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17. J. M. Warren, G. Hirth, *Earth Planet. Sci. Lett.* **248**, 438 (2006).
18. T. V. Gerya, B. Stoeckhert, A. Perchuk, *Tectonics* **21**, 1056 (2002).
19. C. Subarya *et al.*, *Nature* **440**, 46 (2006).
20. N. I. Christensen, *Int. Geol. Rev.* **46**, 795 (2004).
21. T. Seno, *Earth Planet. Sci. Lett.* **231**, 249 (2005).
22. M. Simoes, J. P. Avouac, R. Cattin, P. Henry, *J. Geophys. Res. Solid Earth* **109**, B10402 (2004).
23. D. L. Turcotte, G. Schubert, *Geodynamics* (Cambridge Univ. Press, ed. 2, 2002).
24. S. Ide, G. C. Beroza, D. R. Shelly, T. Uchide, *Nature* **447**, 76 (2007).
25. A. M. Freed, R. Burgmann, *Nature* **430**, 548 (2004).
26. H. Ueda, M. Ohtake, H. Sato, *J. Geophys. Res. Solid Earth* **108**, 2151 (2003).
27. K. Regenauer-Lieb, D. A. Yuen, J. Branlund, *Science* **294**, 578 (2001).
28. J. A. Conder, *Phys. Earth Planet. Int.* **149**, 155 (2005).
29. P. Raterron, Y. Wu, D. J. Weidner, J. Chen, *Phys. Earth Planet. Int.* **145**, 149 (2004).
30. S. Karato, D. C. Rubie, H. Yan, *J. Geophys. Res. Solid Earth* **98**, 9761 (1993).
31. A. Dimanov, G. Dresen, *J. Geophys. Res. Solid Earth* **110**, B07203 (2005).

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A Comprehensive Phylogeny of Beetles Reveals the Evolutionary Origins of a Superradiation

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Beetles represent almost one-fourth of all described species, and knowledge about their relationships and evolution adds to our understanding of biodiversity. We performed a comprehensive phylogenetic analysis of Coleoptera inferred from three genes and nearly 1900 species, representing more than 80% of the world's recognized beetle families. We defined basal relationships in the Polyphaga supergroup, which contains over 300,000 species, and established five families as the earliest branching lineages. By dating the phylogeny, we found that the success of beetles is explained neither by exceptional net diversification rates nor by a predominant role of herbivory and the Cretaceous rise of angiosperms. Instead, the pre-Cretaceous origin of more than 100 present-day lineages suggests that beetle species richness is due to high survival of lineages and sustained diversification in a variety of niches.

The extraordinary diversity of beetles has long fascinated evolutionary biologists (*1*). The strongly sclerotized front wings defining the order Coleoptera (the beetles), which provide protection while retaining the ability of powered flight with the membranous hindwings, may be an evolutionary novelty that promoted extensive diversification (*2*). Beetles appeared around 285 million years ago (Ma) (*2*, *3*), followed by radiations of wood-boring (suborder Archostemata), predacious (Adephaga), and fungivorous (Polyphaga) lineages (*4*) present in the fossil record from the middle Triassic on (*2*, *3*). Their species richness is associated with extreme morphological, ecological, and behavioral diversity (*4*), and diversification of the most species-rich extant lineages may have been driven by co-radiations with angiosperms (*5*) and/or mammals (*6*) and/or geological and climatic change (*7*) occurring since the Cretaceous (145 to 65 Ma).

Studies of phylogenetic relationships within the Coleoptera resulted in a preliminary consensus on the classification, defining 4 suborders, 17 superfamilies, and 168 families (*8*–*10*). However, formal phylogenetic analyses of morphological characters (*11*, *12*) and more recently molecular data (*5*, *13*, *14*) have been limited to subgroups at the family or superfamily level. Because of the sheer size of the group and the complexity of morphological character systems, these analyses have not been applied to the entire order.

We compiled a three gene data matrix providing a complete taxonomic representation for all suborders, series and superfamilies; >80% of recognized families; and >60% of subfamilies (*9*, *10*), which together contain >95% of described beetle species. Sequences for the small subunit ribosomal RNA (18 *S* rRNA) were obtained for 1880 species from de novo sequencing and existing databases. Mitochondrial 16S rRNA (*rrnL*) and cytochrome oxidase subunit I (*cox1*) sequences were added for nearly half of these taxa (table S1) to create a data matrix of rapid, medium, and slowly evolving sequences. Phylogenetic analysis of the combined matrix was performed with a fragment-extension procedure for global sequence alignment followed by tree searches with fast parsimony algorithms (*15*). We tested for long-branch attraction, i.e., the spurious pairing of rapidly evolving lineages, by removing taxa terminal to long branches and assessing trees with a retention index (RI) measure of fit to the traditional classification (table S2) (*15*). The resulting parsimony tree largely agrees with the existing classification at the family and superfamily levels [on average, 95.7% of terminals assigned to a family were recovered as monophyla (table S2)], although our taxon sampling was not comprehensive in some families. Model-based Bayesian methods were applied to a 340-taxon representative subset at the subfamily level.

The trees (Figs. 1 and 2) were rooted with the neuropterid orders, the presumed sister to the Coleoptera (*16*), and recovered the major subdivisions of Adephaga [37,000 known species; posterior probability (pp) = 1.0] and Polyphaga (>300,000 species; $pp = 1.0$) as sisters to the Myxophaga (94 species) plus Archostemata (40 species) (*8*). The Adephaga was divided into two clades containing an aquatic (Hydradephaga; diving beetles and whirligig beetles; $pp = 0.90$) and a terrestrial (Geadephaga; ground beetles and tiger beetles; $pp = 1.0$) lineage, supporting a single terrestrial-to-aquatic transition in this suborder (*13*).

In the strongly supported suborder Polyphaga, five families occupied the basal nodes (Figs. 1 and 2) ($pp = 1.0$). These families include the Decliniidae; the Scirtidae, with aquatic larvae; the Derodontidae, an ecologically diverse family from global temperate zones; and the Eucinetidae and the Clambidae. These ancestral five families were previously considered basal Elateriformia (superfamily Scirtoidea), except for Derodontidae,

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which has been associated with Bostrichiformia (*9*, *10*). All five families exhibit archaic morphological features shared only with Archostemata and Adephaga (*8*, *17*). Their basal position was stable (always *pp* = 1.0) (table S3) when trees were rooted with the neuropterid orders or only with Myxophaga or Adephaga as outgroups.

All superfamilies of Polyphaga were previously grouped into five series (*4*, *9*), of which only the Scarabaeiformia (*pp* = 1.0) and the Cucujiformia (*pp* = 1.0) were strongly supported as monophyletic in this study. Staphyliniformia comprised a paraphyletic basal grade, and both Bostrichiformia and Elateriformia were polyphyletic. Relationships among the five series were poorly supported or unresolved in the consensus tree (fig. S1). Nosodendridae, usually included in Bostrichiformia near Derodontidae (*4*, *9*) but recently associated with Scirtoidea on the basis of thoracic characters (*18*), grouped instead with the nonscirtoid Elateriformia, albeit with low support (fig. S1) (*pp* = 0.59).

Within Elateriformia, the superfamilies Buprestoidea (jewel beetles; *pp* = 1.0), Dascilloidea (*pp* = 1.0), and Elateroidea (click beetles and allies; *pp* = 0.72) were supported. Our data showed that Byrrhoidea, sensu Lawrence and Newton (*9*), is paraphyletic, supporting the division of this clade (*8*) into Byrrhoidea (Byrrhidae, moss beetles; *pp* = 1.0) and Dryopoidea (riffle beetles and water pennies). The Cantharoidea (soldier beetles, fireflies, etc.) fell inside the Elateroidea, and our tree supported that bioluminescence arose repeatedly in beetles, in agreement with structural differences in luciferases (*19*). Scarabaeiformia (chafers, stag beetles, and dung beetles; *pp* = 1.0) is thought to be related to the Staphyliniformia (*4*, *14*, *20*). In our trees, it was part of an unresolved paraphyletic Staphyliniformia including the superfamilies Histeroidea (clown beetles; *pp* = 1.0); Hydrophiloidea (*pp* = 1.0), a clade of both Leiodidae and Agyrtidae (*pp* = 1.0); the Staphylinidae (rove beetles including Silphidae and carrion beetles; *pp* = 0.86); and the Hydraenidae as sister (*pp* = 0.74) to the Ptiliidae (featherwing beetles).

The hyperdiverse Cucujiformia, representing more than half of all beetles and 90 families, was strongly supported as monophyletic (Figs. 1 and 2; *pp* = 1.0). Among the seven established superfamilies, the Lymexyloidea (ship-timber beetles) was found near the base of the Tenebrionoidea (30 families; *pp* = 0.76). The Cleroidea (checkered beetles and allies) was monophyletic (*pp* = 0.70) only when including the Biphyllidae plus Byturidae (*pp* = 1.0). The latter two were formerly classified as Cucujoidea, but their association with Cleroidea is supported by genitalic characters (*11*). The Cucujoidea, comprising 34 families, was polyphyletic, but the Cerylonid series (Figs. 1 and 2 and fig. S3) (*pp* = 1.0) consisting of eight families (*21*) was monophyletic. Apart from the Sphindidae (*pp* = 1.0), the remaining cucujoid families formed a monophyletic clade (*pp* = 0.72) together with the species-rich Curculionoidea (weevils and bark beetles; *pp* = 0.73) and Chrysomeloidea (leaf beetles and longhorns).

Once the relationships among coleopteran families and superfamilies were established, we investigated the origins of beetle diversity. Diversification may be driven by feeding strategy, and we tested the hypothesis that feeding on plants (herbivory), and specifically flowering plants (angiosperms), explains the diversity of beetles (*5*). Predominantly herbivorous clades tend to contain

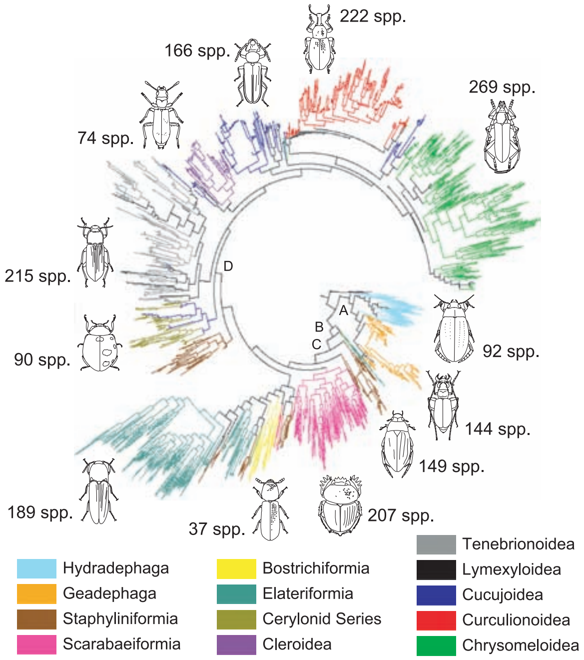

Fig. 1. One of 27 most parsimonious trees obtained from the aligned 1880-taxon matrix. The number of representatives from each major lineage analyzed (in colors) is given. Major clades are denoted by letters: A, Adephaga; B, Polyphaga; C, Polyphaga minus the ancestral five families; and D, Cucujiformia. For full details of the tree, see fig. S4.

Table 1. Comparisons of species richness between clades feeding on living plants and their sister clades with alternative feeding strategies. Restricting the comparisons to those feeding on angiosperms removes contrast 4 and adds two contrasts of angiosperm- versus gymnosperm-feeding lineages within Curculionoidea and two within Chrysomeloidea [table S4; see also (*5*)]. Plant-feeding clades include taxa feeding mainly on rotting vegetation in contrast 7 or in recently dead wood in contrast 8, but probably >70% of species in both clades are herbivorous. Excluding the last two contrasts increases the probability under a Wilcoxon test to *P* = 0.28.

	Plant-feeding	Diet	No. of species	Non–plant-feeding	Diet	No. of species
1	Byturidae	Fruits, flowers	16	Biphyllidae	Fungivorous	195
2	Languriinae	Stem borers	800	Xenoscelinae	Fungivorous, decaying vegetation	100
3	Chrysomeloidea	Herbivorous xylophagous	53,442	Nitidulidae plus Erotylid plus Cucujid series	Mostly fungivorous	7743
4	Curculionoidea	Herbivorous xylophagous	59,340	Brontinae plus Silvaninae plus Priasilphinae	Fungivorous	480
5	Epilachninae	Herbivorous	1051	Coccidulinae plus Chilocorinae plus Scymninae	Predacious	3900
6	Dascillinae	Roots	80	Rhipiceridae	Ectoparasitic on cicadas	57
7	Melolonthinae plus Orphninae plus Rutelinae plus Dynastinae	Herbivorous (and saprophagous)	16,329	Cetoniinae	Saprophagous (detritus)	4121
8	Buprestidae	Xylophagous, herbivorous, roots, leaf miners	14,000	Dryopoidea	Saprophagous, algivorous	3242

more species than nonherbivorous sister clades, but this difference was not significant [Table 1; one-tailed Wilcoxon test on contrasts in log (no. of species), $P = 0.13$] even when we distinguished between angiosperm and gymnosperm feeders ($P = 0.06$) (table S4). Similarly, of 21 significant shifts in diversification rate inferred with a robust equal rates null model (*22*, *23*), only two characterize transitions between angiosperm and gymno-

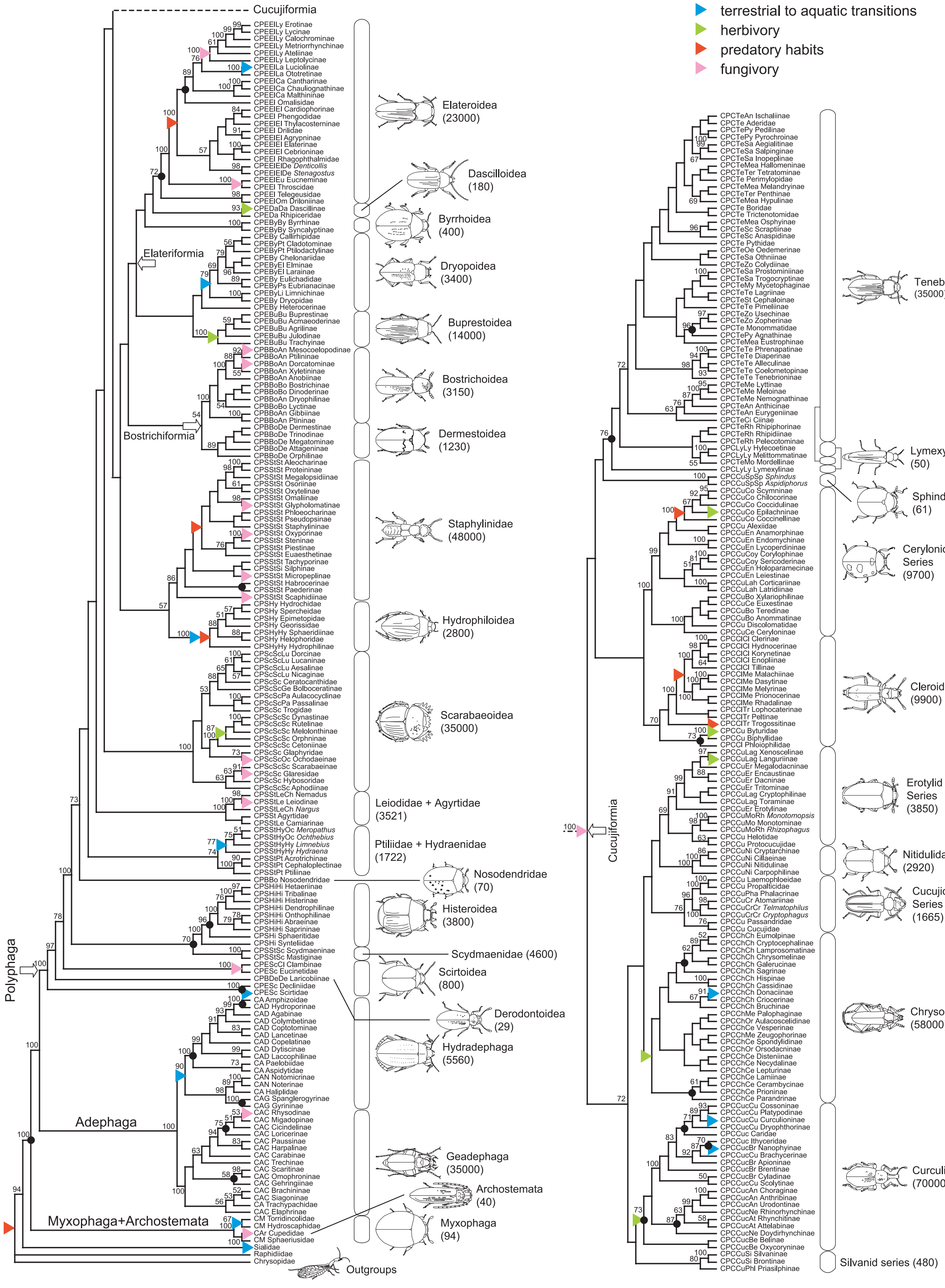

Fig. 2. The phylogeny of Coleoptera at the subfamily level. The tree was selected from the 340-taxon Bayesian analysis based on maximum congruence with the majority-rule consensus (fig. S1). Posterior probability clade support values indicated at nodes >0.5. Approximate known species numbers in terminal taxa are given in parentheses. Black circles mark significant shifts in diversification rate of sister clades (table S5). Colored triangles mark character transitions in lifestyles inferred by parsimony optimization (see figs. S2 and S3 for details).

sperm feeders, whereas the remainder showed no association with transitions to feeding on angiosperms or seed plants (table S5). A significant increase in diversification rate was inferred near the base of the Polyphaga whether herbivorous taxa were included or excluded from the analyses (table S5). Herbivory has played a role in the diversification of some beetle lineages, but the trait per se does not explain why beetles are so diverse.

Fast diversification rates also do not explain beetle diversity. Dating the tree with fossil calibration and penalized likelihood rate-smoothing (Fig. 3 and table S6) (*15*), we estimated net diversification rates across terminal taxa of 0.048 to 0.068 Myear^{-1} (table S7), slightly lower than comparable measures for the angiosperms (0.077 Myear^{-1}) (*24*). However, more than 100 modern beetle lineages were present at the first appearance of crown-group angiosperms dated to <140 Ma on the basis of pollen records (*25*), and less than one-third of extant beetle species are associated with angiosperms (table S8 and fig. S3). Therefore, the extreme diversity of beetles reflects the Jurassic origin of numerous modern lineages, high lineage survival, and the diversification into a wide range of niches, including the utilization of all parts of plants. These switches into new niches occur repeatedly as, for example, the multiple shifts from terrestrial to aquatic habits in the evolutionary history of beetles, which occurred at least 10 times (Fig. 2 and fig. S2).

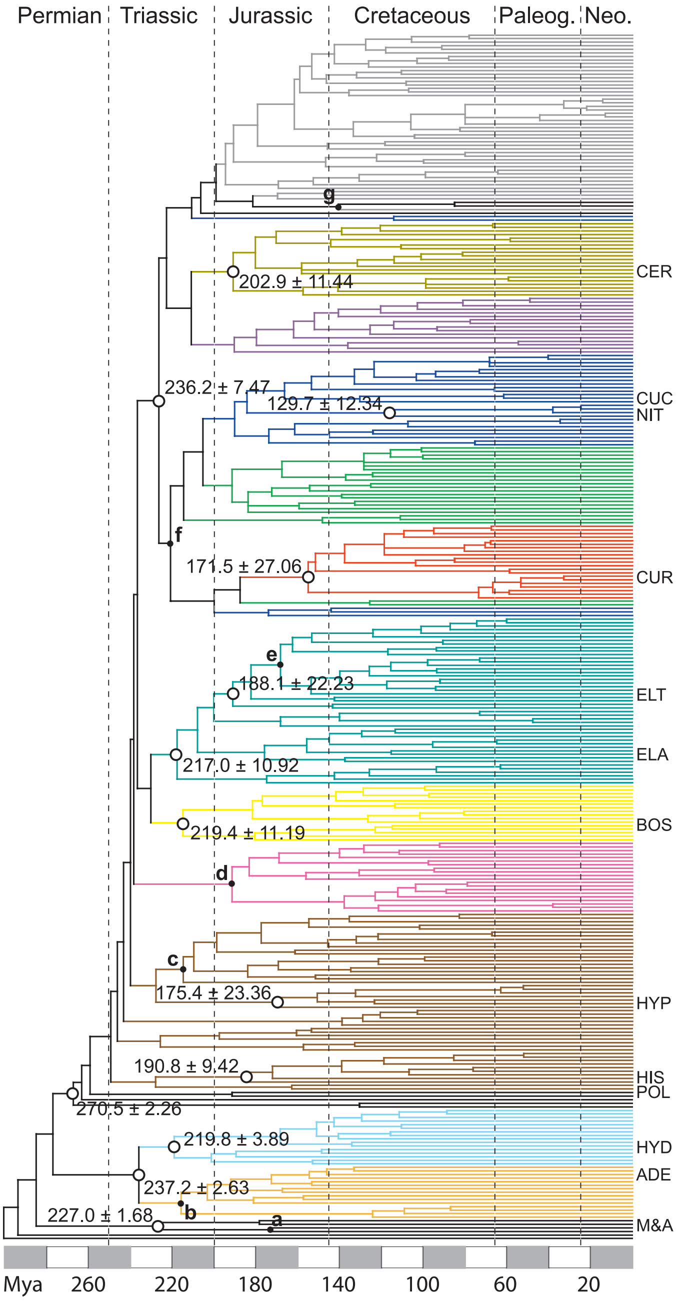

Fig. 3. A dated 340-taxon "all-compatible" consensus tree of Coleoptera from Bayesian analysis was dated with penalized likelihood placing the origin of Coleoptera at 285 Ma (*15*). Estimated number of lineages present at 200 Ma, 36; at 140 Ma, 145; and at 65 Ma, 301 (see also table S7). Colors correspond to the same groups as in Fig. 1. Numbers refer to average ages and 95% confidence intervals (*15*) of selected clades (open circles): CER, Cerylonid series; CUC, Cucujiformia; NIT, Nitidulidae; CUR, Curculionoidea; ELT, Elateroidea; ELA, Elateriformia; BOS, Bostrichiformia; HYP, Hydrophiloidea; HIS, Histeroidea; POL, Polyphaga; HYD, Hydradephaga; ADE, Adephaga; and M&A, Myxophaga and Archostemata. Seven fossil calibration points (table S6) were used to cross-validate rate-smoothing parameters (optimal value = 100) (*15*): point a, Cupedidae; b, *Sogdodromeus* (Geadephaga); c, Staphylinidae; d, *Holcorobeus* (Scarabaeoidea); e, *Elaterophanes* (Elateridae); f, *Cerambycomima* (Chrysomeloidea); and g, *Praemordella* (Mordellidae).

References and Notes

1. S. J. Gould, in *Dinosaurs in a Haystack* (Harmony, New York, 1996), pp. 377–387.
2. R. A. Crowson, *The Biology of Coleoptera* (Academic Press, London, 1981).
3. D. Grimaldi, M. S. Engel, *Evolution of the Insects* (Cambridge Univ. Press, Cambridge, 2005).
4. R. A. Crowson, *Annu. Rev. Entomol.* **5**, 111 (1960).
5. B. D. Farrell, *Science* **281**, 555 (1998).
6. A. L. V. Davis, C. H. Scholtz, T. K. Philips, *J. Biogeogr.* **29**, 1217 (2002).
7. T. L. Erwin, in *Taxonomy, Phylogeny and Zoogeography of Beetles and Ants*, G. E. Ball, Ed. (W. Junk, Dordrecht, Netherlands, 1985), pp. 437–472.
8. R. A. Crowson, *The Natural Classification of the Families of Coleoptera* (Nathaniel Lloyd, London, 1955).
9. J. F. Lawrence, A. F. Newton, in *Biology, Phylogeny, and Classification of Coleoptera: Papers Celebrating the 80th Birthday of Roy A. Crowson*, J. Pakaluk, S. A. Slipinski, Eds. (Museum i Instytut Zoologii PAN, Warzawa, 1995), pp. 779–1066.
10. R. G. Beutel, R. A. B. Leschen, *Coleoptera, Beetles. Volume 1: Morphology and Systematics*, vol. IV of *Handbuch der Zoologie/Handbook of Zoology*, N. P. Kristensen, R. G. Beutel, Eds. (de Gruyter, Berlin, 2005).
11. J. F. Lawrence, A. F. Newton, *Annu. Rev. Ecol. Syst.* **13**, 261 (1982).
12. J. Pakaluk, S. A. Slipinski, Eds., *Biology, Phylogeny, and Classification of Coleoptera: Papers Celebrating the 80th Birthday of Roy A. Crowson* (Muzeum i Instytut Zoologii PAN, Warzawa, 1995).
13. V. L. Shull, A. P. Vogler, M. D. Baker, D. R. Maddison, P. M. Hammond, *Syst. Biol.* **50**, 945 (2001).
14. M. S. Caterino, T. Hunt, A. P. Vogler, *Mol. Phylogenet. Evol.* **34**, 655 (2005).
15. Materials and methods are available as supporting material on *Science* Online.
16. W. C. Wheeler, M. Whiting, Q. D. Wheeler, J. M. Carpenter, *Cladistics* **17**, 113 (2001).
17. J. F. Lawrence, *Spec. Publ. Japan Coleopterological Soc. (Osaka)* **1**, 351 (2001).
18. S. Q. Ge, R. G. Beutel, X. K. Yang, *Syst. Entomol.* **32**, 635 (2007).
19. V. R. Viviani, *Cell. Mol. Life Sci.* **59**, 1833 (2002).
20. J. Kukalova-Peck, J. F. Lawrence, *Can. Entomol.* **125**, 181 (1993).
21. S. A. Slipinski, J. Pakaluk, in *Advances in Coleopterology*, M. Zunino, X. Belles, M. Blas, Eds. (European Association of Coleopterology, Barcelona, 1991), pp. 79–88.
22. T. J. Davies *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **101**, 1904 (2004).
23. J. B. Slowinski, C. Guyer, *Am. Nat.* **134**, 907 (1989).
24. S. Magallon, M. J. Sanderson, *Evol. Int. J. Org. Evol.* **55**, 1762 (2001).
25. E. M. Friis, K. R. Pedersen, P. R. Crane, *Palaeogeogr. Palaeoclim. Palaeoecol.* **232**, 251 (2006).
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Supporting Online Material

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