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DNA Hybridization Evidence of Hominoid Phylogeny: Results from an Expanded Data Set

Charles G. Sibley[1] and Jon E. Ahlquist[2]

[1] Tiburon Center, San Francisco State University, Box 855, Tiburon, California 94920, USA
[2] Department of Zoological and Biomedical Sciences, Ohio University, Athens, Ohio 45701, USA

Summary. The living hominoids are human, the two species of chimpanzees, gorilla, orangutan, and nine species of gibbons. The cercopithecoids (Old World monkeys) are the sister group of the hominoids. A consensus about the phylogeny of the hominoids has been reached for the branching order of the gibbons (earliest) and the orangutan (next earliest), but the branching order among gorilla, chimpanzees, and human remains in contention. In 1984 we presented DNA–DNA hybridization data, based on 183 DNA hybrids, that we interpreted as evidence that the branching order, from oldest to most recent, was gibbons, orangutan, gorilla, chimpanzees, and human. In the present paper we report on an expanded data set totaling 514 DNA hybrids, which supports the branching order given above. The ranges for the datings of divergence nodes are Old World monkeys, 25–34 million years (Myr) ago; gibbons, 16.4–23 Myr ago; orangutan, 12.2–17 Myr ago; gorilla, 7.7–11 Myr ago; chimpanzees–human, 5.5–7.7 Myr ago. The possible effects of differences in age at first breeding are discussed, and some speculations about average genomic rates of evolution are presented.

Key words: Hominoid phylogeny — DNA–DNA hybridization — Statistical tests — Human evolution — Generation time — Average rates of genomic evolution

Introduction

The living members of the Hominoidea are human (*Homo sapiens*), the common chimpanzee (*Pan troglodytes*), the pygmy chimpanzee or bonobo (*Pan paniscus*), gorilla (*Gorilla gorilla*), orangutan (*Pongo pygmaeus*), and the nine species of gibbons (*Hylobates*). The sister group of the hominoids is the Cercopithecoidea (Old World monkeys, macaques, baboons, colobines, etc.). The composition of these groups is not in dispute, but the branching sequence of some of the hominoid lineages and the datings of the divergences are still being debated.

In recent years at least five branching sequences have been proposed for the hominoid genera. All authors have accepted the gibbons as the living descendants of the oldest hominoid branch; the other genera are the descendants of subsequent branchings of the sister clade of the gibbons. Thus, the debates concern the branching sequence among orangutan, gorilla, chimpanzees, and human.

Primarily from morphological evidence, Kluge (1983) proposed that the orangutan lineage is the sister group of the African apes, and Schwartz (1984a,b) suggested that the orangutan and human lineages form a clade that is the sister group of the African apes. However, the consensus from both morphology and molecular evidence is that the orangutan lineage branched next after the gibbon clade, and that the gorilla, chimpanzee, and human lineages form a clade. Therefore, we will consider only the following four branching patterns in which the taxa are listed in the order of branching from oldest to youngest, as in Fig. 1.

1) Gorilla–chimpanzee–human (Fig. 1-1);
2) Human–chimpanzee–gorilla (Fig. 1-2);
3) Chimpanzee–gorilla–human (Fig. 1-3);
4) A trichotomy, from which the three lineages branched simultaneously (Fig. 1-4).

A true trichotomy is unlikely to occur, thus it is

Offprint requests to: C.G. Sibley

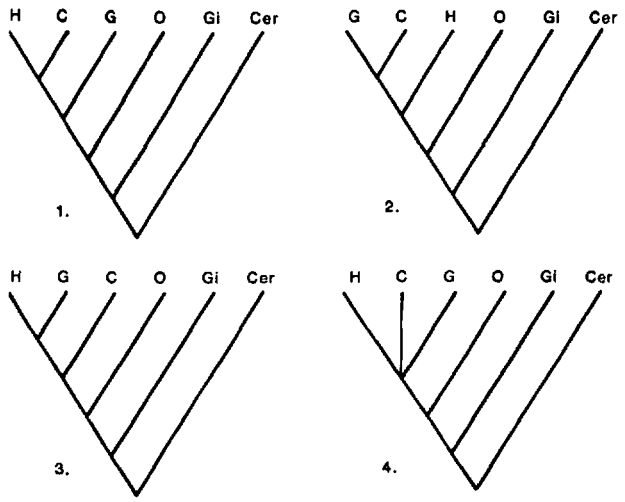

Fig. 1. Four possible hominoid phylogenies. H = human; C = chimpanzees; G = gorilla; O = orangutan; Gi = gibbons; Cer = cercopithecoids.

probable that when a trichotomy is found there are actually two branches so close together that they are not being resolved. Several molecular methods have produced a trichotomy, including comparisons of 2251 bases of the eta locus, one of the five beta-related globin genes (Koop et al. 1986a).

A chimpanzee–gorilla–human sequence (Fig. 1-3) has received little support and is not indicated by any of the molecular data. We will not discuss it further. The principal debate is between the proponents of branching sequences 1 and 2 in Fig. 1. Most morphological studies favor an African ape clade (chimpanzee–gorilla) (Fig. 1-2) as the sister group of the human lineage, but many molecular studies favor a chimpanzee–human clade as the sister group of the gorilla lineage (Fig. 1-1).

Sibley and Ahlquist (1984) presented the results of a DNA–DNA hybridization study of these taxa based on a complete matrix of comparisons. The principal conclusion was that the sequence of branchings was as in Fig. 1-1.

Here we present the results of a larger set of DNA–DNA hybridization values. The 1984 paper was based on 183 delta $T_{50}H$ values; the present set includes the earlier values plus an additional 331, for a total of 514.

Reviews of Some Recent Publications

Ciochon (1985) reviewed 43 classical papers "covering the entire scope of primate and human evolution with particular emphasis on the fossil record." He made a cladistic analysis of 32 "selected" morphological characters of living and fossil hominoids, and concluded that the result "supported by a plurality of contributors" is that depicted in Fig. 1-2.

Gingerich (1984, 1985, 1986) examined the problem of using fossils to date divergences that are then used to calibrate the rate of molecular evolution along divergent lineages. For the primates, Gingerich (1984, p. 67) noted three divergence times that "are sufficiently well established on the basis of fossils to be of importance":

1) Prosimii–Anthropoidea, ca. 55 million years (Myr) ago;
2) Platyrrhini–Catarrhini, ca. 40 Myr ago;
3) Cercopithecoidea–Hominoidea, ca. 25 Myr ago.

Gingerich believes it unlikely that rates of molecular evolution will be linear with absolute time and that they should be expected to be nonlinear. Therefore, in calculating rates the calibration should be scaled against "a minimum of four divergence times . . . for adequate statistical power in testing the linear model" (1986, p. 205). This is clearly desirable, but accurate divergence dates based on fossils are all too rare.

Gingerich (1986, p. 215) calculated the scaling coefficients for several sets of distance data on primates. A coefficient of 1.0 indicates a linear relationship between time and molecular change. For DNA hybridization data the scaling coefficients ranged from 1.08 to 1.75; for immunological distances, 1.42 to 1.54; for amino acid sequences (myoglobin), 1.12 to 1.37; and for nucleotide sequences, 1.83 to 2.61.

It seems likely that the average rate of genomic evolution is affected by generation time, thus it is possible that some of the nonlinearity is due to changes in generation length over the time a lineage has been diverging from its related lineages. Gingerich (1986, p. 219) concludes that "To the extent that amino acid replacements and nucleotide substitutions change in proportion to elapsed evolutionary time . . . this change can be attributed to neutral evolution. However, there is little evidence that molecular change occurs at constant rates, and there is thus little basis for assuming that most molecular change is selectively neutral." However, Kimura (1987) shows that substantial nonconstancy does not invalidate the neutral theory.

Hill (1985) reported an "early hominid from Baringo, Kenya" dated at ca. 5 Myr ago. This is about 1 Myr older than the oldest *Australopithecus afarensis* material from Laetoli, Tanzania, thus nearer the minimum date of 5.5 Myr ago for the split between the human and chimpanzee lineages based on our DNA hybridization data.

Martin (1985) used the tandem scanning reflected light microscope (TSRLM) to examine the fine structure of the tooth enamel in primates. There are three patterns, one of which is shared by apes and

humans. Within these taxa the thickness of the enamel varies. Thick enamel seems to be the primitive condition, which is retained in the human lineage, but the gorilla and chimpanzees have thin enamel. This shared derived condition is proposed as a synapomorphy linking the African apes to one another by Martin (1985) and by Andrews (1987, see below).

Andrews (1985) proposed a classification based on morphological evidence in which *Pan, Gorilla,* and *Homo* are placed in the Homininae, and *Pongo* and *Sivapithecus* in the Ponginae.

Pilbeam (1985) reviewed the fossil evidence and suggested that the divergence of the orangutan lineage occurred between 12 and 15 Myr ago, that the human lineage split from the African apes 5–10 Myr ago, probably in Africa, and that the gibbon branch diverged ca. 20 Myr ago or more.

Scott et al. (1984) compared the sequences of the fetal globin genes in human, gorilla, chimpanzee, and two cercopithecids, *Macaca* and *Papio.* The differences among these taxa were small, but the authors concluded that "human and chimpanzees share the derived feature and thereby a more recent common ancestor" (Scott et al. 1984, p. 387). This analysis involves comparisons of sequences, thus qualifying as trait data under cladistic definitions.

Chang and Slightom (1984), Goodman et al. (1984), and Koop et al. (1986) studied the beta-globin gene family in the primates and other mammals. Koop et al. (1986a) compared the DNA sequences of the eta-globin genes of the hominoids (except gibbons), a macaque (rhesus), a New World monkey (owl monkey), a lemur, and domestic goat. They concluded that their "findings substantially increase the evidence indicative of a human–chimpanzee–gorilla clade with ancestral separations around 8 to 6 Myr ago. We also verify that neutral hominoid DNA evolved at markedly retarded rates" (Koop et al. 1986a, p. 234).

Miyamoto et al. (1987) extended the DNA sequence in the eta-globin gene region to 7.1 kilobase pairs for orangutan, gorilla, common chimpanzee, and human. From a parsimony analysis they concluded that the branching pattern of Fig. 1-1 is supported. This is further evidence that DNA hybridization and DNA sequences produce the same answer to phylogenetic questions.

Hasegawa and his colleagues have examined the molecular data pertaining to hominoid evolution and have subjected them to several statistical analyses. Hasegawa and Yano (1984a) analyzed the mtDNA sequences of 896 nucleotides from five species of hominoids (Brown et al. 1982), bovine (*Bos*), and mouse (*Mus*). They applied the maximum-likelihood method developed by Felsenstein (1981) to these data and concluded that a chimpanzee–human clade, as in our Fig. 1-1, was indicated. They calibrated their data against an assumption that the divergence between primates and ungulates occurred 65 Myr ago, which gave dates for the branch of the gibbon lineage as 13.7 Myr ago, orangutan 11.6 Myr ago, gorilla 3.6 Myr ago, and chimpanzee–human 2.5 Myr ago. Since there are well-identified australopithecine fossils at ca. 3.5 Myr ago, it is clear that the calibration is not correct.

Hasegawa and Yano (1984b) used computer simulation to evaluate the maximum-likelihood and maximum-parsimony methods for inferring phylogenetic trees, based on the sequences of 896 nucleotides of mitochondrial DNA (mtDNA) (Brown et al. 1982), as in Hasegawa and Yano (1984a). They also (Hasegawa and Yano 1984b) analyzed the sequences of the *psi* eta-globin genes of the beta-globin gene family from human, chimpanzee, gorilla, owl monkey, and a lemur that were sequenced by Chang and Slightom (1984), Harris et al. (1984), and Goodman et al. (1984). They assumed a divergence time for the split between the hominoids and New World monkey clade (Ceboidea) as 38 Myr ago, and calculated the divergence of the gorilla lineage as 5.8 Myr ago and that between human and chimpanzee as 5.2 Myr ago. They noted that "these datings seem to contradict . . . those from mitochondrial DNA data, and this discrepancy may suggest the possibility of mitochondrial DNA transfer between proto-human and proto-chimpanzee after the former had developed bipedalism" (Hasegawa and Yano 1984b, p. 4).

Hasegawa et al. (1984) "performed a direct comparison among mitochondrial DNA (mtDNA) sequences" and differentiated between the "numbers of transition and transversion type differences between species . . . ." They assumed a 90-Myr-ago divergence between bovids and primates and concluded that the gibbon lineage split from the other hominoids ca. 19.1 Myr ago, the orangutan lineage 15.9 Myr ago, gorilla 4.9 Myr ago, and the chimpanzee–human divergence 3.4 Myr ago.

Hasegawa et al. (1985) used a "new statistical method for estimating divergence dates" and calibrated the mtDNA data by assuming a 65-Myr-ago divergence between primates and ungulates. Using a "generalized least-squares method" they calculated dates of 92.3 Myr ago for the mouse divergence, 13.3 Myr ago for the gibbon branch, 10.9 Myr ago for the orangutan branch, 3.7 Myr ago for the gorilla branch, and 2.7 Myr ago for the divergence between the human and chimpanzee lineages. They noted the problem of reconciling their dates with the 3.7-Myr-ago date for the fossils of *Australopithecus afarensis* and suggested (Hasegawa et al. 1985, p. 160) that "mtDNA was transferred through hybridization between a proto-human and a proto-chimpan-

zee after the former had developed bipedalism." Hasegawa et al. (1985, p. 172) also discussed the datings from other studies and concluded that "The DNA sequence data presently available for setting our molecular clock are limited and the clock cannot always determine which one of the various possibilities discussed in this paper is the truth. . . . In this paper we have demonstrated that chimpanzee and human are far more closely related genetically than is generally believed."

Hasegawa et al. (1987) estimated divergence dates among primates by a molecular clock analysis based on the sequence of the eta-globin pseudogene, and calibrated by setting the divergence date between the Catarrhini and Platyrrhini at 38 Myr ago. This gave divergence dates of 25.67 Myr ago for the Old World monkeys (based on *Macaca*), 13.11 Myr ago for the orangutan lineage, 5.87 Myr ago for the gorilla split, and 5.16 Myr ago for the divergence between the chimpanzee and human branches. They placed 95% confidence intervals on each separation that gave a span of 4.10–7.83 for the gorilla branch, and 3.13–6.98 for the chimpanzee–human branch. They suggested that the eta-globin pseudogene evolved more rapidly early in primate evolution, later decreased in rate in the anthropoid lineage, and decreased even more in the hominoid lineage compared with the rate in the cercopithecoids. If these rate changes are correct, they may reflect the increase in the age of first breeding that accompanied the evolution of the higher primates.

Nei and Tajima (1985) developed a mathematical theory for the evolutionary change of restriction endonuclease cleavage sites, and evaluated the probabilities of various types of restriction site changes. They found that parsimony methods often make erroneous inferences about the construction of phylogenetic trees unless the number of substitutions per site is less than 0.01 for all branches. They reexamined the mtDNA sequence data of Ferris et al. (1981) and concluded (Nei and Tajima 1985, p. 189) that their analysis "does not support Templeton's [1983] conclusions regarding the phylogenetic tree for man and apes and the molecular clock hypothesis." Nei and Tajima (1985, p. 200) showed that Templeton's preference for the chimpanzee–gorilla–human branching sequence (Fig. 1-2) is no better supported than is the trichotomy (Fig. 1-4). Nei and Tajima (1985, p. 202) also showed that "contrary to Templeton's conclusion, the molecular clock hypothesis cannot be rejected by his data."

Willard et al. (1985) compared the DNA base sequences of the zeta 1 globin gene, plus an additional 1 kb of flanking sequences, in human and "chimpanzee" (presumably *Pan troglodytes*). The zeta 1 globin genes in both species contain two introns and there are 12 differences between the 650-bp single-copy sequences in the first introns of the two genes. This difference is 1.85%, which is close to the 1.63% we (Sibley and Ahlquist, this manuscript) found between the total single-copy genomes of *Homo* and *Pan*. Since the introns are believed to be under little selective pressure, this difference presumably reflects the rate of accumulation of neutral, or nearly neutral, mutations.

O'Brien et al. (1985) included data on hominoid relationships in a DNA–DNA hybridization study of several mammalian taxa. They obtained an average delta value of 1.80°C between human and gorilla, 1.85 between human and common chimpanzee, and 1.95 between chimpanzee and gorilla. These delta values "were based on 2–9 hybridizations." The authors did not claim that these data resolved the branching order among the African ape and human lineages. [See Ruvolo and Smith (1986, p. 287) for additional comments.]

Hixson and Brown (1986) sequenced 955 nucleotide positions from the mtDNAs of the common chimpanzee, pygmy chimpanzee, gorilla, and orangutan, and compared them with the human sequence. They concluded that the data "support the notion of an approximately equidistant relationship among chimpanzees, gorilla, and man, with the orangutan much less closely related. However, inference from a shared deletion suggests that the gorilla and the chimpanzees may be more closely related to one another than they are to man" (Hixson and Brown 1986, p. 1).

Bishop and Friday (1985, 1986) analyzed the 896-nucleotide mtDNA sequence of Brown et al. (1982) by the maximum-likelihood method and concluded that the chimpanzee–human–gorilla sequence (Fig. 1-1) was the most likely to be the correct tree: "It is of interest, however, that [the best fitting tree has] been regarded as the 'best' pattern by Sibley and Ahlquist (1984) on the basis of parsimony analysis of DNA hybridization data" (Bishop and Friday 1986, p. 152). They cited other studies both supporting and not supporting the branching pattern of Fig. 1-1, and concluded (Bishop and Friday 1986, p. 154) that "The trees with the greatest likelihoods . . . both group *Homo sapiens* and *Pan* together, and are uncertain about the location of *Pongo*. The [best fitting] tree . . . happens to correspond with the 'best fit' tree of Sibley and Ahlquist (1984). . . ."

Lanave et al. (1986) used "a stationary Markov model" to estimate the transition and transversion rates for 720 bp in the mtDNA sequences of pairs of hominoid species. They supported the *Pan–Homo* clade and noted (Lanave et al. 1986, p. 278) that "Sibley and Ahlquist (1984) . . . arrived at the same quantitative branching order we obtained. . . ."

Koop et al. (1986b) mapped and sequenced the epsilon globin gene and seven surrounding Alu re-

peat sequences in orangutan and compared them with the homologous sequences in *Homo*. The average divergence for all noncoding regions was 3.2%, which "corresponds closely to the . . . 3.6% . . . estimated from DNA hybridization . . . (Sibley and Ahlquist 1984)" (Koop et al. 1986b, p. 98).

Saitou and Nei (1986) analyzed 1834 bp of human, chimpanzee, and gorilla mtDNAs using six different tree-making methods. All gave "the same phylogenetic tree in which humans and chimpanzees are most closely related" (Saitou and Nei 1986, p. 189).

Sakoyama et al. (1987) compared the sequences of 2200 bp in the immunoglobulin epsilon-chain genes of human, chimpanzee, and orangutan, and calculated divergence dates based on the hominoid–cercopithecoid split as 30 Myr ago. The mean dates were 6.4 ± 2.6 Myr ago for human–chimpanzee, and 17.3 ± 4.5 Myr ago for human–orangutan, thus "· · · generally consistent with those" of Sibley and Ahlquist (1984) (Sakoyama et al. 1987, p. 1083).

Goldman et al. (1987) used two-dimensional electrophoresis to compare 383 radiolabeled fibroblast polypeptides among the hominoid genera and a cercopithecoid (*Macaca fascicularis*). Their data produced the same branching sequence as in our 1984 and present studies. To calibrate the genetic distances they used the fossil-based dating of the orangutan branch at 13 Myr ago and calculated the split between hominoids and cercopithecoids as 37 Myr ago, the gibbon branch as 20–25 Myr ago, the gorilla divergence soon after that of the orangutan (ca. 12 Myr ago), and the chimpanzee–human divergence as "about 8 Myr ago."

Ruvolo and Pilbeam (1986) briefly reviewed the molecular and fossil data of hominoid evolution and supported a *Pan–Homo* clade.

Martin (1986) reviewed six possible phylogenetic trees for the hominoids and assumed that morphological variation "should form the basis for the assessment of their relationships." He made a cladistic analysis of 109 skeletal and 13 soft tissue characters, of which 43 were identified as derived within the clade. Martin followed Andrews (1986) in his decision that molecular evidence is insufficient to support the resolution of the branching pattern within the African ape–human clade. Martin accepted the combined morphological and molecular evidence for an African ape–human clade, but concluded (Martin 1986, p. 173) that

> Within the African ape/human clade there is only one character (a spatulate upper lateral incisor) supporting a chimp/human clade, and no evidence for a gorilla/human clade. The shared derived possession of secondarily reduced enamel thickness, in addition to shared specializations relating to knuckle-walking, is convincing evidence for an African ape clade. . . .

Martin (1986, p. 175) presented the cladist's standard view of molecular distance data as based on

> 'similarity', which was considered, in some unspecified way, to be equivalent to phylogenetic affinity. In general, scientists working from a molecular data base have continued to rely on similarity to reconstruct phylogeny and some have argued for a chimp/human clade within the African ape/human clade on that basis. The molecular data do not lend themselves to phylogeny reconstruction which is best achieved on the basis of the shared possession of derived characters. The molecular data have been analysed in a cladistic framework by Andrews (this volume [1986]), who found that they support an African ape/human clade, but were not definitive concerning relationships within that clade. A further problem with the molecular data is that they show rather low levels of variability among members of the great ape and human clade, and especially among the African ape/human clade. In view of the greater known morphological variability within these clades it seemed likely that morphology would provide a better basis for resolving great ape and human relationships at the present time. Morphological data also have the major advantage that they can often be applied to fossil material.

It is important to recognize that DNA–DNA hybridization does not measure "similarity" in the same sense that morphological similarity is judged by the human eye. DNA distance values are measures of median sequence divergence and, unlike morphological characters, they are not subject to the effects of convergent evolution. Neither is it true that because distance data cannot be partitioned into primitive and derived traits that they, therefore, cannot be used to reconstruct phylogeny. These false notions have repeatedly been used by cladists to discredit distance data without offering proof that the phylogenies produced by such data are wrong. It is the *complexity* of DNA sequences that gives DNA–DNA hybridization its ability to separate analogy from homology, a property lacking in the few characters used by morphologists, as in the present example of the branching pattern of the hominoids. Martin (1986) is satisfied that he has obtained the correct pattern on the basis of two characters of uncertain phylogenetic value, namely, tooth enamel thickness and knuckle-walking. In addition, he acknowledges one character (spatulate upper lateral incisor) that supports a chimpanzee–human clade. In effect, he is basing his conclusion on a difference of 1 character out of 123 to define an African ape clade.

For comparison, consider the following: The single copy genome of the higher primates contains ca. 2×10^9 base pairs. The delta $T_{50}H$ between the divergences of the gorilla lineage and the chimpanzee–human lineage is 0.7. Using the generally accepted relationship of delta 1.0 = 1% base mismatch, the delta 0.7 translates into 0.007%, or ca.

14×10^6 base pair mismatches, as the difference that accumulated during the time between the divergences of the gorilla lineage and the chimpanzee–human clade. By the same logic, the delta 1.6 between living chimpanzees and humans indicates a base pair mismatch of ca. 32×10^6, and between living gorillas and either chimpanzees or humans, ca. 46×10^6 mismatches (delta 2.3).

Jones (1986) reviewed the results of several studies including molecular structure, blood groups, cell surface polymorphisms, chromosomes, proteins, DNA sequences, DNA hybridization, and mtDNA. He concluded (1986, p. 327) that "the new information on molecular anatomy may mean that we still know rather little about the genetic changes involved in the origin of man."

Andrews (1986, p. 107) defines three "levels" of competence in molecular methods in terms of "their results rather than their methods."

1) Electrophoresis and immunodiffusion "provide indices of change for whole proteins without necessarily knowing exactly what is changing. Similar investigations of DNA structure by hybridization techniques detect changes in the DNA molecule without knowing exactly which nucleotides have changed. All these methods produce distance statistics as evidence of similarity or dissimilarity between taxa."

2) "At a second level, protein alteration is specified by amino acid sequencing, so that changes in protein can be defined in terms of known changes in amino acids at specific loci."

3) "In some cases, the DNA coding for the amino acids may also be known, and this is the third level of analysis, DNA sequencing. An approach to this is made by restriction enzyme mapping, which breaks the DNA chains into identifiable segments, but the most interesting results are from the actual sequencing data of DNA chains."

These statements reflect Andrews's preference for trait data that can be analyzed by cladistic procedures. Distance data are unacceptable to some cladists, and Andrews's three "levels" reveal this predilection. The notion that protein sequences are more informative than the data of DNA hybridization is erroneous; the difference lies not in whether "exactly what is changing" is known, but in the *complexity* of the data and whether they can distinguish analogy from homology. DNA hybridization data are enormously complex, indexing essentially the entire information content of the genome, while a protein sequence provides indirect information about a few hundred to a few thousand bases. Cladists prefer such data because they can be partitioned into primitive and derived characters, providing one has homologous sequences of the same protein from appropriate taxa.

DNA sequences may be superior to DNA hybridization data, but the meager available evidence suggests that the two methods will give the same answer to any given question (Britten 1986; Miyamoto et al. in press). Until more sequences of comparable taxa are available this question is moot. It may be some time before the answer is clear because sequencing remains a slow process in spite of improvements in the instrumentation.

There is no obvious advantage in knowing "exactly what is changing" unless one is committed to the Hennigian procedure. We find the measurement of the *median sequence divergence* values between single-copy DNAs to be able to reproduce any definitely known pattern of relationships, and to reveal errors due to convergence. This is a claim that cannot be made for morphological characters, however they may be analyzed.

Andrews repeats his criticism of distance data (1986, pp. 110–111) and cites our (Sibley and Ahlquist 1984) DNA hybridization data on the hominoids. He discredits our "association between chimpanzee and man within the African ape and man group" by claiming that the standard deviations are so large that the "result cannot be regarded as significant." On page 124 Andrews (1986) repeats this assertion, in spite of the fact that we performed a *t*- test for the difference and found it to be statistically significant. For additional evidence on this point we refer the reader to the statistical analysis of our data by Felsenstein (1987) and the statistical studies of our data by Lausen and Degens (1986) and Degens and Lausen (1986).

Andrews (1986, p. 112) presented a "theoretical example applied to DNA hybridization data" in which he set up an unrealistic arrangement of hypothetical characters that omits the principal reason that DNA hybridization data are able to reveal phylogeny, namely, *complexity*. It is not reasonable to think that a set of nine units, arranged to prove a preconceived result, has any relevance to reality. DNA hybridization depends on the comparison of millions to billions of base pairs, and a realistic model must take this into account.

Andrews (1986, p. 120) states that the mtDNA analysis of Hasegawa et al. (1984) obtained dates for the hominoid branchings that "are quite similar to those obtained from DNA hybridization." If the reference is to our data (Sibley and Ahlquist 1984), we disagree. Hasegawa et al. (1984) calculated the gorilla branch as 4.9 Myr ago and the chimpanzee–human divergence as 3.4 Myr ago; our dates for these branches were 8–10 Myr ago for the gorilla branch and 6.3–7.7 for the chimpanzee–human branch. We do not regard the two sets as "quite similar" since our dates are twice as long ago as those of Hasegawa et al. (1984). The latter did obtain

dates similar to ours for the gibbon and orangutan branches, but the orangutan was based on fossils, not on molecules.

Andrews (1986, pp. 124–125) discussed the properties of sequence data, distance data, and morphology as sources of evidence. He claimed that sequence data, even the short sequences of proteins, are better than distance data. However, Andrews acknowledges that morphological characters are not an attractive alternative because "we can do little more than guess about convergence." In spite of his animadversion for distance data, Andrews concluded (1986, p. 125) that "the balance of evidence is in favour of the chimpanzee/man link, but clearly the evidence leaves a lot to be desired."

Andrews (1987, p. 24) again reviews the morphological and molecular methods as evidence of hominoid phylogeny and states his "bias in favour of the morphological end of the spectrum."

We have little quarrel with most of Andrews's (1987) discussion of DNA hybridization data and will comment on only a few points. On page 31 (Andrews 1987) he notes that "The problem with DNA–DNA hybridization data is the same as for any distance statistic, that it is not possible to isolate the changes that are being measured, and so is inherently untestable." We disagree that the method is untestable. The ultimate test will be congruence with DNA sequences and, to date, the few examples available suggest that the two methods will give the same answer. They should, because DNA–DNA hybridization may be viewed as an indirect method of sequencing. The success of the technique depends on the fact that only homologous sequences that are complementary at 75–80% of their base pairs will form thermally stable duplexes at the standard criterion of 60°C. The use of fragments with an average length of ca. 500 bases precludes the formation of duplexes between short sequences that are similar by chance or by convergence. This is because a DNA sequence of 500 bases can occur in 10^{301} different arrangements, which is more than the number of ultimate particles in the Universe. It is inconceivable that chance, or convergence, could produce the 75–80% of base pairing required for the production of a stable duplex at 60°C. Thus, in effect, the two genomes that are hybridized "sequence" one another, and the median melting temperature of the hybrid molecules tells us the percentage of mismatch between them. This is true because a delta value of 1°C is produced by a mismatch of ca. 1% of the bases (Bonner et al. 1973; Britten et al. 1974; Jacobs et al. 1983). We do not know exactly which bases are mismatched, but the percentage of mismatch is as useful for phylogeny reconstruction as the actual sequence.

Andrews (in press, p. 31) states that:

> Similar characters, be they complex morphological characters or nucleotide substitutions, may be inherited unchanged from a remote common ancestor, in which case they are primitive retentions, or they may be inherited from the last common ancestor, in which case they are derived. The small number of derived characters uniquely shared between two closely related species would be swamped by the much larger number of shared primitive characters unless the latter can be eliminated from consideration, but this is exactly what has not been done by the hybridization method.

Figure 2 illustrates why DNA hybridization data may be used to reconstruct phylogeny, regardless of rate and without confusing primitive characters and derived characters. The common ancestor of a and b is defined by the base sequences at x. The distance from a to b, measured by DNA hybridization, equals the sum of the autapomorphous changes in lineages x–a and x–b. The fact that only autapomorphous changes are measured negates the effects of symplesiomorphy and synapomorphy, which are constants at x. If there is a difference in rate, for example in lineage x–b, it will be detected by comparisons between a and c and between b and c because branch lengths x–y and y–c are constant for these measurements.

Our studies of the hominoids (Sibley and Ahlquist 1984 and the present paper) satisfy these conditions. If the technique does not provide a valid basis for phylogeny reconstruction, why does it agree with the overwhelming evidence of the branching pattern of the hominoids produced by other methods, including morphology? The *only* branching still being seriously debated is that of the position of the chimpanzees—are they closer to humans or to gorillas? Miyamoto et al. (1987) have now provided sequence data that support our tree of the hominoids, including the chimpanzee–human clade. Will Andrews, and others, accept this as the validation of the DNA hybridization evidence?

Andrews (1987, pp. 43–44) concludes that there are only two morphological characters that link chimpanzees and gorillas, namely, knuckle-walking and thin tooth enamel, "therefore, there is very substantial morphological evidence supporting a link between chimpanzees and gorillas. . . . In contrast to all of the possible cladistic groupings so far discussed, there is no morphological evidence whatever supporting a cladistic relationship between humans and chimpanzees." Andrews (1987, p. 45) again claims that DNA hybridization "produces a distance statistic which fails to distinguish character homology, and this must raise questions about the reliability of the DNA–DNA hybridization results." Again, we consider this to be a false criticism based on the assumption that all distance data are incom-

petent and that only character data can "distinguish character homology." Andrews, and other critics, must prove that our results are consistently wrong, not merely that they prefer traits that can be partitioned into primitive and derived states. Andrews (1987, pp. 48–49) concludes that

> The morphological evidence and some molecular interpretations show chimpanzees and gorillas to be more closely related, while some of the molecular evidence shows humans and chimpanzees to be more closely related. Amino acid sequencing and DNA–DNA hybridization provide the strongest support for the human–chimpanzee link, while gross morphology supports the African ape grouping. DNA sequencing [= mtDNA] which ought to provide the answer to this problem, produces ambiguous results: different interpretations of the same data lend support to both alternatives. In view of this, and the strength of the morphological evidence, my conclusion here is that chimpanzees and gorillas share a common ancestry separate from humans, and together form the sister-group to humans.

Thus, in two subsequent papers, Andrews (1986, 1987) comes to different conclusions based on the same evidence.

Pilbeam (1986) reviewed essentially the same evidence used by Andrews. With reference to enamel thickness, Pilbeam noted that although *Ramapithecus* and *Australopithecus* share thick enamel, they have proved to be phylogenetically distant from one another and "the similarity of *Ramapithecus* and *Australopithecus* relative to the chimpanzee tells us nothing about their relationship. (In fact, *Australopithecus* and the chimpanzee are more closely related than either is to *Ramapithecus*)" (Pilbeam 1986, p. 296). Pilbeam reviewed the DNA hybridization data and concluded that "humans and chimpanzees are probably closest relatives (p. 301)." He also noted (p. 301) that

> The precise relationship between any of these genetic differences and time is less clear. As Gingerich (1985) has shown, there is no way that relationship can be known except by using geological or paleontological events to estimate splitting times, and results are clearly dependent on the quality of those estimates, few of which are very good. The rate test is not a test of linearity. It is indeed unlikely that any system is linear. But even if they are not linear the local pattern for hominoids over the last 20 million years of evolution is likely to be sufficiently close to linear for us not to have to worry too much. These results have not been accepted without a fight. Many morphologists still react strongly, as though "molecular" systematics puts us all out of a job. Quite the contrary. If we can infer relationships correctly, using whatever methods can be shown as consistent (as DNA hybridization patterns of hominoids are, when judged by the rate test), we have a better guide to the interpretation of anatomy.
>
> Given that the branching sequence indeed links hominids and chimpanzees, with gorillas more distant, this may well indicate that protohominids included knuckle-walking in their behavior, but it does not necessarily mean they closely resembled chimpanzees. For example, just from knowledge of living hominoids one would not have predicted *Australopithecus*. This is because not all possible character states are represented in living species, because past species are usually novel combinations of character states, and because we often have only a hazy idea of polarities—what is primitive and what is derived. As an example of the difficulties, consider enamel thickness once more. In hominids [= human lineage] it is thick, while chimpanzees and gorillas have thin enamel, which was believed to be a derived homology implying a chimpanzee–gorilla link. There is histological and fossil evidence that thick enamel is primitive (Martin 1985; Pilbeam 1985), a character state retained in hominids. But if humans and chimpanzees are closest, the thin enamel of chimps and gorillas must be a homoplasy [= convergent]. It looks as though parallelism is widespread among hominoids in many systems.
>
> To reconstruct the protohominid pattern we clearly need both molecular and comparative records, and we need to play those records off against the fossils.

It is obvious that the search for the correct branching sequence of the hominoid lineages has been controversial. Although a consensus has developed about the relative branchings of the gibbon and orangutan lineages, the debate about the branching sequence of the human, chimpanzee, and gorilla lineages continues. The publication of our paper 4 years ago (Sibley and Ahlquist 1984) elicited considerable interest, pro and con. It was the first study using DNA hybridization that presented a complete matrix of distance values based on the averages of five or more comparisons for each node, and it was the first to present molecular evidence for a *Pan–Homo* clade. We had examined many similar situations among birds and assumed that the results would be accepted readily. Our naive assumption proved to be wrong, and the debate intensified rather than abated. The most vigorous attack on our study was presented by Templeton (1985); the rebuttal was made by Saitou (1986), Ruvolo and Smith (1986), and Fitch (1986). Templeton (1986) responded. We will not review these interesting papers, but suggest that those interested in the problem consult the originals.

The critique by Templeton (1985), and the subsequent debates, stimulated us to make over 300 additional DNA–DNA comparisons among the hominoids, mostly among the chimpanzee, gorilla, and human single-copy genomes. The present paper is the report on this expanded study, and we offer this larger data set for examination and analysis by those so inclined. Lausen and Degens (1986), Degens and Lausen (1986), and Felsenstein (1987) have made statistical analyses of this expanded data set that support the tree depicted in Figs. 1-1, 3, and 7.

Methods and Results

For descriptions of our methods see Sibley and Ahlquist (1983, 1984).

Tables 1–7 and Figs. 1–8 present our data. As in

Table 1. Complete set of 514 delta $T_{50}H$ values among the hominoids. This table includes the 183 values from Sibley and Ahlquist (1984), plus 331 from subsequent comparisons. The data are arranged in six columns: (1) tracer species, e.g., Pt; (2) tracer preparation number, e.g., 2; (3) driver species, e.g., Pp; (4) driver preparation number, e.g., 1; (5) experimental set, e.g., 786; (6) delta $T_{50}H$ value, e.g., 0.4.

Pt	2	Pp	1	786	0.4
					0.6
				842	0.5
		Hs	1b	785	1.5
				786	1.5
					1.7
				842	1.6
					1.7
					1.8
					1.8
		Gg	2	785	2.0
				786	2.1
					2.3
				842	1.8
					2.1
					2.1
					2.2
		Po	2	785	3.8
				786	3.0
					3.2
				842	3.5
					3.7
					3.8
					3.8
		Hy	1	786	5.0
					5.5
		Hl	2	842	4.6
					4.7
					4.8
					5.1
		Ca		786	6.3
					6.9
				842	7.0
		Ph		786	7.4
					7.8
				842	7.2
					7.7
		Mm		842	7.1
					7.3
		Pn		842	7.8
Pt	3a	Hs	1c	848	1.7
				1152	1.4
					1.4
					1.5
					1.6
					1.6
					1.6
					1.6
					1.6
					1.7
					1.7
					2.1
				1163	1.1
					1.3
					1.4
					1.4
					1.4
					1.4
					1.5
					1.6
					1.7
					1.9
					2.0
					2.2
				1166	1.5
		Gg	3	848	2.5
				1163	1.9
					2.0
					2.0
					2.2
					2.3
					2.4
					2.4
					2.5
					2.6
		Gg	4	1152	2.1
					2.1
					2.1
					2.1
					2.2
					2.2
					2.2
					2.3
					2.3
					2.4
					2.5
					2.6
				1163	1.9
					1.9
					2.5
				1166	2.4
		Po	2	848	3.2
				1166	3.6
					3.7
					3.8
					3.8
					3.9
		Hl	1	848	5.5
		Hl	2	1166	4.1
					4.8
					5.1
					5.1
		Ca		848	7.3
				1166	7.7
		Ph		848	7.1
					7.4
				1166	6.5
		Mm		848	7.2
				1166	7.3
		Ms		1166	7.9
		Pp	1	Pt	2	790	1.1
				Pt	3a	843	0.5
							0.8
							0.9
							1.0
						849	0.4
				Gg	2	790	2.6
						843	2.1
							2.3
							2.4
							2.8
				Gg	3	849	2.3
				Hs	1b	790	1.5
					1c	843	1.6
							1.8
							1.8
							1.9
							2.0
							2.0
							2.1
							2.1
						849	1.5
		Po	2	790	3.5
				843	3.6
					3.7
					3.7
					4.1
				849	3.6
		Hy	1	790	4.2
		Hl	2	843	5.1
					5.2
					5.2
					5.3
				849	4.2
		Ca		790	6.7
				843	6.9
					7.2
				849	6.5
					6.8
		Ph		790	6.9
				849	6.6
					7.4
		Mm		843	7.4
					7.7
				849	7.4
Hs	1a	Pp	1	581	1.6
					1.6
		Pt	1	581	1.8
					1.9
		Gg	1	581	2.6
					2.8
		Po	1	581	3.8
					3.9
		Hl	1	581	4.8
					4.8
		Hy		581	4.2
					4.8
		Ca		581	7.6
					7.6
		An		581	7.0
					7.1
		Mm		581	7.3
					7.3
		Nl		581	6.9
Hs	1c	Pt	3a	844	1.4
					1.5
					1.6
					1.6
				847	1.8
				1151	1.7
					1.7
					1.7
					1.8
					1.8
					1.9
					2.0
					2.0
					2.1
					2.1
				1154	1.3

Table 1 continued on pp. 108 and 109

Table 1. Continued

			1.5					847	5.2						3.8
			1.5				Hl	3	1165	4.0					4.0
Pt	3b	1154	1.1				Mm		844	7.1					4.0
			1.3							7.5					4.1
			1.4						847	8.0				869	3.8
			1.5				Ph		844	7.3			Hl	2	864 4.6
			1.5							7.4					4.6
			1.5						847	7.7					4.6
			1.6							7.7					4.7
			1.9						1165	6.7					5.3
			2.0				Pn		844	7.6				869	4.5
		1165	1.2				Ca		1165	7.1			Mm		864 7.5
			1.5				Ms		1165	7.5				869	7.2
			1.5	Gg	2		Pt	2	785	1.9					7.4
Pp	1	844	1.3						828	1.7			Ph		864 6.9
		847	1.7						831	2.1				869	6.3
		1165	1.7							2.1					6.9
Gg	2	844	2.4							2.1			Ca		869 7.2
			2.5							2.3			An		869 6.5
			2.6				Pp	1	828	2.2	Gg	4	Pt	3a	1153 2.0
			2.6						831	2.2					2.0
Gg	3	847	2.5							2.4					2.1
		1165	1.7							2.5					2.2
			1.8							2.6					2.2
			1.9				Hs	1b	785	2.2					2.4
			2.0						828	2.4					2.4
			2.0						831	2.2					2.4
			2.2							2.3					2.5
			2.3							2.3					2.5
			2.3							2.7					2.6
		853	2.3				Po	2	785	3.3					2.6
Gg	4	1151	2.0						828	3.1				1164	2.1
			2.0						831	2.9					2.5
			2.1							3.0				1167	2.0
			2.1							3.6			Pt	3b	1164 2.1
			2.1							3.7					2.1
			2.2				Hy	1	785	4.8					2.2
			2.2						828	4.4					2.2
			2.2						831	4.3					2.2
			2.2							4.4					2.2
			2.4							4.5					2.2
			2.4							4.9					2.2
			2.4				Ca		785	7.9					2.3
		1154	1.6						828	6.5					2.4
			1.9							7.0			Hs	1c	1153 1.9
			1.9						831	7.2					2.2
			2.3							7.3					2.2
			2.5							7.4					2.2
Po	2	844	3.3							7.4					2.3
			3.5				Mm		828	7.3					2.3
			3.7							7.5					2.4
			3.9				Ph		828	7.0					2.5
		847	3.8				Pn		828	6.2					2.6
Po	3	1154	3.6	Gg	3		Pt	3a	864	1.9					2.6
			3.5							2.1					2.7
			3.7							2.3					2.7
			3.7							2.3				1164	2.1
			3.7						869	1.9					2.1
			4.0				Pp	1	864	2.0					2.2
			4.2				Hs	1c	864	2.1					2.2
		1165	3.1							2.1					2.2
Hl	2	844	4.9							2.2					2.3
			4.9							2.4					2.3
			4.9						869	2.3					2.4
			5.0				Po	3	864	3.5					2.4

Table 1. Continued

					2.4					3.5								4.6
					2.5					3.7						Hs	1c	856	4.7
					2.5				Hs	1a	790	3.2							4.8
				1167	2.0						822	2.9							4.9
					2.3							3.1							5.2
		Po	3	1167	3.2							3.8						859	4.3
					3.3				Hs	1b		3.5							4.5
					3.4						809	3.6				Gg	3	856	4.2
					3.6							3.8							4.5
		Hl	3	1167	4.5				Gg	2	790	3.7							5.0
					4.6						809	3.7							5.0
					4.6							3.7						858	5.1
					4.7							3.8						859	4.9
		Ca		1167	7.8						822	3.0							5.4
		Mm		1167	7.2				Hl	1	809	4.9				Po	3	859	4.3
		Ms		1167	7.4							5.1							4.3
					7.5							5.3							4.4
		Ph		1167	7.3				Hy	1	790	4.6							4.5
Po	1	Pt	1	679	3.7						809	5.0							5.3
					3.9							5.0				Mm		856	6.6
					4.1							5.2							7.1
		Pp	1	679	3.4						822	5.0				Ph		856	6.8
					3.6				Ph		790	6.8						859	7.5
					3.6						822	6.9				Pn		856	6.7
		Hs	1a	679	3.1							7.3							7.5
					3.5				An		809	7.2		Ph	1	Pt	3b	1200	7.2
					4.1							7.4						1201	7.2
		Gg	1	679	3.5						822	7.4				Pt	4	1200	7.9
					3.6				Ca		822	7.4						1201	7.3
					3.9				Mm		822	6.8				Hs	1a	1200	7.9
		Hl	1	679	4.4				Pn		822	7.2				Hs	1b	1200	7.3
					4.5	Hy	1		Pt	2	785	4.9						1201	7.1
					5.2				Hs	1b	785	5.1				Hs	1c	1200	7.0
		Hy	1	679	4.8				Gg	2	785	4.5						1201	7.2
					5.0				Po	2	785	4.7				Gg	5a	1200	7.5
					5.1				Ca		785	7.2						1201	7.4
		An		679	7.8	Hl	2		Hy	1	856	1.7				Gg	5b	1200	7.3
					7.8							1.8						1201	7.1
Po	2	Pt	2	790	3.4							1.9				Po	3	1200	7.6
				809	3.4							2.4							8.2
					3.4				Pt	3a	856	4.1						1201	7.7
					3.5							4.5							7.9
				822	2.9							4.7				Hl	1	1201	7.0
		Pp	1	790	3.0							5.2							
				809	3.3						859	4.5							

Tissues from four individuals of *Pan troglodytes* were used. These are labeled Pt 1, Pt 2, Pt 3, and Pt 4. Tissues from Pt 3 were used to make two different DNA extracts; they are labeled Pt 3a and Pt 3b. The other DNA extracts follow the same coding. Only one sample each of *Pan paniscus* and *Homo sapiens* were used, but three tracers were prepared from the human tissues, Hs 1a, 1b, and 1c. Tissues from five individual gorillas, two orangutans, and two lar gibbons were used

Abbreviations: Pt, *Pan troglodytes*; Pp, *Pan paniscus*; Hs, *Homo sapiens*; Gg, *Gorilla gorilla*; Po, *Pongo pygmaeus*; Hy, *Hylobates lar*; Hs, *Hylobates syndactylus*; Ph, *Papio hamadryas*; Ca, *Cercopithecus aethiops*; An, *Allenopithecus nigroviridis*; Mm, *Macaca mulatta*; Ms, *Macaca silenus*; Pn, *Pygothrix nemaeus*; Nl, *Nasalis larvatus*

1984, we conclude that the phylogeny depicted in Figs. 1-1, 3, and 7 is supported by the data, and that it is highly probable that it represents the correct reconstruction of the phylogeny of the hominoid primates. We also conclude that the divergences occurred within the ranges of the datings indicated in Fig. 8, as follows: *Pan troglodytes–Pan paniscus*, 2.4–3.4 Myr ago; *Pan–Homo*, 5.5–7.7 Myr ago; *Gorilla*, 7.7–11.0 Myr ago; *Pongo*, 12.2–17.0 Myr ago; *Hylobates*, 16.4–23.0 Myr ago; Cercopithecoidea, 25.0–34.0 Myr ago. The minimum date in each pair is based on the fossil dating for the cercopithecoid branch as 25 Myr ago, and the maximum date is based on the fossil dating of the *Pongo* branch as 17.0 Myr ago.

Although there are differences in the age at first breeding among the living hominoids (Table 7), there is no detectable "generation time effect" in the data.

Table 2. Unfolded matrix of 510 delta $T_{50}H$ values, with the standard deviation (SD), standard error of the mean (SE), and number of observations (n) per cell. The four comparisons between *Hylobates lar* and *Hylobates syndactylus* were omitted. Comparisons between the two species of *Hylobates* and other hominoids, and comparisons between the hominoids and the cercopithecoids, have been combined.

	Pp	Pt	Hs	Gg	Po	Hy + Hl	Cercop.
Pan paniscus	0.00	0.78	1.70	2.42	3.70	4.87	7.04
Pygmy chimpanzee		0.28 SD	0.19 SD	0.25 SD	0.21 SD	0.52 SD	0.39 SD
		0.11 SE	0.08 SE	0.10 SE	0.09 SE	0.21 SE	0.12 SE
		$n = 6$	$n = 6$	$n = 6$	$n = 6$	$n = 6$	$n = 11$
Pan troglodytes	0.50	0.00	1.61	2.22	3.60	4.94	7.27
Common chimpanzee	0.10 SD		0.23 SD	0.22 SD	0.29 SD	0.40 SD	0.43 SD
	0.06 SE		0.04 SE	0.04 SE	0.08 SE	0.12 SE	0.10 SE
	$n = 3$		$n = 32$	$n = 33$	$n = 13$	$n = 11$	$n = 18$
Homo sapiens	1.58	1.65	0.00	2.21	3.69	4.75	7.35
Human	0.16 SD	0.26 SD		0.28 SD	0.27 SD	0.37 SD	0.33 SD
	0.07 SE	0.05 SE		0.05 SE	0.07 SE	0.12 SE	0.08 SE
	$n = 5$	$n = 32$		$n = 33$	$n = 15$	$n = 10$	$n = 18$
Gorilla gorilla	2.32	2.20	2.32	0.00	3.52	4.62	7.16
Gorilla	0.22 SD	0.21 SD	0.19 SD		0.37 SD	0.23 SD	0.43 SD
	0.09 SE	0.03 SE	0.03 SE		0.09 SE	0.06 SE	0.09 SE
	$n = 6$	$n = 36$	$n = 37$		$n = 16$	$n = 16$	$n = 24$
Pongo pygmaeus	3.44	3.54	3.46	3.61	0.00	4.94	7.27
Orangutan	0.24 SD	0.37 SD	0.38 SD	0.27 SD		0.27 SD	0.35 SD
	0.09 SE	0.13 SE	0.12 SE	0.10 SE		0.07 SE	0.10 SE
	$n = 7$	$n = 8$	$n = 10$	$n = 8$		$n = 14$	$n = 11$
Hylobates spp.		4.64	4.79	4.82	4.58	0.00	7.00
Gibbons		0.35 SD	0.32 SD	0.39 SD	0.38 SD		0.37 SD
		0.13 SE	0.12 SE	0.14 SE	0.16 SE		0.14 SE
		$n = 7$	$n = 7$	$n = 8$	$n = 6$		$n = 7$
Cercopithecid spp.		7.40	7.30	7.33	7.85	7.00	0.00
Old World monkeys		0.34 SD	0.35 SD	0.17 SD	0.26 SD		
		0.17 SE	0.16 SE	0.08 SE	0.13 SE		
		$n = 4$	$n = 5$	$n = 4$	$n = 4$	$n = 1$	

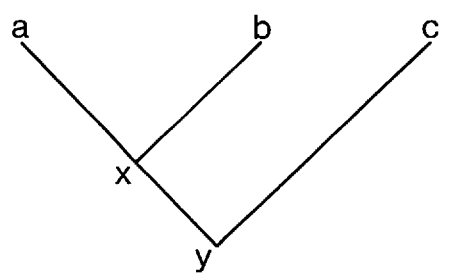

Fig. 2. See text for explanation

Felsenstein (1987) concluded that there is "no detectable departure from a molecular clock." Thus, the same average rate of nucleotide substitution apparently occurred along each of the hominoid lineages. See below for further discussion of these and related topics.

Discussion

The branching sequence is the first step in phylogeny reconstruction, but it leads to a consideration of the dates of the divergences, the rates of evolution along the branches, and the factors that determine rates and dates.

In our 1984 paper we used Pilbeam's (1983) estimate of the branching of the orangutan clade as 13–16 Myr ago to calibrate the DNA "clock." The results of this calibration were as follows:

> The delta $T_{50}H$ value for the *Pongo* divergence is 3.7. If we use Pilbeam's (1983) 16 MYA date for this node, delta $T_{50}H$ 1.0 = 4.3 MY, and if we use 13 MYA for the pongine divergence date, delta $T_{50}H$ 1.0 = 3.5 MY. If we use these two calibrations to calculate probable ranges of the hominoid divergence nodes we obtain the following: *Pan troglodytes–Pan paniscus*; 2.4–3.0 MYA, *Homo*, 6.3–7.7 MYA; *Gorilla*, 8.0–9.9 MYA; *Pongo*, 13–16 MYA; *Hylobates*, 18.2–22.4 MYA; *H. lar–H. syndactylus*, 7.7–9.5 MYA; Cercopithecoidea, 27–33 MYA (Sibley and Ahlquist 1984, p. 13).

We assumed that the DNA clock was running at the same average rate along all lineages, and we were surprised to find that the 4.3 calibration factor was close to those we had obtained from avian data, based on dates of divergences probably caused by geological events during the breakup of Gondwanaland. This seemed to confirm our belief in a "uni-

Table 3. Unfolded matrix of the 450 delta $T_{50}H$ values used by Felsenstein (1987) for his statistical analysis. Felsenstein used the comparisons between the hominoids and the Hamadryas baboon (*Papio hamadryas*), whose single-copy DNA was used as one of the tracers, but he excluded the comparisons between the hominoids and cercopithecoids, whose DNAs were not used as tracers. This eliminated 64 delta values between hominoid tracer taxa and the driver DNAs of the species of *Cercopithecus, Allenopithecus, Macaca, Nasalis,* and *Pygathrix*. See Table 4. The four delta values between *Hylobates lar* and *H. syndactylus* are included; they average delta $T_{50}H$ 1.95.

	Pp	Pt	Hs	Gg	Po	Hy	Hl	Ph
Pan paniscus Pygmy chimpanzee	0.00	0.78 0.28 SE 0.11 SE $n = 6$	1.70 0.19 SD 0.08 SE $n = 6$	2.42 0.25 SD 0.10 SE $n = 6$	3.70 0.21 SD 0.09 SE $n = 6$	4.20 $n = 1$	5.00 0.45 SD 0.20 SE $n = 5$	6.97 0.40 SD 0.23 SE $n = 3$
Pan troglodytes Common chimpanzee	0.50 0.10 SD 0.06 SE $n = 3$	0.00	1.61 0.23 SD 0.04 SE $n = 32$	2.22 0.22 SD 0.04 SE $n = 33$	3.60 0.29 SD 0.08 SE $n = 13$	5.25 $n = 2$	4.87 0.40 SD 0.13 SE $n = 9$	7.30 0.43 SD 0.16 SE $n = 7$
Homo sapiens Human	1.58 0.16 SD 0.07 SE $n = 5$	1.65 0.26 SD 0.05 SE $n = 32$	0.00	2.21 0.28 SD 0.05 SE $n = 33$	3.69 0.27 SD 0.07 SE $n = 15$	4.50 $n = 2$	4.81 0.35 SD 0.12 SE $n = 8$	7.36 0.41 SD 0.18 SE $n = 5$
Gorilla gorilla Gorilla	2.32 0.22 SD 0.09 SE $n = 6$	2.20 0.21 SD 0.03 SE $n = 36$	2.32 0.19 SD 0.03 SE $n = 37$	0.00	3.52 0.37 SD 0.09 SE $n = 16$	4.55 0.24 SD 0.10 SE $n = 6$	4.67 0.23 SD 0.07 SE $n = 10$	6.88 0.36 SD 0.16 SE $n = 5$
Pongo pygmaeus Orangutan	3.44 0.24 SD 0.09 SE $n = 7$	3.54 0.37 SD 0.13 SE $n = 8$	3.46 0.38 SD 0.12 SE $n = 10$	3.61 0.27 SD 0.10 SE $n = 8$	0.00	4.96 0.18 SD 0.06 SE $n = 8$	4.90 0.37 SD 0.15 SE $n = 6$	7.00 0.26 SD 0.15 SE $n = 3$
Hylobates syndactylus Siamang gibbon		4.90 $n = 1$	5.10 $n = 1$	4.50 $n = 1$	4.70 $n = 1$	0.00		
Hylobates lar Lar gibbon		4.60 0.36 SD 0.15 SE $n = 6$	4.73 0.31 SD 0.13 SE $n = 6$	4.87 0.40 SD 0.15 SE $n = 7$	4.56 0.42 SE 0.19 SE $n = 5$	1.95 0.31 SD 0.15 SE $n = 4$	0.00	7.15 $n = 2$
Papio hamadryas Hamadryas baboon		7.40 0.34 SD 0.17 SE $n = 4$	7.30 0.35 SD 0.16 SE $n = 5$	7.33 0.17 SD 0.08 SE $n = 4$	7.85 0.26 SD 0.13 SE $n = 4$		7.00 $n = 1$	0.00

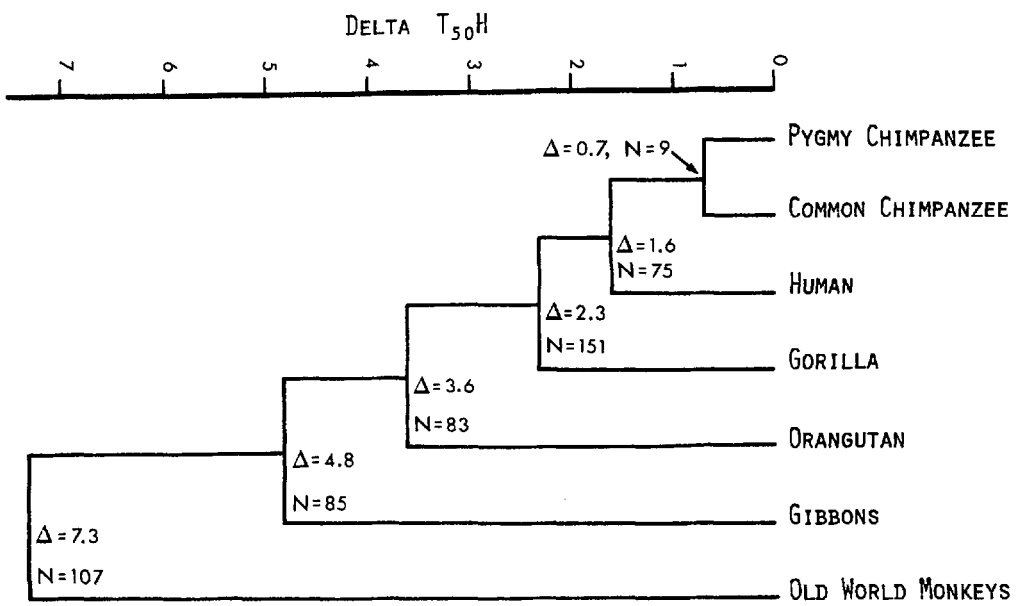

Fig. 3. Phylogeny of the hominoid primates as determined by average linkage clustering of delta $T_{50}H$ values derived from DNA–DNA hybridization

Table 4. Unfolded matrix of the 64 delta $T_{50}H$ values between hominoid tracers and the cercopithecid species whose DNAs were used only as drivers. These 64 values were not used by Felsenstein (1987) for his statistical analysis.

	Ca	An	Mm	Ms	Pn	Nl
Pan paniscus Pygmy chimpanzee	6.82 0.26 SD 0.12 SE $n = 5$		7.50 0.17 SD 0.10 SE $n = 3$			
Pan troglodytes Common chimpanzee	7.04 0.52 SD 0.23 SE $n = 5$		7.22 0.10 SD 0.05 SE $n = 4$	7.9 $n = 1$	7.8 $n = 1$	
Homo sapiens Human	7.43 0.29 SD 0.17 SE $n = 3$	7.05 $n = 2$	7.44 0.34 SD 0.15 SE $n = 5$	7.5 $n = 1$	7.6 $n = 1$	6.9 $n = 1$
Gorilla gorilla Gorilla	7.3 0.41 SD 0.14 SE $n = 9$	6.5 $n = 1$	7.35 0.14 SD 0.06 SE $n = 6$	7.45 $n = 2$	6.2 $n = 1$	
Pongo pygmaeus Orangutan	7.4 $n = 1$	7.52 0.27 SD 0.12 SE $n = 5$	6.8 $n = 1$		7.2 $n = 1$	
Hylobates syndactylus Siamang gibbon	7.2 $n = 1$					
Hylobates lar Lar gibbon			6.85 $n = 2$		7.10 $n = 2$	
Papio hamadryas Hamadryas baboon						

Ca, *Cercopithecus aethiops*; An, *Allenopithecus nigroviridis* (= *Cercopithecus nigroviridis*); Mm, *Macaca mulatta*; Ms, *Macaca silenus*; Pn, *Pygathrix nemaeus*; Nl, *Nasalis larvatus*

form average rate" of DNA evolution, which was based mainly on the large data base we had developed for birds. As we noted (Sibley and Ahlquist 1984, p. 13): "The range of dates considered from 16 to 80 MYA suggests that the regression is linear, and since the birds and the primates lie on the same regression line, it appears that the same average rate of DNA evolution occurs in both groups." Alas, this simple idea has been destroyed by facts, some of which we have provided. It is now clear that the age at first breeding, which is related to generation time, is also related to the rate of genomic evolution.

The effect of generation time on the average rate of genomic evolution has been debated since Laird et al. (1969), Kohne (1970), and Kohne et al. (1972) suggested that generation length determines the rate of accumulation of mutations in DNA, and therefore influences the average rate of genomic evolution. This proposal was based on the observation that by correcting for generation time, certain data from DNA comparisons became compatible in relation to the times of divergence of their lineages. It appeared that the average rate of molecular evolution is faster in species with short generation times, and vice versa. Some of the first evidence came from DNA hybridization comparisons between the single-copy DNAs of *Rattus* and *Mus*, which were believed to have diverged ca. 10 Myr ago, but which produced delta values of ca. 15–20, thus much greater than expected if rodents had evolved at the same average rate as primates or other large mammals. When the rat–mouse delta values were corrected for their shorter generation times in relation to the DNA hybridization data for cow–sheep, human–chimpanzee, and human–gibbon, which were also corrected for generation time, a remarkably constant rate, expressed as nucleotide substitutions per generation, was obtained. It was concluded that rodents are evolving at least 10 times as fast as the artiodactyls and the hominoids.

Kortlandt (1972) was among those advocating a generation time effect and the value of biochemical data for the reconstruction of the hominoid phylogeny. He reviewed the controversy between the molecular and fossil evidence prior to 1972 and developed an argument for the correction of the datings based on "biochemical" data in relation to different generation times. He noted (Kortlandt 1972, p. 32) that in all current studies the distance values from immunological data have

Table 5. Folded matrix of Table 2

	Pp	Pt	Hs	Gg	Po	Hy
Pan paniscus Pygmy chimpanzee						
Pan troglodytes Common chimpanzee	0.69 0.27 SD 0.09 SE $n = 9$					
Homo sapiens Human	1.64 0.18 SD 0.05 SE $n = 11$	1.63 0.24 SD 0.03 SE $n = 64$				
Gorilla gorilla Gorilla	2.37 0.23 SD 0.07 SE $n = 12$	2.21 0.21 SD 0.02 SE $n = 69$	2.27 0.24 SD 0.03 SE $n = 70$			
Pongo pygmaeus Orangutan	3.56 0.25 SD 0.07 SE $n = 13$	3.58 0.31 SD 0.07 SE $n = 21$	3.60 0.33 SD 0.07 SE $n = 25$	3.55 0.34 SD 0.07 SE $n = 24$		
Hylobates spp. Gibbons	4.87 0.52 SD 0.21 SE $n = 6$	4.82 0.40 SD 0.09 SE $n = 18$	4.76 0.34 SD 0.08 SE $n = 17$	4.69 0.30 SD 0.06 SE $n = 24$	4.83 0.34 SD 0.08 SE $n = 20$	
Cercopithecid spp. Old World monkeys	7.04 0.39 SD 0.12 SE $n = 11$	7.29 0.41 SD 0.09 SE $n = 22$	7.34 0.33 SD 0.07 SE $n = 23$	7.18 0.41 SD 0.08 SE $n = 28$	7.43 0.41 SD 0.11 SE $n = 15$	7.05 0.34 SD 0.12 SE $n = 8$

Table 6. Folded matrix of Table 3

	Pp	Pt	Hs	Gg	Po	Hy	Hl
Pan paniscus Pygmy chimpanzee							
Pan troglodytes Common chimpanzee	0.69 0.27 SD 0.09 SE $n = 9$						
Homo sapiens Human	1.64 0.18 SD 0.05 SE $n = 11$	1.63 0.24 SD 0.03 SE $n = 64$					
Gorilla gorilla Gorilla	2.37 0.23 SD 0.07 SE $n = 12$	2.21 0.21 SD 0.02 SE $n = 69$	2.27 0.24 SD 0.03 SE $n = 70$				
Pongo pygmaeus Orangutan	3.56 0.25 SD 0.07 SE $n = 13$	3.58 0.31 SD 0.07 SE $n = 21$	3.60 0.33 SD 0.07 SE $n = 25$	3.55 0.34 SD 0.07 SE $n = 24$			
Hylobates syndactylus Siamang gibbon	4.20 $n = 1$	5.13 0.32 SD 0.19 SE $n = 3$	4.70 0.46 SD 0.26 SE $n = 3$	4.54 0.22 SD 0.08 SE $n = 7$	4.93 0.19 SD 0.64 SE $n = 9$		
Hylobates lar Lar gibbon	5.00 0.45 SD 0.20 SE $n = 5$	4.76 0.39 SD 0.10 SE $n = 15$	4.78 0.33 SD 0.09 SE $n = 14$	4.75 0.32 SD 0.08 SE $n = 17$	4.74 0.42 SD 0.12 SE $n = 11$	1.95 0.31 SD 0.15 SE $n = 4$	
Papio hamadryas Hamadryas baboon	6.97 0.40 SD 0.23 SE $n = 3$	7.34 0.38 SD 0.12 SE $n = 11$	7.33 0.36 SD 0.11 SE $n = 10$	7.08 0.36 SD 0.12 SE $n = 9$	7.49 0.51 SD 0.19 SE $n = 7$		7.10 0.36 SD 0.21 SE $n = 3$

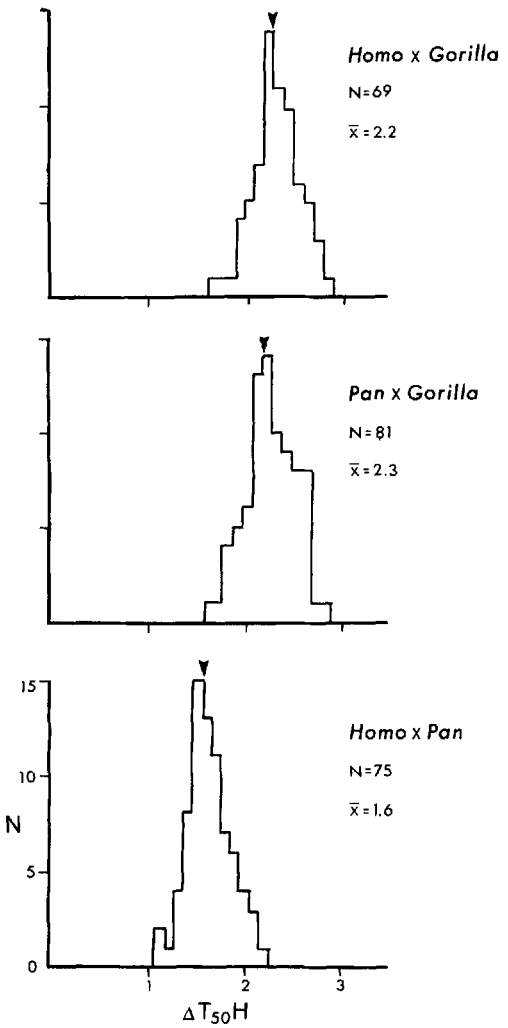

Fig. 4. Histograms of delta $T_{50}H$ values among *Homo, Pan,* and *Gorilla.* Note that the *Homo* × *Gorilla* and *Pan* × *Gorilla* distributions are essentially identical, but that they differ from the *Homo* × *Pan* distribution. Arrows = mean.

been expressed in millions of years. However, the time-table of the evolutionary process proceeds not by calendar years, but by generations. The rate at which new mutations spread through the population depends on the age at puberty and the duration of the fertile life span. Thus a phyletic rate of evolution which is constant when expressed in hundredthousands (*sic*) of generations would be decelerating when expressed in millions of years if the generation cycle time in the meantime gradually lengthens as a result of this same evolution. Whether or not this actually happened during the Mio-Pliocene we cannot know, of course. However, an indirect estimate of the prolongation of the life cycle during phylogeny may be derived from Schultz' (1969) data on extant primates.

These data indicate that, with increasing cephalization and body-size, the average duration of the generation cycle time increases at about 30% at each subsequent level of organization.

Kortlandt (1972, p. 33) proposed that

> we must convert a time-table based upon a supposedly constant rate of evolutionary change *per calendar-unit* into a time-table based upon a supposedly constant rate of evolutionary change *per generation-unit.* This can be done by postulating that the generation cycle time has steadily increased by $\sqrt[30]{1.3^3}$ per million years, while the total number of generations over the past 30 million years has remained the same. That is, we have to assume that the biochemical "clock" is slowing down by 2.65% per million years.

By applying his correction to the hominoids, Kortlandt (1972, p. 33) dated the Old World monkey branch at 40 Myr ago, the gibbon branch at 30 Myr ago, the orangutan branch at 24 Myr ago, and the trichotomy among gorilla, chimpanzees, and human at 12 Myr ago.

An effect of generation time on the rate of molecular evolution was widely accepted, although doubted by some (Sarich and Wilson 1973; see Wilson et al. 1977, p. 592, for a review). Kimura (1983, pp. 81, 246–248, 310) discussed the problem and, although favoring rate constancy per year, found reasons to suggest that generation time may have an effect on rates of molecular evolution. Although Benveniste (1985) saw no evidence of a generation time effect in certain DNA comparisons among primates, he suggested (p. 377) that "the key parameter in generation time may be the number of cell replications per year in the germ line instead of gestation lengths. . . . The rates of molecular evolution may also not always be constant."

Wu and Li (1985) found rate differences between rodents and other mammals that they attributed to a generation time effect. They compared the base sequences of 11 homologous genes from *Mus, Rattus,* and *Homo* with those of one or more other mammals (pig, cow, goat, dog), which were used as reference species in relative rate tests, which avoid assumptions about divergence times. Wu and Li (1985) found that the coding regions of the 11 genes from the two rodents are evolving much faster than those of humans, and they ascribed the difference to the shorter generation times in the mouse and rat. "The ratio of the number of nucleotide substitutions in the rodent lineage to that in the human lineage is 2.0 for synonymous substitutions and 1.3 for nonsynonymous substitutions. Rodents also evolve faster in the 5′ and 3′ untranslated regions of five different mRNAs; the ratios are 2.6 and 3.1 respectively" (Wu and Li 1985, p. 1741).

Like Benveniste (1985), Wu and Li (1985, p. 1744) proposed that the generation time effect "depends more on the number of DNA replications or cell cycles per unit time in the germ line than on the number of generations per unit time." They concluded that DNA–DNA hybridization studies are able to detect the effect because the technique measures the average rate across the genomes, and thereby detects many weakly constrained regions such as

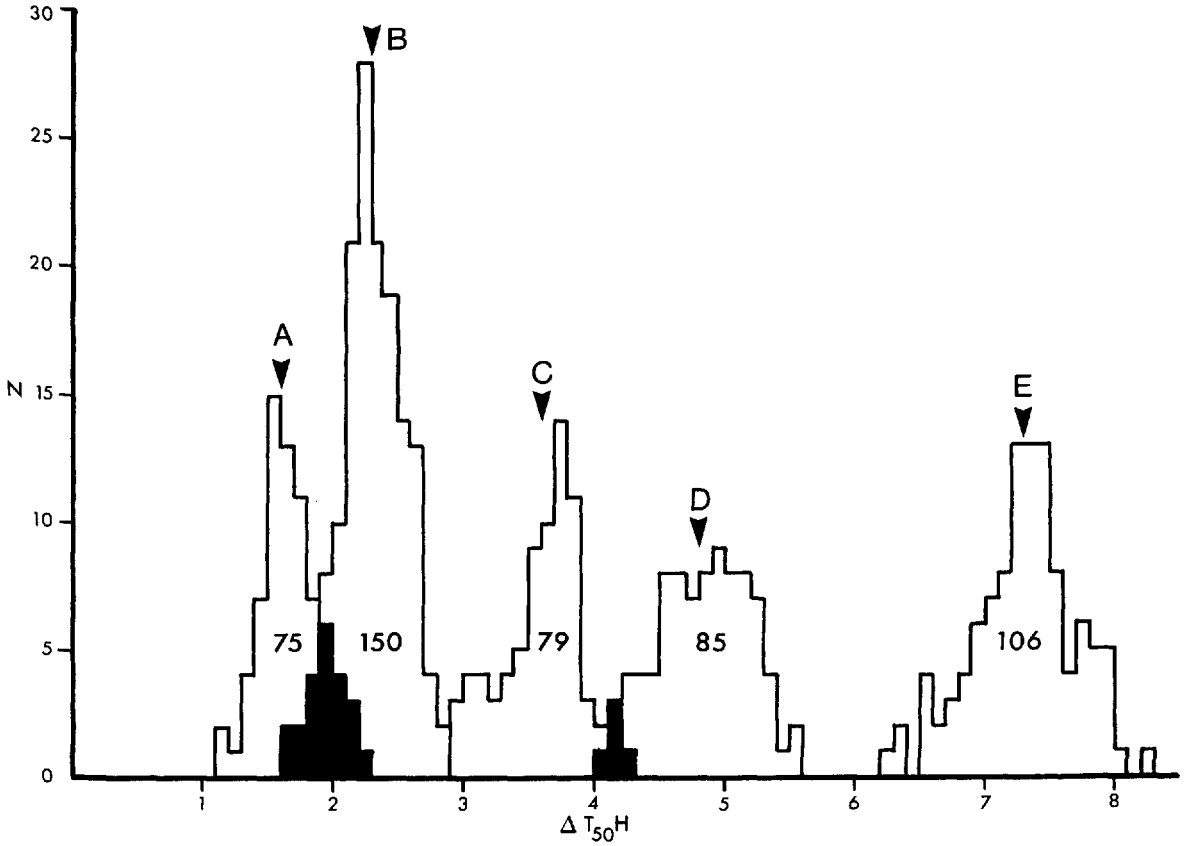

Fig. 5. Frequency distributions of actual DNA–DNA hybrids for the data depicted in Fig. 3. **A** All human × chimpanzee comparisons. **B** All human + chimpanzee × gorilla comparisons. **C** All orangutan × human + chimpanzee + gorilla comparisons. **D** All gibbon × human + chimpanzee + gorilla + orangutan comparisons. **E** Comparisons between species of the Cercopithecidae and species of the Hominoidea. Arrows = mean.

synonymous sites and introns, i.e., neutral, or near-neutral, mutations. Studies using single proteins, which are coded for by structural genes that are under more severe selection, would not be expected to show such a clear generation time effect. This may explain why Sarich and Wilson (1973) and Wilson et al. (1977) did not see the effect in their immunological comparisons of such proteins as albumin and transferrin.

In several papers we have suggested that there is a uniform average rate (UAR) of genomic evolution in birds (e.g., Sibley and Ahlquist 1983, 1985). The evidence for this hypothesis came from our DNA hybridization studies of birds, and from the similar average genomic rates (AGRs) we estimated for the hominoids (Sibley and Ahlquist 1984). Most groups of passerines, and many nonpasserine lineages, seem to be evolving at the same AGR, within the limits of experimental error. However, recently we have discovered that several groups of nonpasserine birds (e.g., ratites, loons, albatrosses, penguins, cranes, herons, flamingos, ibises, pelicans, storks, bustards, birds of prey, parrots, and others) have AGRs slower than those of the passerines and other nonpasserines. Furthermore the amount of the slowdown appears to be proportional to age at first breeding. The number of bird species having delayed maturity is approximately 1300; thus about 14% of the 9000 living species may be expected to show a slowdown compared with species that begin to breed at 1 year of age.

Conversely, in a few taxa, for example the estrildine finches (Passeridae: Estrildinae), which begin to breed before 1 year of age and may breed more than once per year thereafter, a faster AGR has been found than for those that breed first at the age of 1 year.

Sibley and Ahlquist (1983) reported the results of experiments that seemed to show that generation time has no effect on the AGR, but the design of these experiments was flawed because the reference species were also birds with delayed maturity (heron, plover); thus the branches in relative rate tests were approximately equal, although they show the effect to a small degree. These experiments should be ignored in view of the currently available evidence.

Britten (1986) reviewed the evidence for different rates of DNA sequence evolution in different taxonomic groups and concluded (p. 1393) that "examination of available measurements shows that rates of DNA change of different phylogenetic groups

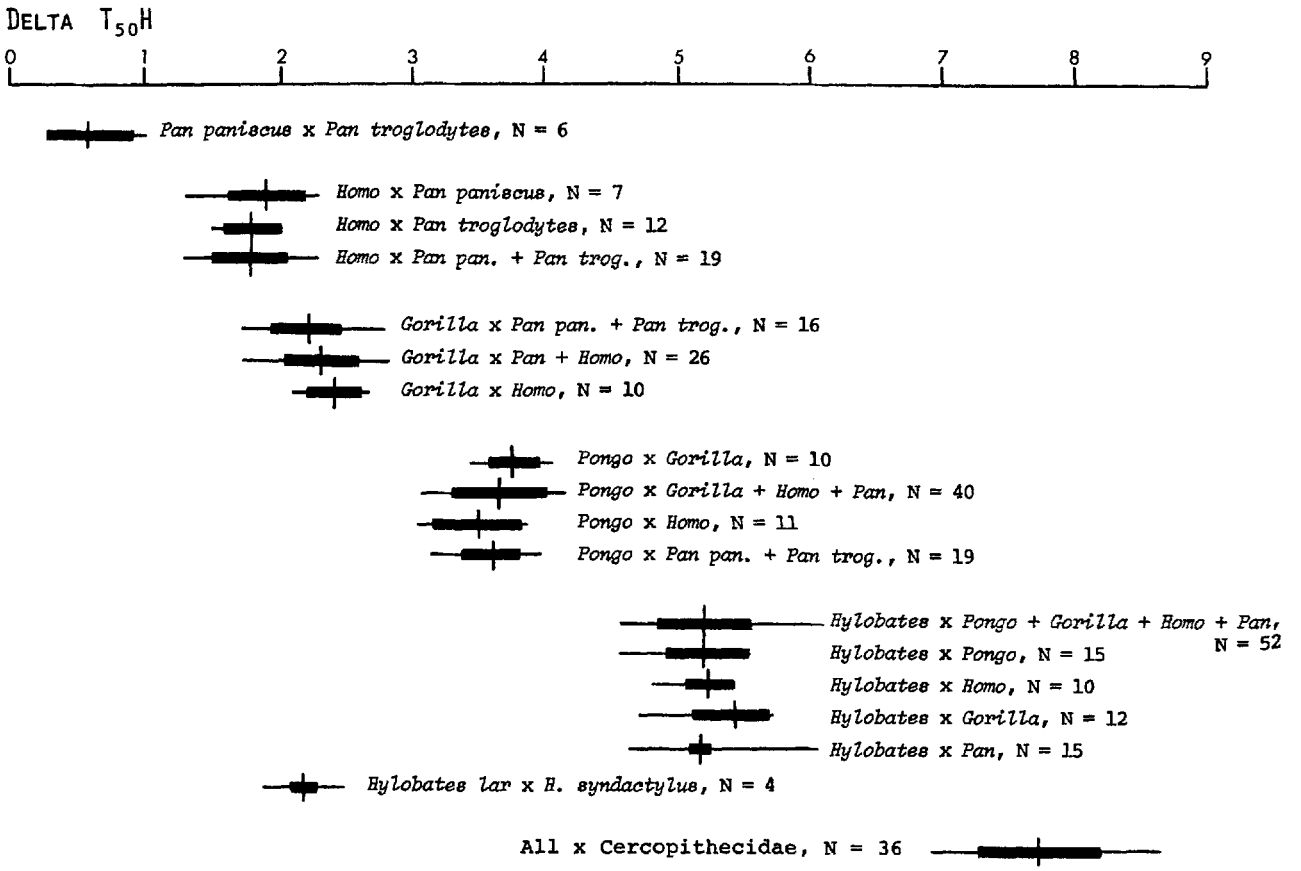

Fig. 6. Comparisons of delta $T_{50}H$ values among the hominoid taxa, and between hominoids and cercopithecids. Vertical lines = means; solid black rectangles = standard deviations; horizontal lines = ranges of delta values.

differ by a factor of 5." He noted that the slowest rates occur in the hominoids "and some bird lineages, while faster rates are seen in rodents, sea urchins, and drosophila." Britten suggested that the differences in rates "is probably due to evolutionary variation and selection of biochemical mechanisms such as DNA replication or repair."

Catzeflis et al. (1987) confirmed the observations of previous investigators who concluded that rodents are evolving much faster than primates and other large mammals with longer generation times. The average genomic rate of DNA evolution in muroid rodents was calculated as 10 times as fast as in the hominoids, thus a delta $T_{50}H$ of 1.0 = 0.40 Myr, in contrast to the estimate by Sibley and Ahlquist (1984) of delta 1.0 = 3.5–4.3 Myr in the hominoids. The average rate of change in muroid rodents is ca. 2.5%/Myr; in hominoids it is ca. 0.24%/Myr.

We conclude that the average genomic rate of evolution differs in different lineages of animals and must be taken into account when calculating divergence times from DNA–DNA hybridization data. It may be that birds show the effect so clearly because most species begin to breed at 1 year of age and breed annually thereafter. The exceptions to this pattern of breeding provided the evidence that delayed maturity is correlated with the average rate of genomic evolution. The tube-nosed seabirds (Procellariidae), including the albatrosses, petrels, diving petrels, shearwaters, fulmars, and storm petrels, provide an illustration of the relationship between the AGR and delayed maturity.

The procellariids are morphologically uniform, and their monophyly is not in question. DNA comparisons among the procellariids also indicate that the members of this group are each others' closest relatives. However, DNA–DNA distance measurements from members of other groups to the different subgroups of the procellariids differ considerably.

We have made comparisons between the procellariids and labeled species in many outgroups, some as distant as a coly (*Urocolius indicus*: Coliiformes), a gallinule (*Porphyrio porphyrio*: Gruiformes), and a francolin (*Francolinus natalensis*: Galliformes), and all give the same relative differences.

The procedure for determining the amount of shortening of branch lengths is as follows:

1) Make comparisons to several outgroups; it does not matter which ones; it is only necessary to show that the distances to outgroups are larger than any distance between two procellariids (Fitch 1986).

2) In relative rate tests, it is clear that the dis-

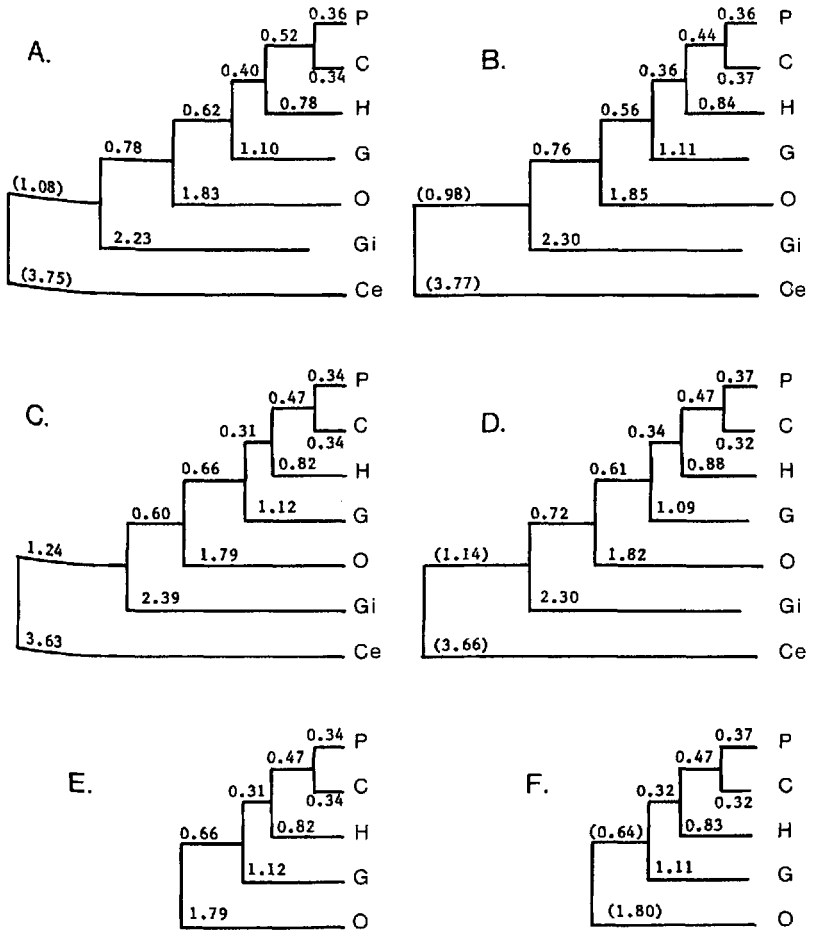

Fig. 7. Phylogenetic trees based on different methods of clustering. **A** Distance Wagner method. **B** Distance Wagner analysis, provided by Dr. Paul A. DeBenedictis, using a least squares fit of distance measures to the Steiner tree topology generated by Daniel Faith's modification of the distance Wagner algorithm; folded matrix, unweighted for sample size. **C** PHYLIP, clock assumed; folded matrix, weighted for number of values, negative branch lengths allowed, same taxa and data as for A and B. **D** Same as C, but clock not assumed. **E** Five highest taxa only used; PHYLIP, clock assumed; unfolded matrix, weighted for number of values, negative branch lengths allowed. **F** Same as E, except clock not assumed.

tances from the outgroups to the different subgroups of procellariids differ and that the differences are correlated with the age at first breeding of the procellariids. Thus, the albatrosses have the shortest branches, shearwaters next shortest, storm petrels next, and diving petrels last.

3) By using the branch length of a species that breeds at the age of 1 year as a standard, it is possible to correct the short branches so that all are equal in length. The amount of the correction is proportional to the age at first breeding.

4) This principle applies to all groups of birds.

The relationship between age at first breeding and generation time is illustrated by an example kindly provided by Dr. Bertram G. Murray, Jr. (personal communication). Using data from studies of the Prairie Warbler (*Dendroica discolor*) (Nolan 1978) and the Northern Gannet (*Sula bassanus*) (Nelson 1978), Murray developed life history tables and made the following calculations.

In the Prairie Warbler the age at first breeding is 1 year and the generation time is 2.76 years, whereas in the Northern Gannet the average age at first breeding is 5 years, and the generation time is 12.79 years. In these two species the ratio of generation time to age at first breeding is about the same, 2.76 and 2.56. The age at first breeding is an obvious concept; generation time is the time required for the population to turn over, or replace itself, weighted for age-specific contributions. Murray made certain simplifying assumptions, and concluded that generation time is the most useful measure, but one difficult to obtain for many species because the data are not available. It is probably safe to use the age at first breeding as a measure of differences in the rate of population turnover. From the two examples cited above it is not certain whether the similarity in the ratios between generation time and age at first breeding is a general constant or a coincidence. However, if the average age at first breeding for the human lineage during the past ca. 8 Myr is ca. 13 years, and the generation time is ca. 30 years, the ratio is 2.31, not far from the 2.76 and 2.56 for the two species of birds. It seems clear that several demographic factors, including longevity, number of offspring, spacing of breeding periods, ability to feed self and young, etc., are correlated with the age at first breeding.

This raises the question of the age at first breeding in the hominoids. Table 7 presents the data for some species of primates. Jolly (1985) gave the age at menarche for female chimpanzees as ca. 13 years,

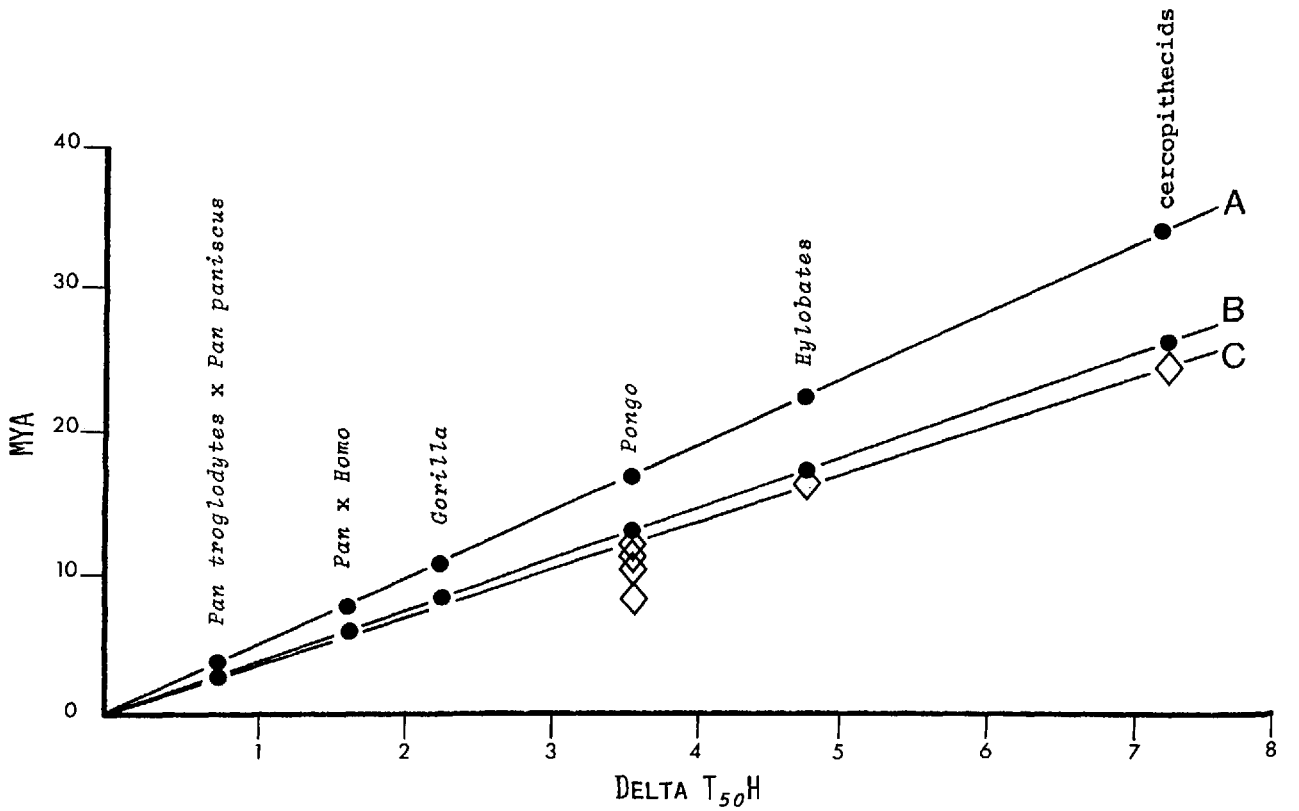

Fig. 8. Relationships between delta $T_{50}H$ values and absolute time. Solid circles = branch nodes based on DNA hybridization data (Fig. 2); open diamonds = dated fossils. Regression A: delta 1.0 = 4.76 Myr, assuming the orangutan lineage branched 17 Myr ago. Regression B: delta 1.0 = 3.64 Myr, assuming the orangutan lineage branched 13 Myr ago. Regression C: delta 1.0 = 3.42 Myr, assuming the cercopithecid lineage branched 25 Myr ago.

and for humans 11–13 years if well nourished but older if poorly nourished. Well-fed captive rhesus monkeys (*Macaca*) reach menarche at 3–5 years but in the wild at 6–8 years. Other factors, including rank in the social hierarchy, also affect the age at menarche and the age at first breeding.

The most accurate data for the common chimpanzee are probably those of Goodall (1986, p. 84) who noted that males are 12–13 years old before they begin to produce sperm. Females are 10–11 years old at first mating, but menarche occurs at 11–12 years, and the first birth at ca. 12–13 years. These ages must be close to the averages for the human lineage over the span of time since its separation from the chimpanzee and gorilla lineages. Thus, although the average age at first breeding is likely to remain uncertain, it seems probable that it is about the same for the lineages leading to humans, chimpanzees, and gorillas. Therefore, we should not expect to find significant differences in the average rates of genomic evolution among these three groups.

The delta $T_{50}H$ for the hominoid–cercopithecoid divergence is 7.2, which equates (roughly) with a 7.2% base pair mismatch between the single-copy genomes of the two lineages. Thus, the average rate of divergence since the hominoids branched from the cercopithecoids, ca. 25–34 Myr ago, has been between 0.21%/Myr and 0.29%/Myr. If 30 Myr ago is a reasonable estimate for the divergence date, the average rate = 0.24%/Myr. This is only one-tenth the rate in the muroid rodents (Catzeflis et al. 1987), which begin breeding at the age of ca. 2–3 months.

From Table 7 and the discussion above, it seems that the average age at first breeding in the living hominoids is ca. 10 years and in the living cercopithecoids ca. 5 years. Although the averages over the past 30 Myr along each branch probably differ from these values, we will use them to speculate, as follows.

Because the age at first breeding in the cercopithecoids is half as long as that in the hominoids, we assign one-third of 7.2% (= 2.4%) to the hominoid lineage, two-thirds of 7.2% (= 4.8%) to the cercopithecoid lineage, and use 30 Myr ago as the divergence time between them. Thus, the hominoid lineage has evolved at an average rate of 2.4/30 = 0.08%/Myr, and the cercopithecoid lineage has evolved at an average rate of 0.16%/Myr.

These values may be translated into the approximate number of base substitutions by assuming that the single-copy genome of the anthropoids contains ca. 2×10^9 base pairs. Thus, 7.2% of $2 \times$

Table 7. Ages at first breeding for some primate taxa

Taxa	Age at first breeding	Source
Galago demidovii, galago	ca. 8–9 mo	Macdonald 1984
Perodicticus, potto	ca. 1 yr	Macdonald 1984
Tarsius bancanus, tarsier	ca. 1 yr	Macdonald 1984
Lemur, lemur	2 yr	Napier and Napier 1985
Saimiri, squirrel monkey	2–3 yr	Napier and Napier 1985
Macaca, macaque	6+ yr	Napier and Napier 1985
Macaca, macaque	5–6 yr	Richard 1985
Cercopithecus, vervet	3–7 yr	Macdonald 1984
Papio, baboon	4–8 yr	Macdonald 1984
Hylobates, gibbon	6+ yr	Richard 1985
Hylobates, gibbon	7–8 yr	Napier and Napier 1985
Pongo, orangutan	6–7 yr	Napier and Napier 1985
Pan troglodytes, chimpanzee	12–13 yr	Goodall 1986
Pan troglodytes, chimpanzee	13 yr	Jolly 1985
Gorilla, gorilla	8–10 yr	Napier and Napier 1985
Homo sapiens, human	14 yr	Napier and Napier 1985
Homo sapiens, human	11–13 yr	Jolly 1985

$10^9 = 144 \times 10^6$ for the total base substitutions along both lineages. One-third of this number ($= 48 \times 10^6$) occurred along the hominoid branch and two-thirds ($= 96 \times 10^6$) along the cercopithecoid branch. These values translate into 1.6×10^6 base substitutions per million years, or 1.6/year for the hominoids and 3.2/year for the cercopithecoids.

These calculations are speculative and may be grossly in error, but they suggest that it may be possible to arrive at fairly accurate values when the data become more accurate.

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