

CONCEPTS AND COMMENTS

PHYLOGENETIC APPROACHES TO THE STUDY OF IMMUNOLOGY AND PARASITOLOGY: SOME COMMENTS ON POTENTIAL RESEARCH PROGRAMS

DALTON S. AMORIM and DALMO S. AMORIM*

*Departamento de Biologia, Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto,
Universidade de São Paulo, 14040-901 Ribeirão Preto, SP, Brasil*

**Departamento de Clínica Médica, Faculdade de Medicina de Ribeirão Preto,
Universidade de São Paulo, 14049-900 Ribeirão Preto, SP, Brasil*

True revolutions in areas of basic knowledge are usually followed by technological revolutions in the corresponding applied areas. The emergence of new explanatory theories, in physics, for example, from Ptolemy to Einstein, has resulted in new and more powerful ways of solving practical problems. This imbricated development between basic knowledge and technology is easily observable in the medical sciences. Explanatory models for different kinds of human diseases have probably been proposed since man started to reason. Earlier proposals were based on superstitious beliefs and/or unclear concepts. During the last centuries more and more complex models have become available. At present, the development of medical sciences depends as much on the progress of medicine itself, as on the progress of related areas, such as chemistry, biology, physics, engineering, etc. Altogether, this development of a basic understanding of nature has resulted in an obvious success of the human ability to promote cures.

Comparative biology, in particular, has recently experienced a theoretical revolution which as yet has not reached applied areas. The theory of biological evolution, a model to explain biological diversity, arose in the mid 19th century, structured on the idea of an interrelation between inheritance and modification with time. However, it took nearly a century for a general method of phylogenetic analysis to appear which permitted the precise reconstruction of genealogical relationships among groups of organisms. This method, originally developed by the German entomologist Willi Hennig (1-3), was later refined by others (see 4,5).

Commonsense indeed recognizes that there exists in nature a hierarchy of similarities among organisms. However, it is frequently argued that such similarities have a considerably random distribution among biological groups. One of the most important contributions of Willi Hennig was to show that from the evolutionary point of view there are different kinds of similarities shared by organisms, and that the distribution of

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Correspondence: Dr. Dalton de Souza Amorim, Departamento de Biologia, Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto, USP, Av. Bandeirantes, 3900, 14040-901 Ribeirão Preto, SP, Brasil.

vous cells (as in sleeping sickness), or a variety of cells (as in Chagas' disease) or some other alternative cell type?

- What was the plesiomorphic intermediate host in the ancestral species involving a vertebrate and an invertebrate: a hirundinean, a hemipteram, or a dipteran or some other host?
- Are there common biochemical mechanisms involving the attack against host cells in the group of parasites as a whole?
- What are the features (mainly biochemical) which are exclusively associated with each kind of disease?
- What are the enzymes synapomorphic for the Trypanosomatidae as a whole, i.e., derived proteins shared by all trypanosomatids and only by them?
- What are the proteins characteristic for the Trypanosomatidae and what is their distribution in the family?

This kind of analysis is certainly not a solution for all diseases. Immunological data, and not phylogenetic analysis, provide solutions for diseases caused by viruses, bacteria, protozoans, and multicellular parasites. However, the precise understanding of the phylogenetic relationships among groups of species and of the history of modifications of each biological trait could certainly be an important database for the proposal of reliable models of evolution of the parasite condition in different groups.

The information on the generality of immunological traits may be of extreme help in applied immunological studies. Ideally, it would be preferable to "attack" parasites by fighting traits of broader rather than of more restrict generality in a group, since this eventually could result in the cure of more than one disease or form of a disease at a time. Additionally, this would permit indirect access to information about groups that have not been (or that in some instances hardly could be, such as man) studied for particular traits. Indeed, the method enables us to bring order, the phylogenetic kind of order, to our knowledge about biological features.

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diagrams in which the connection between the stems is given by shared apomorphic traits (i.e., a "phylogenetic tree" constructed under a strict phylogenetic method). One of the more powerful uses of cladograms is to make predictions about the distribution of any biological character in groups of organisms. In practice, the information about particular characteristics is almost always available for only part of the whole set of species belonging to a group. The cladogram permits the use of the available information to make predictions about the distribution of characters in the whole group, including the species which have not been examined directly for those characters. For example, if a biochemical trait (e.g., a response to a certain drug) is observed in species of primates and species of marsupials, it can be inferred that all eutherians (mammals, man included, except Ornitorrincha) will present that same feature (or a condition derived from it). Features shared by man and some birds, on the other hand, can be predicted to occur at least in all amniotes, i.e., all mammals, all birds, and all reptiles. In fact, the determination of the precise level of origin of the whole set of human features - there are supposedly 100,000 genes in the human species - would certainly modify very strongly our knowledge about man - actually, our models of human systems and of their evolutionary development.

A possible application of this method to human problems is the study of monophyletic groups of parasites. The Trypanosomatidae may well illustrate these possibilities. This family of flagellate protozoans includes a number of genera, many of them parasites of plants, invertebrates, and/or vertebrates, including man. Some of the more dramatic tropical diseases are caused by trypanosomatid species such as those of the genera *Trypanosoma* and *Leishmania*.

Many efforts have been made to characterize and treat these diseases. Characterization of species of Trypanosomatidae using molecular biology techniques has advanced considerably (e.g., 6-11). However, the application of the method of phylogenetic analysis to reconstruct the evolution of the family using the enormous amount of information available has been far less common (e.g., 12-14) and much work is still to be done. Such a study would not only recover the relationships among genera, species, subspecies, and "strains", but also indicate the generality of each of the morphological, life cycle, and - more important - immunological features of the group. Examples of questions to be answered using this approach are listed below:

- Did the parasitic condition originate once or more than once in the history of the Trypanosomatidae?
- Did the parasitic life cycle which includes one invertebrate and one vertebrate originate once or more than once in the history of the family?
- Did the parasitism of man originate only once in the history of the Trypanosomatidae?
- Did the original ancestral species of Trypanosomatidae attacking a vertebrate (if this condition appeared only once) affect derm cells (as in leishmaniosis), ner-

similarities is actually not random. The terms *plesiomorphy* and *apomorphy* have been used (3) to designate, respectively, the primitive and the derived states of any pair of homologous features, in which the former was the pre-existing condition which was modified to generate the latter, more recent one in a transformation series. From the point of view of the similarities shared by groups of organisms, we can distinguish among: 1) *symplesiomorphies*, or primitive traits shared by a given set of species and derived from an ancestral species which is ancestral not only to the group of species considered; 2) *synapomorphies*, or derived traits shared by a set of species and not shared by any other species, which must have originated in an ancestral species which is not ancestral to any other species; and 3) *homoplasies*, or traits which are similar to each other, but have derived independently during evolution.

If we are able to distinguish these kinds of similarities among the characteristics of a group, we can reconstruct the genealogical history of such group, i.e., we can determine which species or groups of species have exclusive common ancestrals. Indeed, synapomorphies are precisely the kind of similarity which is evidence of common exclusive ancestry (for more details of the method, see 2-4). The presence of hair, for example, is an apomorphy shared by all mammals (i.e., it is a synapomorphy of mammals), the corresponding plesiomorphy being the presence of derm scales. The presence of an amnion during the development of the embryo is synapomorphic for all amniotes ("reptiles", birds, and mammals). The quadruped condition, on the other hand, is a symplesiomorphy (the human biped condition being the corresponding apomorphy) of the group composed by all mammals except man. The presence of wings is a homoplasy between birds, bats, and pterygote insects. Among these characters, the presence of hair is evidence that mammals are a monophyletic group, i.e., that the species referred to as mammals have an ancestral species which is not ancestral to any other group. The presence of the amnion is evidence of monophyly of the amniotes. The quadruped condition and the presence of "wings", on the other hand, are not at all indications that "all mammals except man" or "birds, bats, and pterygote insects", respectively, have exclusive ancestral species. Actually, very few of the biological traits known have been definitely classified as synapomorphic, symplesiomorphic, or homoplastic conditions.

Phylogenetic analysis arose historically within the context of basic systematics. Phylogenetic analyses have used morphological traits more frequently, probably because morphological traits are easier to access. However, other kinds of traits may and should be used in phylogenetic analyses. Once a reliable cladogram of a group has been reconstructed using a strict method of phylogenetic analysis, the evolution of all traits produced by the genotype, i.e., morphological, physiological, ethological, biochemical, ecological, etc., can be readily understood. The reason is that there is a single history of phylogenetic relationships of the species and all biological features are generated and modified within this system of ancestral-descendant relationships, i.e., the species successively divided in space and time.

There are important consequences of the application of this method to medical research. Phylogenetic knowledge is expressed in cladograms. Cladograms are branching

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