The Concept of Virulence:
Interpretations and Implications

R. Poulin and C. Combes

The study of parasite virulence, from both a theoretical and an empirical perspective, has blossomed in the past few years[1–4]. The realization that virulence can evolve rapidly, and that it may be influenced by medical practices and social behaviour, has led to its inclusion in classical epidemiological frameworks and to progress in our understanding of the evolution of infectious diseases[5–7].

The concept of virulence, however, is far from perfect. The many definitions, and the ways they are applied, have been influential in shaping our approach to the study of parasite evolution. In particular, the notion of 'genes for virulence' appearing in the recent literature can potentially become instrumental in guiding future research. Because of its importance as a conceptual tool, it is perhaps time to look at the idea of virulence more closely.

What is Virulence?

The word 'virulence' has the same Latin origin as the word 'virus', which originally refered to any pathogen that caused disease. Virulence should therefore refer specifically to a property of the pathogen. According to modern definitions, virulence is the ability of a pathogen to multiply and cause harm to its host[8]. The majority of microbiologists studying parasites appear to use virulence according to this definition. Ecologists and evolutionary biologists sometimes apply a stricter definition of virulence, where harm done to the host takes on a greater importance than parasite fitness itself[8–10]. The issue is not merely one of semantics, however: many of the deeper problems experienced with the notion of virulence may come from this narrower definition.

Operational definitions of virulence used in recent theoretical or empirical studies of parasite evolution have focused on parasite-induced effects on host fitness. In models, virulence is always defined precisely and taken as the main fitness component of the parasite. Empirical studies have generally focused on the variation in damage done to hosts among parasite strains or species, and have also used narrow definitions of virulence. These vary among studies, but usually consist of either mortality rate or lifetime reproductive success of infected hosts relative to uninfected hosts[2,3,11–15], or even more indirect measures, such as the effect of the parasite on the bilateral symmetry of the host[16]. All these definitions are host centred, and are roughly synonymous with the ultimate pathogenic effects of parasites on host fitness. One of the main problems with the concept of virulence comes from these measurements of pathogenicity as surrogates for virulence. Pathogenicity is a property of the host–parasite association[10]. Parasite-induced reductions in survival or reproduction are the product not only of parasite actions, but also of host responses. Many studies of virulence tacitly assume that hosts are monolithic entities, but forget that they also evolve (if at a slower rate than parasites). There is considerable intra- and interspecific variability among hosts in their ability to deal with parasitic infections. The pathogenic effects of a parasite are therefore contingent upon not only which host species, but also which host individual it infects. Effects are also contingent upon the nutritional status of the host or, because of possible synergy among parasites, which other parasite species are also present.

What does the use of these definitions of virulence add to our understanding of parasite evolution? We could obtain substantially different estimates of the virulence of an individual parasite by using different hosts[17], even if the expression of the genes of the parasite within these hosts was the same. Therefore, phenotypic plasticity in parasite exploitation of host resources, as a response to variation in the host environment, is amplified by the use of effects on host fitness as an indirect measure of virulence (environmental conditions and host genes will modulate the pathogenicity of the parasite). Thus, current measures of virulence may be poor reflections of actual parasite actions.

Virulence is often only one of many components of parasite fitness, and it may not be the most significant one. Sometimes, the damage done to the host is incidental and of little concern for parasite fitness. This has been pointed out before[18], but is generally ignored. There is no doubt that the pathogenic effects of parasites on hosts, which are at the core of much recent work on virulence, are important for medicine or wildlife ecology. However, they are perhaps not the best parameters to track in evolutionary studies that are supposedly focused on the parasite itself.

Genes for Virulence?

Current interest lies in how the aggressiveness of the exploitation of host resources by the parasite evolves under certain conditions, and whether medical or veterinary interventions can modify its evolution. Effects on hosts are not part of the strategy of the parasite: there are no genes in the parasite genome specifically coding for a certain level of host damage. One should therefore be careful when speaking of 'genes for virulence', even in mathematical models. The only exceptions are cases where harming the host can directly facilitate parasite transmission, a phenomenon referred to as host manipulation[19,20]. For example, some parasites increase the susceptibility of their intermediate host to predation (therefore increasing host mortality rate) in order to facilitate their own transmission to definitive hosts. In such systems, genes that specifically code for harm to the host can be selected; ensuing reductions in host fitness are adaptive for the parasite and can be viewed as part of the extended phenotype of the parasite. Only these genes can truly be called 'virulence genes' because they have been selected for their immediate virulent effect. In all other systems, there are no specific genes for pathogenicity, and virulence is not truly a part of the parasite phenotype (although it may be a necessary by-product rather than an accident). In summary, virulence can be the specific product of manipulation genes in some cases, or the side-effect of exploitation genes in most other cases. (Ironically, whereas there are no genes for virulence in parasite genomes, there are specific genes for reductions in parasite fitness in host genomes, specifically coding for the destruction of the parasite.)

The idea of genes for virulence in parasite genomes loses even more credibility in the context of gene-for-gene models of pathogen virulence and host

 0169-4758/99/$ – see front matter PII: S0169-4758(99)01554-9

resistance, which have been particularly used in the study of plant diseases[21,22]. Resistance of a plant to a pathogen is only possible when dominant resistance genes are present in the plant and corresponding avirulence genes are present in the pathogen. What this really means is that the pathogen is eliminated when the proteins encoded by avirulence genes are recognized by the immune system of the host. It is then illogical to speak of avirulence genes when the same genes would lead to disease and virulence in hosts not capable of recognizing their products. Pathogens do not express avirulence genes with the intention of triggering defense responses from the host; the primary function of these genes is not clear, but they must confer some advantage to the pathogen instead of coding specifically for its destruction[22,23]. True genes for avirulence would quickly be eliminated from parasite genomes. Avirulence is therefore a profoundly misleading term that, in the context of gene-for-gene evolution, only adds confusion to the issue of virulence evolution.

What Should Virulence Be?

The concept of virulence, however defined, is a property of the host–parasite interaction[10], often treated as a property of the parasite[2,3]. Yet it is measured indirectly through its effects on host fitness: the presumed feature of one organism is measured by its effect on another organism. It seems preferable to reformulate our working definition of virulence as a direct expression of parasite genes in order to study its evolution.

What investigators are seeking is the actual genetically based strategy of a parasite that specifies the rate at which host resources are exploited, how this strategy would fare against other possible strategies in terms of the number of parasite offspring it can produce, and how various agents of selection determine which strategy is currently optimal. Exploitation of host resources has harmful consequences for hosts, but since host genetics and many environmental variables also modulate the expression of pathogenic effects, pathogenicity is a poor substitute for a more direct measure of virulence. Ideally, we would attempt to measure directly the rate at which parasites exploit host tissues: that is, quantify what the parasite actually does rather than its consequences. In most host–parasite systems, this is very difficult. An alternative, also preferable to the study of pathogenicity, would be to combine an investigation of parasite-induced effects on hosts with some measurement of parasite fitness, such as transmission success, rates of asexual multiplication or rates of propagule production (eg. Refs 24,25). If we are to understand parasite evolution more clearly, we need to focus on measures of parasite success in addition to host fitness.

A Shift in Emphasis

The use of the term 'virulence' and the lack of another, better, term for the actual exploitation strategy of the parasite have guided the study and understanding of parasite evolution in one direction, preventing the exploration of others. It will be impossible, and unnecessary, to drop the word virulence and replace it with another; it comes with a lot of baggage and is here to stay. What is necessary, though, is to recognize that: (1) virulence is used as a host-centred measure of a phenomenon that is a feature of neither host nor parasite but of the host–parasite complex; and (2) that it is not a direct expression of parasite (or host) genes and, although it is the result of selective processes, it is not an easily tractable target of selection. The genotype of the parasite is expressed in the actions of the parasite within the host, whereas virulence is a consequence of the phenotype of the parasite. Another informative focus of study would be a parasite-centred measure of how aggressively the parasite exploits host resources, as an expression of a genetic strategy that maximizes the fitness of the parasite.

We do not propose to abandon the term virulence. Instead, we would like to see more emphasis on the forgotten part of the original definition of the word; that is, the ability of the parasite to multiply. Pathogenicity and parasite fitness are not independent, but the former is not a perfect reflection of the latter. Interpretations regarding parasite evolution based strictly on observations of parasite-induced reductions in host fitness are subject to error. It might also be wise to stop talking of virulence (and avirulence) genes in parasite genomes. Investigators of parasite virulence and its evolution should recognize these and other limitations imposed by a host-centred working definition of virulence. Progress will come mainly from good experiments that integrate the various facets of host and parasite fitness, and not just the traditional measurements of virulence.

Acknowledgements
We are grateful to Dieter Ebert, Michael Hochberg and Andrew Read, who provided useful and insightful comments on an earlier version of this essay, despite not agreeing with all of its content.

References
1 Combes, C. (1995) *Interactions Durables,* Masson, Paris
2 Ebert, D. and Herre, E.A. (1996) *Parasitol. Today* 12, 96–101
3 Frank, S.A. (1996) *Q. Rev. Biol.* 71, 37–78
4 Poulin, R. (1998) *Evolutionary Ecology of Parasites,* Chapman & Hall
5 Anderson, R.M. and May, R.M. (1991) *Infectious Diseases of Humans* Oxford University Press
6 Ewald, P.W. (1994) *Evolution of Infectious Disease* Oxford University Press
7 Stearns, S.C., ed. (1999) *Evolution in Health and Disease* Oxford University Press
8 Calow, P., ed. (1998) *The Encyclopedia of Ecology and Environmental Management,* Blackwell Science
9 Read, A.F. (1994) *Trends Microbiol.* 2, 73–76
10 Read, A.F. *et al.* (1999) in *Evolution in Health and Disease* (Stearns, S.C., ed.), pp 205–215, Oxford University Press
11 Toft, C.A. and Karter, A.J. (1990) *Trends Ecol. Evol.* 5, 326–329
12 Herre, E.A. (1993) *Science* 259, 1442–1445
13 Ewald, P.W. (1995) *J. Parasitol.* 81, 659–669
14 Ebert, D. and Mangin, K.L. (1997) *Evolution* 51, 1828–1837
15 Hochberg, M.E. (1998) *Evolution* 52, 1865–1868
16 Agnew, P. and Koella, J.C. (1997) *Proc. R. Soc. London Ser. B* 264, 9–15
17 Jaenike, J. (1996) *Evolution* 50, 2241–2247
18 Ebert, D. and Hamilton, W.D. (1996) *Trends Ecol. Evol.* 11, 79–82
19 Combes, C. (1991) *Am. Nat.* 138, 866–880
20 Poulin, R. (1995) *Int. J. Parasitol.* 25, 1371–1383
21 Hammond-Kosack, K.E. and Jones, J.D.G. (1995) *Can. J. Bot.* 73, S495–S505
22 Van den Ackerveken, G. and Bonas, U. (1997) *Trends Microbiol.* 5, 394–398
23 Bonas, U. and Van den Ackerveken, G. (1997) *Plant J.* 12, 1–7
24 Ebert, D. (1994) *Science* 265, 1084–1086
25 Imhoof, B. and Schmid-Hempel, P. (1998) *Parasitology* 117, 331–336

Robert Poulin *is at the Department of Zoology, University of Otago, PO Box 56, Dunedin, New Zealand. Claude Combes is at the Centre de Biologie et d'Ecologie Tropicale et Méditerranéenne, Laboratoire de Biologie Animale (UMR 5555 CNRS), Université de Perpignan, 66860 Perpignan cedex, France.* ***Tel: +64 3 479 7983, Fax: +64 3 479 7584, e-mail: robert.poulin@stonebow.otago.ac.nz***