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10. The last two terms of Eq. 1 are important if determining the optimal dip of the thrust fault. Ignoring them results in the incorrect conclusion that the optimal dip for the thrust is 90° (*2*).
11. Alternatively we can express ψ in terms of R and Δ: $\sin\psi = R(\cot^2\Delta + R^2 \sin^2\Delta)^{-1/2}$
12. Alternatively, $\tan\phi = [-1 \pm (1 + 4S(1 - S) \tan^2\zeta)^{1/2}]/2\tan\zeta\ (1 - S)$ when $S \neq 1$ and $\theta = \zeta$ when $S = 1$.
13. The ratio works out to $\tau_s/\tau_t = [\sin^2\Delta + \sin^2 2\Delta (\sin^2\phi - S)^2/\sin^2 2\phi]^{-1/2}$, which reduces to R using Eq. 6.
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Rejection of the "Flying Primate" Hypothesis by Phylogenetic Evidence from the ϵ-Globin Gene

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Whether the bat suborder Megachiroptera (megabats) is most closely related to the other suborder of bats, Microchiroptera (microbats), or whether Megachiroptera is the sister group of order Primates has been an issue of much debate. Should all bats be classified into a monophyletic order (Chiroptera) or do bats have diphyletic origins, and are the megabats actually "flying primates"? These questions were addressed by phylogenetic analysis of ϵ-globin gene sequences from a number of primates and other eutherian mammals. Results of parsimony analysis not only support bat monophyly, but the strength of Chiroptera grouping is comparable to that supporting the monophyly of the prosimian primate suborder Strepsirhini (galago and lemur). Furthermore, 39 derived nucleotide sequence changes are uniquely shared by the megabat (*Cynopterus sphinx*) and microbat (*Megaderma lyra*) versus three commonly shared by the megabat, primates, and Dermoptera or flying lemur (*Cynocephalus variegatus*), and only two shared by either megabat and primates, or by megabat and flying lemur.

Debate over chiropteran origins began as early as the 1700s when Linnaeus (*1*) first placed the bats with the order Primates in mammalian taxonomy. The classical hypothesis (*2*)—a monophyletic grouping of megabats, suborder Megachiroptera, with microbats, suborder Microchiroptera, in order Chiroptera—is based on an array of morphological traits including common wing structure, cranial vascular features, and fetal membranes (*3*). Furthermore, Novacek proposed that Dermoptera (flying lemur) and Chiroptera are most closely related to each other and that they should be included in a superorder Archonta with Primates and Scandentia (tree shrews) (*4*).

In contrast, the diphyly of bats or "flying primate" hypothesis advocates that flight evolved twice in mammals, once in the early descent of Microchiroptera, and again later in the lineage leading to the Megachiroptera from a common lineage shared with Primates and Dermoptera (*5, 6*). Pettigrew's analysis of neural anatomy in the visual and motor pathways led him to conclude that the brains of Primates, Dermoptera, and Megachiroptera share important derived features that are absent in Microchiroptera (*5, 7*). Additional evidence for the diphyly of bats is the presence of a glans penis, found only in Dermoptera, Megachiroptera, and Primates (*8*).

Elucidating the true phylogeny of Chiroptera has relevance to the origins of Primates, Dermoptera, and Scandentia. It also provides a framework for exploring evolutionary processes, because both megabats and microbats share similar wing structures, whereas megabats and primates share similar neural pathways. Thus, one set of these shared traits represents homoplasy (superficial similarity due to convergence or reversal). Morphological evidence has failed to define accurate phylogenetic relationships between megabats, microbats, and other eutherian mammals (*3, 6, 7*).

The evidence on bat origins from earlier molecular studies have been inconclusive as well (*9, 10*). In seeking more definitive molecular evidence, we have analyzed a data set of DNA sequences representing the ϵ-globin gene from 11 primates, flying lemur, tree shrew, megabat, microbat, rabbit, and goat. Our study provides molecular evidence from a nuclear gene directed at answering whether megabats share a more recent common ancestor with primates or microbats.

The ϵ-globin gene in mammals is the 5′-most member of the β-globin gene cluster that arose from a series of tandem duplications, the first of which occurred about 200 million years ago (Ma) and led to the embryonically expressed proto-ϵ gene and the postnatally expressed proto-β gene. By the time of the first placental mammals (90 to 100 Ma), further tandem duplications resulted in five gene loci linked in the order 5′-ϵ-γ-η-δ-β-3′ (*11*). In placental mammals, the ϵ gene has been much less prone to undergo further tandem duplications than have the other β-type globin genes. Therefore, it is well suited for the study of phylogenetic relationships because the problem of comparing paralogous genes (genes derived from duplication events) is largely avoided. In addition, the majority of the sequence data is noncoding, hence it is not under selective constraints that may result in functional homoplasies.

Sequence analysis encompassed 17 orthologous genes (genes derived from speciation events). The data set consisted of sequences from previously published data, lambda library subclones, and polymerase chain reaction (PCR)–generated clones

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(*12*). Six sequences are represented by a 4.1-kb Eco RI subcloned fragment (Fig. 1). Four of these, human (*13*), orangutan (*14*), galago (*15*), and rabbit (*16*), were sequenced previously, whereas two others, gibbon and capuchin monkey, were sequenced for this study. Tarsier (*17*), lemur (*18*), and goat (*19*) were also previously published lambda-derived sequences spanning 1.8 kb of the gene proper beginning just upstream of the promoter and ending about 100 bp downstream of the polyadenylation signal. Four other sequences each

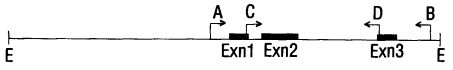

Fig. 1. Map of the ϵ-globin gene region. Primers C and D were used when amplification reactions failed from primers A and B because of mismatches to DNA from species that are more distantly related to the primates. Three PCR-derived clones from three independent PCR reactions were sequenced from each species. Where sequence discrepancies existed between clones, more than three cloned inserts were sequenced and the consensus was used for comparative analysis.

1.8 kb in length from common chimpanzee, pygmy chimpanzee, gorilla, and rhesus monkey were amplified by PCR from primers A and B (Fig. 1). Four more PCR-generated sequences, each 1.2 kb in length, from flying lemur, tree shrew, megabat, and microbat were amplified from primers C and D (Fig. 1). Initial sequence alignments were obtained by the pairwise alignment algorithm of Smith and Waterman (*20*) as modified by Goodman *et al.* (*11*), and alignment for all 17 sequences was completed by hand. Gaps were inserted where they maximized the alignment by increasing similarities that could be attributed to common ancestry.

Evolutionary reconstruction began with pairwise sequence comparisons over shared nucleotide positions to obtain estimates of divergences. The divergence values in Table 1 compare the major primate clades [the anthropoids, the strepsirhines (galago and brown lemur), and the tarsier (the prosimian primate that may be closest cladistically to anthropoids) (*17*)], flying lemur, tree shrew, megabat, microbat, rabbit, and goat. Flying lemur diverges less from the primates (23.5% to 28.7%) than it does from the nonprimates (32.9% to 38.8%), indicating that Dermoptera may be the sister taxon of Primates. This finding is consistent with recent paleontological evidence (*21*). The two bats diverge less from each other than either does from any other species, indicating a recent common ancestry. Furthermore, the divergence value for the two bats (23.5%) is essentially equal to divergence values observed within the primates themselves (24.5% to 28.9%), which is congruent with the hypothesis that megabats and microbats are monophyletic. In contrast, interordinal divergence values range between 33% (flying lemur:tree shrew) and 47.8% (rabbit:goat).

This divergence matrix was then used to construct trees by the UPGMA method (*22*) and the neighbor-joining (NJ) method (*23*). The UPGMA tree placed flying lemur among the primates, joining it to the anthropoids, followed successively by strepsirhines, tarsier, tree shrew, rabbit, a bat clade, and goat. The NJ tree grouped all the primates together; in succession, these were then joined by flying lemur, a tree shrew–rabbit clade, a bat clade, and goat. These topologies were used as input trees with the full 4.2-kb sequence alignment to generate a number of alternative branching arrangements by two branch-swapping parsimony programs, a global swap program PTRFC (*24*) and an all trees program PTRALL (*25*).

The lowest length (LL) tree joins rabbit to tree shrew which then joins the primates, the primate–tree shrew–rabbit clade is next joined by flying lemur, then by the bat clade, and by goat (Fig. 2). The grouping of tree shrew and rabbit agrees with a previous study in which amino acid sequences of α and β hemoglobin were used (*10, 25*). The "all trees" program was used to assess how strongly these ϵ sequences support the monophyly of bats as well as other phylogenetic relationships. In the LL tree (Fig. 2), the numbers within circles on each stem represent the minimum number of nucleotide substitutions that must be added to the maximum parsimony score to break up that clade. Note that the number of nucleotide substitutions required to break up the bats (31) is comparable to that required to break up the strepsirhines (37). The other strongly supported monophyletic groupings are the catarrhines (27), the anthropoids (65), and the grouping of primates, flying lemur, tree shrew, and rabbit (23).

The most parsimonious tree was then compared with opposing hypotheses. Figure 3A shows the LL tree and its score, and Fig. 3, B to E, shows alternative hypotheses of Primate-Dermoptera-Chiroptera relationships tested by parsimony

Table 1. Sequence divergence values between primate and nonprimate species. Divergence values were calculated from the 1.2-kb region where all species were represented over all positions. Values represent actual divergence corrected for superimposed mutations (*28*). For each pairwise comparison, average divergence values are given for the eight anthropoid primates and two strepshirhine primates (galago and lemur).

Species	Sequence divergence values								
9. *C. hirus*									—
8. *O. cuniculus*								—	.47843
7. *M. lyra*							—	.46788	.40968
6. *C. sphinx*						—	.23532	.43371	.36553
5. *T. glis*					—	.38630	.41426	.40359	.47023
4. *C. variegatus*				—	.32909	.33712	.35254	.33648	.38756
3. Strepsirhine			—	.25853	.34569	.33644	.37390	.34401	.39509
2. *T. syrichta*		—	.28857	.28645	.36912	.41781	.41526	.35889	.43112
1. Anthropoid	—	.26774	.24534	.23513	.33184	.32294	.34394	.33819	.38989
	1	2	3	4	5	6	7	8	9

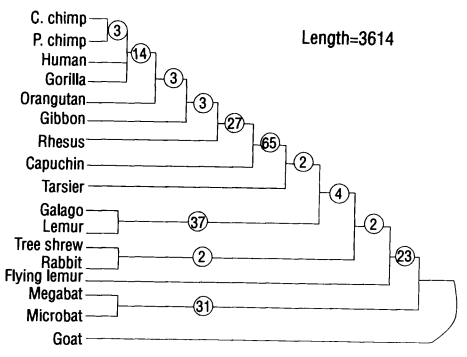

Fig. 2. Maximum parsimony tree with strength of grouping results based on a number of eight-branch trees. First the global swap (PTRFC) program was used with the full sequence alignment to generate the most parsimonious or lowest-length (LL) tree—that is, the tree with the fewest number of hypothesized evolutionary changes. This LL tree was then submitted to the "all trees" (PTRALL) program to determine whether a lower score could be obtained. For PTRALL, subtrees can be designated within which no branch swaps are allowed, producing a starting tree with eight terminal taxa. Therefore, a terminal taxon can either be an exterior node (one of 17 extant species) or a designated subtree. PTRALL examines all possible 10,395 unrooted trees for these eight terminal taxa. Further exhaustive branch swaps by the PTRALL program, with starting trees having different sets of eight terminal taxa, failed to lower the score found by PTRFC. Due to limited data and a very high degree of sequence similarity between chimpanzee, human, and gorilla, no resolution of the trichotomy was observed in this analysis.

and the resulting scores. Novacek's hypothesis of Chiroptera-Dermoptera monophyly in a superorder Archonta, which also contains Primates joined to Scandentia (Fig. 3B), requires postulating 52 changes more than the most parsimonious tree. Three of Pettigrew's "flying primate" hypotheses or the diphyly of bats (Fig. 3, C to E), each require greater than 100 postulated changes over the LL tree. For Pettigrew's trees, alternative topologies were tested with respect to the position of lagomorph and microbats and for each the lowest scoring topology is presented.

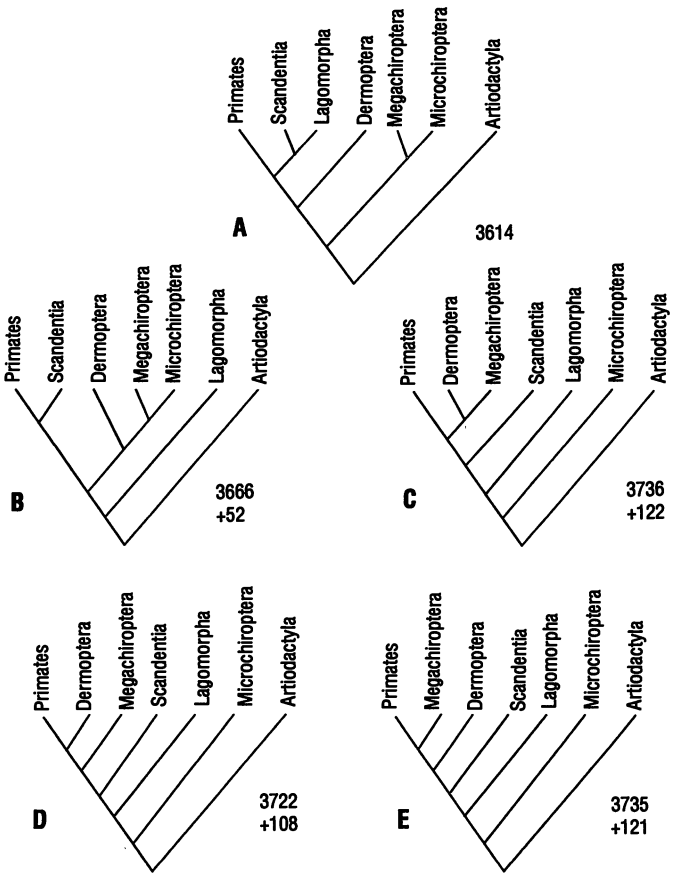

Fig. 3. Comparison of the most parsimonious tree versus alternative hypothetical topologies. Groups represented are order Primates, tree shrew (family Scandentia), flying lemur (order Dermoptera), megabat (suborder Megachiroptera), microbat (suborder Microchiroptera), rabbit (order Lagamorpha), and goat (order Artiodactyla). (**A**) The most parsimonious tree and its score; (**B**) Novacek's hypothesis with its parsimony score and the difference between that tree's score and the lowest score; and (**C** to **E**) three of Pettigrew's hypotheses, scores, and differences.

Table 2. Positions with synapomorphic sequence characters supporting various Megachiroptera groupings. Abbreviations are listed in (*12*) and Primates = Pri. The positions in bold type denote where there are no convergent nucleotides between the bats and any other species. To determine whether the number of positions supporting one hypothetical tree over another is significant, we applied the winning-sites method (*29*). The winning-sites test determined whether 39 supportive positions were significantly greater than the 11 or 3 positions supporting alternative hypotheses. In comparing Csp:Mly (39 positions) versus Csp:Chi (11) by a binomial distribution, $p = 4.51 \times 10^{-5}$. In comparing Csp:Mly (39) versus Csp:Pri/Cva (3), $p = 2.82 \times 10^{-9}$. Therefore, support for the monophyletic grouping of the bats is significantly greater than either competing alternative.

Sequence positions						
Csp:Mly			Csp:Cva	Csp:Pri/Cva	Csp:Pri	Csp:Chi
2388	**2846**	**3539–43**	2560	2830	2861	2390
2396	**2893**	**3583–87**	2821	3213	2891	2415
2400	**3219**	3625		3614		2448
2405	3231	**3626**				2615
2416	**3441**	**3627**				2629
2477	**3476**	3628				2872
2731	**3494**	**3629**				3199
2838	3495	3630				3396
2779	**3496**	3637				3685
2782	**3502–04**	**3688–89**				3721–22
2811–14	**3511**	**3743**				3750
2815	**3523**	3763				
2816	3536	3830				

Over the 1200 nucleotide positions representing the megabat, microbat, tree shrew, and flying lemur, there are 39 different, single or contiguous positions where the sequences uniquely group the two bats (*C. sphinx* and *M. lyra*) (Table 2). It is worth noting that just one of these putative synapomorphic positions or shared derived nucleotides, site 2477, is within exon 2, which is the only coding region represented in these species. This would argue against any selected convergences accounting for the similarity in the two bat sequences. In contrast, only two positions support either a megabat–flying lemur clade or a megabat-primate clade and only three support a megabat–primate–flying lemur clade. Because the first tree to break up Chiroptera joined the megabat to goat, the number of putative synapomorphic characters supporting that grouping was tabulated. The megabat and goat share only 11 such nucleotides (Table 2).

Additional molecular data involving different representatives of Megachiroptera and Microchiroptera also support Chiroptera monophyly. In recent studies of the mitochondrial 12S ribosomal RNA and the cytochrome oxidase I and II genes, the megabat *Pteropus capestratus* and the microbat *Brachyphylla cavernarum* were included in one analysis and the megabat *Rousettus leschenaulti* and the microbat *Phyllostomus hastatus* were included in another (*26*). Another study comparing sequences of the interphotoreceptor retinoid binding protein gene from primates and a number of nonprimate species, including the megabat *Pteropus hypomelanus* and the microbats *Tonatia bidens* and *Tonatia silvicola*, found a monophyletic Chiroptera to be the most parsimonious solution (*27*).

Pettigrew's view that microbats are not closely related to primates may be more correct than Novacek's, which places all the bats in the superorder Archonta with primates, but we find that neither megabats nor microbats are closely related to primates. Although the question of which mammalian order is the sister group of primates was not resolved in this study, evidence based on divergence values and parsimony analysis demonstrates that Megachiroptera is not the sister group of the primates and that a more likely candidate is one of flying lemur, tree shrew, or rabbit (*21*). Morphological homoplasies must have occurred during the descent of Primates and Chiroptera either in wing structure, present only in the two suborders of bats, or in some neural pathways common only to primates and megabats. According to a diphyletic hypothesis, powered flight in mammals evolved twice,

but due to anatomical constraints much homoplasy resulted. Pettigrew argues that "functionally obscure" neurological features are less apt to be under selective constraints and similarities between primates and megabats are due to recent common ancestry rather than convergence (*7*). Our ϵ-globin noncoding DNA data strongly oppose the "flying primate" view of wing convergence during the descent of two separate bat lineages, in support of the classical hypothesis of a monophyletic Chiroptera and a common origin of mammalian flight.

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Maternal-Effect Selfish Genes in Flour Beetles

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A previously unknown class of dominant, maternal-effect lethal *M* factors was found to be widespread in natural populations of the flour beetle, *Tribolium castaneum*, collected on several continents. Such factors are integrated into the host chromosomes at variable locations and show the remarkable property of self-selection by maternal-effect lethality to all hatchlings that do not inherit a copy of the factor itself. Offspring are rescued by either paternally or maternally inherited copies. The *M*-bearing chromosome is thereby perpetuated at the expense of its non-*M* homolog. *M* factors that map to different regions of the genome do not rescue one another's maternal-effect lethality. Factors expressing these properties are predicted to spread in a population, even in the absence of any additional selective advantage. Similar factors also occur in the related species *T. confusum*.

Much of an animal's genome may be composed of parasitic or "selfish" DNA that serves no immediate function for the host but rather exploits the host for its own propagation (*1*). Parasitic DNA may thrive and spread by replicative transposition, by segregation distortion, by mechanisms involving supernumerary chromosomes (*2*) or by other non-Mendelian mechanisms. We report a novel mechanism by which a selfish gene (or gene complex) may facilitate its own propagation at the expense of its unselfish homolog. We show evidence for the widespread distribution in nature of a chromosomally integrated factor that confers maternal-effect lethality to all progeny that do not inherit a copy of the factor itself. To our knowledge similar mechanisms are unknown in the animal kingdom.

In a screen for hybrid dysgenesis (*3*) between geographically diverse strains of *T. castaneum*, we found numerous cases of reciprocal hybrid female semisterility. One example of such bidirectional, female-specific semisterility, observed in hybrids between strains collected in Singapore (SP) and the United States (US) (*4*), is documented in Fig. 1A. Crosses within each strain and reciprocal crosses between strains are fertile. F_1 hybrid males from either interstrain cross are fertile when backcrossed to females from either parental strain. F_1 hybrid females from either interstrain cross are fertile when crossed to SP males, but semisterile when crossed to US males. This semisterility is due to a preadult mortality rate of 50 to 80% among the

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