.6.2 Mathematical models and their uses

We expect that most practical problems in the management of wildlife diseases will involve more than a single host and one of its pathogens. Begon and Bowers (1995) recognized that this was likely to be an area of growth in the mathematical modelling of wildlife diseases in the report from the Newton Institute meeting. Their view has been confirmed, not only in wildlife epidemiology but in ecology as a whole. However, they also recognized that extending simple host-pathogen models to more than two species raises formidable problems (Hudson and Greenman 1998). The number of parameters to be estimated grows very rapidly as more species are included in the model, and data to estimate these parameters is usually hard to obtain (see § 9.2.2). In addition, it is often difficult to decide how many species must be included to provide a realistic representation of the situation. The latter difficulty is particularly problematic in other ecological contexts, such as food web analysis (eg Yodzis 1998), but it may be less so in epidemiology, because many systems are relatively well defined.

One solution to this set of problems, which has been used successfully in other contexts such as the management of fisheries under uncertainty, is to concentrate on the analysis of a relatively simple mathematical model of the system but also to examine whether the conclusions drawn from this model are robust if additional complexity is added. This is analogous to the use of spatially implicit models to analyse the effects of spatial heterogeneity in epidemiology. Although such models can be mathematically tractable, their realism needs to be tested using more complex models, in systems where this is possible (Bolker and Grenfell 1996).

It should be appreciated that even the scientific stakeholders may have to make trade-offs in the modelling process. Theoretical ecologists prefer relatively simple models which are mathematically tractable and which can be generalized to apply to a wide range of problems. By contrast, wildlife managers will be most comfortable with models that bear a close resemblance to the specific problem that is being addressed. Clearly a spirit of compromise is needed in these circumstances. However, if mathematical models are to be used for management purposes, then the individual parameters of the model have to be interpretable and estimable. In general, the individual variables need to be measurable, although new statistical tools (§ 9.2.1), permit the estimation of “hidden” (i.e. unobservable) variables.

9.6.3 Implementing models and management

The appropriate trade off between complexity and simplicity is intimately linked to the use appropriate use of the models. Developing an agreed mathematical model can help to improve understanding of a problem, because it forces stakeholders to be specific about the mechanisms, which they believe are responsible for the problem. When these proposed mechanisms are recast into mathematical form it often becomes obvious that some do not, and perhaps cannot, provide a satisfactory explanation of the available observations and these can then be discarded.

However, the main use of models in applied situations is to make predictions and to provide advice for decision-making. Although more complex models usually provide a better explanation of the available data than simpler models, they often have only limited value for prediction and advice, because individual

parameters will be rather poorly estimated (Hilborn and Mangel 1997). As a result, predictions made from these models will have greater uncertainty associated with them than the predictions of relatively simple models. Nevertheless, it is important to test the robustness of the predictions of these simpler models to violations of their basic assumptions. If the addition of complexity to the basic model results in very different predictions, then this should immediately raise doubts about the suitability of the simple model for management purposes. This kind of sensitivity analysis can also be used to assign priorities for future research. If predictions are particularly sensitive to changes in certain parameters in the model, then additional research is clearly required to improve the estimates of these parameters. This research may involve laboratory and field experiments as well as additional data collection.

It is unlikely that all stakeholders will be able to agree on one basic model. However, developing more complex models, which allow the implications of different scenarios to be investigated, can accommodate the views of different parties. It will rarely be possible to estimate all, or even many, of the parameters of these complex models. But feasible ranges of values can usually be agreed, and it is then possible to see what outcomes these might lead to. Again, if certain scenarios result in predicted outcomes, which are very different from those obtained with the simple models, then further research is clearly required.

The modelling process can be considered a success if it helps to ensure that resources are used efficiently both in management and research relating to the problem, and if it help to identify the most effective control or preventative measures. For the latter, some kind of risk evaluation may be most appropriate, so that probabilities can be assigned to the potential outcomes of different management strategies.

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