Commentary

Hominoid systematics: The soft evidence

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Phylogenetic analysis (determining evolutionary relationships among organisms) is an absolutely necessary enterprise in developing descriptions and explanations of evolutionary events. Over the past several decades new approaches to analyzing classical morphological data (cladistics) (1), and the increasing availability of newer kinds of data (molecular or genetic) (2, 3), have transformed phylogenetic analysis into a vigorous and sometimes contentious area of evolutionary biology. Arguments among systematists often have been characterized as involving morphology versus molecules (4), but in most cases results from these two kinds of data agree; arguments develop only where there are conflicts. One of the most interesting recent debates has involved relationships among the Hominoidea (humans, chimpanzees, bonobos, gorillas, orangutans, gibbons, and siamangs), one of the more intensively studied groups of mammals. Hominoid relationships have been debated over the past century and more, with strong differences of opinion and interpretation crystallizing most recently around the relationships of the African apes. Are the chimps (including bonobos) more closely related to humans or to gorillas? Molecular data now overwhelmingly support a chimp-human relationship, whereas morphological features are still invoked by many primatologists to support a chimp-gorilla relationship (5). The Gibbs *et al.* (6) paper on hominoid relationships in this issue of PNAS raises several very important issues because it involves an extensive morphological data set, which unequivocally supports, like the genetic data, a chimp-human clade.

Until quite recently most evolutionary relationships were inferred from the morphology of hard tissues (bones and teeth) and soft tissues (including muscles, blood vessels, nerves, integument), by using the so-called cladistic approach developed by Hennig (1). In this approach the phenotype is described as a series of characters, which exist in two or more states. These can be of a presence/absence sort or can form a graded series. Character states are categorized as earlier or more primitive within a lineage (plesiomorphic) and later or more or less derived (apomorphic). Hennig's insight was noting that, in a three-taxon problem, taxa A and B will be more closely related to each other (forming a monophyletic group) than either are to taxon C if A and B share derived character states. The sharing of primitive character states, or of similarities evolved convergently or in parallel, can tell us nothing about relationships. This cladistic approach contrasts with the more traditional phenetic approach in which overall similarity (involving an aggregation of characters) is used to infer relationships.

Until the late 1950s perhaps the most favored hominoid phylogeny linked the large or great apes (chimps, gorillas, orangs) together as a monophyletic group (7), a conclusion based on phenetics or overall similarity. But at that time the first systematic molecular studies of hominoids began. Morris Goodman (8) showed that when genetic distance between species was estimated by the intensity of immunological cross-reactions in a series of serum proteins humans and the African apes (gorillas and chimps) formed a triad of closest relatives, and hence were inferred to be monophyletic, with orangs and gibbons plus siamangs more distantly related. This conclusion became in part widely accepted among morphologists, although it was modified in that humans were linked to a lineage including both chimps and gorillas, supposedly closer to each other, rather than having the three equally related as the genetic evidence had shown (9, 10).

What about strictly morphological approaches to the problem? During the 1980s and 1990s at least six major phylogenetic studies of living hominoids were completed by using dominantly hard and soft tissue morphological data (10–15). They reached five different conclusions as to relationships among the hominoids! And marked disagreements about phylogeny characterized not only analyses of the living hominoids, but also the hominoid fossil records of the (mainly Miocene) apes (16–20) and the (mainly Pliocene) early hominins (21). This embarrassing lack of agreement passed almost unnoticed.

After Goodman's initial work the molecular database expanded enormously. Sarich and colleagues' studies (22, 23) in the 1960s and 1970s confirmed that the relationships of chimps, gorillas, and humans were indeed best then characterized as a trichotomy. By the early 1980s further work from Goodman's group, first with amino acid sequences (24) then with nucleotide sequences (25), began to suggest rather surprisingly a close chimp-human relationship (surprising because of the marked phenotypic similarity of chimps and gorillas). This relationship was confirmed, after some debate, by DNA–DNA hybridization data showing a clear chimp-human relationship (26).

The accumulation of sequence data on a large number of genes, including the entire mitochondrion, has amply confirmed this result. As Ruvolo (27–29) has noted these analyses have several important strengths. They are objective, in that different laboratories will generate the same sequence data from the same genes; they are readily analyzed objectively both cladistically and phenetically; genes on different chromosomes, or distant enough on the same chromosome to segregate independently will provide gene trees that are independent assays of evolutionary relationships (species trees); and well-understood genetic and molecular processes (ancestral polymorphism, gene conversion) exist to explain gene tree conflicts. Ruvolo's analysis (29) of many independent genes demonstrates overwhelmingly that chimps and humans are closest relatives. And hominoids are not the only group in which there are surprising conflicts between morphologically based expectations and more recent genetic data. Among primates a good example is the Papionini, the Old World monkey tribe comprising baboons (*Papio*) and their closest relatives. The dog-faced baboons and mandrills (West African "forest baboons") have long been considered monophyletic relative to shorter-faced species, but genetic data now strongly support a tree in which baboons and mandrills are quite distantly related,

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their long faces having evolved in parallel (30).

Two important recent papers use these robust genetic trees to address issues involving the use of phenotypic data in phylogenetic analyses. For example, Collard and Wood (31) used a set of hard tissue morphological characters frequently used in analyzing early hominin relationships to assess the relationships of both living hominoids and papionins. These characters failed, and failed quite badly, to retrieve the highly probable genetic-based trees. But in marked contrast to this, the analysis of Gibbs *et al.* (6) using a very extensive soft tissue data set (mainly muscles, nerves, blood vessels) unambiguously derives the "molecular" tree in which chimps and humans are closest relatives. The 197 characters used in their analysis were culled from the literature and came from studies in which phylogenetic relationships were generally not a primary goal, often from a time when a specifically chimp-human link was a gleam in no one's eye.

Why did these soft tissue data set give the highly probable hominoid tree whereas other kinds of morphological analyses, particularly those using just hard tissue characters, yielded such discordant, ambiguous, and mostly incorrect results?

Phylogenetic relationships are genetic relationships, and the history of using morphological characters in systematics is one of aspiring to "read" phenotypic data in ways reflecting underlying genetic patterns of relatedness. Ideally, different morphological characters should be independent estimates of relationships, just as sequence data on genes from independently segregating units are. But in practice there are problems with morphological data; problems of ambiguity, arbitrariness, and selectivity, all contributing to making the exercise difficult and problematic. Little attention has been paid until recently to the problems of character definition.

For example, Cartmill (32) showed that different anatomical terminologies applied to the same anatomical region can generate different character lists, and hence different phylogenetic conclusions. In my estimation this problem is much more widespread than generally is recognized and certainly applies to analyses of the Miocene ape fossil record with which I am most familiar. No objective rules appear to govern how any particular region is described (atomized, parsed), how many characters are used to describe an area (these factors apply particularly to hard tissue characters), and how many and which characters are to be used to describe a taxon. Each systematist is free to decide how to characterize an organism (morphologically although not genetically), and there is no way to choose between "my" characters and "your" characters. One recent excellent systematics primer notes: "The experienced systematist- . . . may be in a position to judge which are good characters. . . but this may be difficult to justify to others and implies that the phylogeny is already known" (ref. 33, p. 41).

Sarich (ref. 34, pp. 108–109) characterized a significant part of the problem: "There are always going to be apparently conflicting signals. . . that support the picture you have already decided on intuitively (and which in fact may be correct), even though another worker, possibly equally experienced, can recognize and choose another signal, equally explicit, that supports quite another picture.

". . . there is no objective, unbiased way of randomly choosing and, perhaps even more important, counting characters of anatomy. It is at this point that one has to emphasize that one has to know one's organisms very well indeed to know what characters are phylogenetically relevant and which are not. But how do you choose when, as will almost always be the case, different phylogenies can be supported by different character sets? Parsimony will not do, I am afraid, because that would assume the characters were randomly chosen—or, perhaps, chosen because of their phylogenetic utility. But how do you judge such utility without already having come to some conclusions as to the phylogeny involved? I cannot see how you can, and therefore conclude that one cannot legitimate the statistical testing of various phylogenetic hypotheses derived from anatomical data using cladistic reasoning."

The notion of "independence" for morphological data also presents fundamental problems because of the (long and well-known) phenomena of pleiotropy and multifactoriality; the link between genes and morphology is exceedingly complex. For example, particular genes can be implicated in the development of very different body parts: mouse strains with *hoxa-11*/*hoxd-11* double mutants show phenotypic changes in the forelimb, kidneys, and vas deferens (35). Further, different genes may be involved in the same character; for example, mutations in different *Hox* genes (*hoxa-11*, *hoxd-11*, *hoxd-13*) can produce identical phenotypic effects in mouse distal forelimb elements (36). Hence in analyzing phenotypic features we do not know which part and how much of the genetic base is being assayed, and hence cannot know about independence in the sense that genes in different linkage groups are independent estimators of evolutionary relatedness.

There are, of course, some ameliorating points. Criteria are being proposed to improve selection of morphological characters (37, 38). Among other criteria, units for analysis (characters) should be at the least natural (morphogenetically coherent), independent assays of relationships, heritable, and objective (in the sense that their description should permit no ambiguity). There is growing recognition that developmental processes are important in the definition of appropriate analytical units. As but one example, McCollum (39) recently argued that many facial characters used to assess early hominin relationships are expressions of just a couple of underlying developmental processes; thus phylogenetic analyses based on many facial traits that are likely to be merely reflections of a small number of underlying processes may well give unreliable results. A now classic example of the use of a developmental perspective in systematics is that of Davis (40), who was able to show based on a developmental approach to morphology that giant pandas were related to bears rather than to lesser pandas, a result amply confirmed subsequently with genetic data (41). The rapidly expanding understanding of developmental processes can reduce subjectivity in describing complex morphologies and help us describe better characters (39, 42). But we are still faced with the problem of how to generate not just characters but different character states so that the decisions about what is primitive and what is derived so necessary for cladistic analyses can be made.

Another recent approach to the problem involves so-called total evidence (43). This approach proposes to use all available characters, morphological and genetic. A problem here is comparability; in what sense are morphological and genetic "characters" equivalent? Because any particular morphological character may reflect several to many genes, or one gene affect several to many characters, how are these comparable to a gene? Are genetic characters single genes or single nucleotides? How are these very different kinds of evidence to be combined? Should the 197 soft tissue characters of hominoids (6), plus the 240 mostly hard tissue characters of another recent study (44), be combined with 14 or so independent genes (29) or with the many thousands of bases of sequence represented by those genes? The approach inevitably mixes not just oranges with apples but oranges with apple puree.

In the opinion of a growing number, the ultimate test of the validity of any morphological analysis or of the value of particular characters or kinds of characters is going to be congruence, or lack thereof, with multiply supported, robust, gene-based trees (6, 31). This presents something of a problem for us paleontologists.

So, back to hominoid soft tissues (6). Why do they produce a result compatible with gene-based trees? They are easy to describe relatively objectively. Selectivity is not a problem with this data set. Most of the characters are discrete and "natural" morphogenetic units. Epigenetic effects are minimal and will not affect the number of muscle bellies or the branching patterns or distributional topography of nerves or arteries. Finally, as the authors (6) note, origins and insertions of muscles tend to be conservative; muscle gross morphology and relations are conserved regardless of evolutionary changes in hard tissue. We now need to see how generalizable this result is by testing other groups for which there are robust hypotheses of relationships.

1. Hennig, W. (1966) *Phylogenetic Systematic* (Univ. Illinois Press, Urbana).
2. Goodman, M. & Tashian, R. E., eds. (1976) *Molecular Anthropology* (Plenum, New York).
3. Hillis, D. M., Moritz, C. & Mable, B. K., eds. (1996) *Molecular Systematics* (Sinauer, Sunderland, MA), 2nd Ed.
4. Patterson, C., ed. (1987) *Molecules and Morphology in Evolution: Conflict or Compromise?* (Cambridge Univ. Press, Cambridge, U.K.).
5. Pilbeam, D. (2000) in *The Primate Fossil Record*, ed. Hartwig, W. (Cambridge Univ. Press, Cambridge, U.K.), in press.
6. Gibbs, S., Collard, M. & Wood, B. (2000) *Proc. Natl. Acad. Sci. USA* **97,** 11130–11132.
7. Schultz, A. (1968) in *Perspectives on Human Evolution*, eds. Washburn, S. L. & Jay, P. C. (Holt, Rinehart, & Winston, New York), pp. 122–195.
8. Goodman, M. (1962) *Ann. N.Y. Acad. Sci.* **102,** 219–234.
9. Simpson, G. G. (1963) in *Classification and Human Evolution: Viking Fund Publications in Anthropology no. 37,* ed. Washburn, S. L. (Wenner-Gren Foundation, New York), pp. 1–31.
10. Andrews, P. & Martin, L. (1987) *J. Hum. Evol.* **16,** 101–118.
11. Kluge, A. G. (1983) in *New Interpretations of Ape and Human Ancestry,* eds. Ciochon, R. & Corruccini, R. (Plenum, New York), pp. 151–177.
12. Schwartz, J. H. (1984) *Nature (London)* **308,** 501–505.
13. Groves, C. P. (1986) in *Comparative Primate Biology: Vol. 1 Systematics, Evolution, & Anatomy,* eds. Swindler, D. & Erwin, S. (Liss, New York), pp. 187–217.
14. Hartwig-Scherer, S. (1993) Ph.D. dissertation (University of Zürich, Zürich, Switzerland).
15. Braga, L. (1995) Ph.D. dissertation (University of Bordeaux I, Bordeaux, France).
16. Andrews, P. (1992) *Nature (London)* **360,** 641–646.
17. Begun, D. R. (1994) *Yrbk. Phys. Anthropol.* **37,** 11–64.
18. Cameron, D. W. (1997) *J. Hum. Evol.* **33,** 449–477.
19. Moyà-Solà, S. & Köhler, M. (1995) *J. Hum. Evol.* **29,** 101–139.
20. Schwartz, J. (1990) *J. Hum. Evol.* **19,** 591–605.
21. Corruccini, R. S. (1994) in *Integretive Approaches to the Past: Paleoanthropological Advances in Honor of F. Clark Howell,* eds. Corruccini, R. S. & Ciochon, R. L. (Prentice–Hall, Englewood Cliffs, NJ), pp. 167–183.
22. Sarich, V. M. & Wilson, A. C. (1967) *Science* **158,** 1200–1203.
23. Sarich, V. M. & Cronin, J. E. (1976) in *Molecular Anthropology,* eds. Goodman, M. & Tashian, R. E. (Plenum, New York), pp. 141–170.
24. Goodman, M., Romero-Herrera, A. E., Dene, H., Czelusniak, J. & Tashian, R. E. (1982) in *Macromolecular Sequences in Systematic and Evolutionary Biology,* ed. Goodman, M. (Plenum, New York), pp. 115–191.
25. Goodman, M., Porter, C. A., Czelusniak, J., Page, S. L., Schneider, H., Shoshani, J., Gunnell, G. & Groves, C. P. (1998) *Mol. Phylog. Evol.* **9,** 585–598.
26. Caccone, A. & Powell, J. R. (1989) *Evolution* **43,** 925–942.
27. Ruvolo, M. (1994) *Am. J. Phys. Anthropol.* **94,** 89–114.
28. Ruvolo, M. (1995) *Am. J. Phys. Anthropol.* **98,** 218–232.
29. Ruvolo, M. (1997) *Mol. Biol. Evol.* **14,** 248–265.
30. Disotell, T. (1996) *Evol. Anthropol.* **5,** 18–24.
31. Collard, M. & Wood, B. (2000) *Proc. Natl. Acad. Sci. USA.* **97,** 5003–5006.
32. Cartmill, M. (1982) *Geobios* **6,** 279–287.
33. Smith, A. B. (1994) *Systematics and the Fossil Record* (Blackwell Scientific, Oxford).
34. Sarich, V. M. (1993) in *Mammal Phylogeny: Placentals,* eds. Szalay, F., Novacek, M. & McKenna, M. (Springer, New York), pp. 103–114.
35. Davis, A. P., Witte, D. P., Hsieh-Li, H. M., Potter, S. S. & Capecchi, M. R. (1995) *Nature (London)* **375,** 791–795.
36. Davis, A. P. & Capecchi, M. R. (1994) *Development (Cambridge, U.K.)* **120,** 2187–2198.
37. Atchley, W. R. & Hall, B. K. (1991) *Biol. Rev.* **66,** 101–157.
38. Lieberman, D. E. (1999) *Evol. Anthropol.* **7,** 142–151.
39. McCollum, M. A. (1999) *Science* **284,** 301–305.
40. Davis, D. (1964) *The Giant Panda: A Morphological Study of Evolutionary Mechanisms. Fieldiana: Zoology Memoirs Vol. 3* (Chicago Natural History Museum, Chicago).
41. O'Brien, S. J., Nash, W. G., Wildt, D. E., Bush, M. E. & Benveniste, R. E. (1985) *Nature (London)* **317,** 140–144.
42. Lovejoy, C. O., Cohn, M. J. & White, T. D. (1999) *Proc. Natl. Acad. Sci. USA* **96,** 13247–13252.
43. Kluge, A. (1989) *Syst. Zool.* **38,** 7–25.
44. Begun, D. R., Ward, C. V. & Rose, M. D. (1997) in *Function, Phylogeny, and Fossils: Miocene Hominoid Evolution and Adaptations,* eds. Begun, D. R., Ward, C. V. & Rose, M. D. (Plenum, New York), pp. 389–416.