



Fig. 4. Alternative rootings evaluated with Bayes factors (BF) and the Shimodaira–Hasegawa (SH) test. Constraint trees used to evaluate alternative rootings are depicted on the left. Rootings 1–3 correspond to the first rooting considered by Brady *et al.* (20), but with alternative arrangements of *Martialis* and Leptanillinae. Rootings 4–7 correspond to Brady *et al.*'s rootings 2–5. For each alternative rooting, the corresponding value of $2\ln(\text{BF})$ is given for the comparison of that rooting against the maximum posterior probability (MPP) rooting 1. Values lower than -10 can be interpreted as strong evidence against the alternative. *P* values resulting from the comparison of alternative rootings to the maximum likelihood (ML) rooting 1 using the SH test are also given. Rootings significantly worse than the ML rooting would have *P* values < 0.05 .

the poneroids was recovered, even if the outgroups, *Martialis*, the leptanillines, both the outgroups and *Martialis*, or both the outgroups and the leptanillines were removed from the analysis (Figs. S1 and S2).

Our phylogenetic analyses, combined with the inferred biology of *M. heureka*, suggest that the most basal extant ant lineages are cryptic, hypogaecic foragers, rather than wasp-like, epigaecic foragers (Fig. 3). This finding is congruent with recent molecular studies (20–23), which previously suggested the Leptanillinae, another subfamily of subterranean predators, to be sister lineage to all extant ants. This result has puzzled ant systematists for two reasons. First, Wilson *et al.*'s (16, 17) classic study of the Mesozoic amber ant *Sphecomyrma* postulated that the ancestral ant was a large-eyed, wasp-like, ground forager, creating a strong expectation that the most basal extant ant lineages would also be epigaecic foragers, presumably similar to *Sphecomyrma*. Second, the Leptanillinae share common morphological (10, 38) and behavioral (39, 40) characteristics with the Amblyoponinae, implying the monophyly of this group (20). In contrast, our results and recent molecular systematic studies (20–23) suggest that blind, subterranean, specialized predators, like *Martialis*, the Leptanillinae, and some poneroids, evolved early during ant diversification. We hypothesize, that once these hypogaecic predators adapted to their specialized subterranean environment, their morphology and biology changed little over evolutionary

time because their hypogaecic habitat has likely been ecologically stable and provided a refuge from competition with other, more recently evolved, ants. It is important to note that no definitive statement about the morphology and life history of the ancestral, Mesozoic ant can be derived from our current knowledge about the surviving basalmost ant lineages, because the relative probabilities of evolutionary transitions between epigaecic and hypogaecic habits are uncertain. On the basis of the combined evidence of morphological, phylogenetic, and Mesozoic fossil data, the possibility remains that the ancestral ant was a large-eyed, wasp-like, epigaecic forager (16–19), rather than a blind, specialized, hypogaecic predator.

The Mesozoic fossil *Sphecomyrma freyi* has been interpreted as a representative of the stem-group ants and ant ground-plan characteristics have been derived from its morphology, suggesting that the most recent common ancestor (MRCA) of ants could be similar to tiphoid wasps (16–18). By considering both the topology of our inferred phylogeny (Fig. 3), and the timing of ant diversification presented in previous studies (20, 21), the Sphecomyrminae could alternatively be placed within the subfamilies grouped in the formicoid clade or as stem-group members of the formicoid subfamilies rather than the Formicidae as a whole (16–19). However, the morphology-inferred sister group relationship of *Sphecomyrma* to all extant ants (14) and the absence of hypogaecic ant fossils in Cretaceous deposits (11, 24, 41–50) do not support this hypothesis.

Conclusion. *Martialis heureka* exhibits a mosaic of plesiotypic ant characters and derived morphological adaptations to its hypogaecic predaceous lifestyle. The discovery of *Martialis* and our phylogenetic analyses support the hypothesis that several of the most basal extant ant lineages are hypogaecic foragers, suggesting that hypogaecic foraging evolved at least thrice independently during the early evolution of ants or that the ancestral ant was a hypogaecic forager. These evolutionary scenarios are congruent with the dynastic succession hypothesis for ant evolution (5, 20, 21), proposing a ground-associated ant ancestor, derived from a wasp-like, aculeate predator that radiated into specialized soil, leaf litter, and arboreal habitats potentially coinciding with the diversification of the angiosperms. *Martialis* and other soil-dwelling basal ants might therefore be relicts of early ant diversification, which survived in an environmentally stable refuge habitat, permitting them to retain plesiotypic characteristics because of reduced selective pressures. The exact nature of the ancestral ant remains uncertain, given that the propensity for repeated evolution of a hypogaecic lifestyle may be higher than for reevolution of an epigaecic lifestyle. Interestingly, *M. heureka* is a unique Neotropical representative of an extant, basal ant lineage. This discovery hints at a wealth of species, possibly of great evolutionary importance, still hidden in the soils of the remaining rainforests. Nocturnal leaf litter sampling and subterranean traps would be promising additions to future biodiversity surveys.

Materials and Methods

Photomicrographs were taken using a JVC KY-F70 digital camera mounted on a Leica Z6 APO dissecting scope. Composite images were assembled from 20 sequential photographs with Syncroscopy AutoMontage (v 5.0) software. DNA was extracted from the right front leg using a QIAGEN DNA Micro Kit. Two nuclear ribosomal genes and one single-copy nuclear gene were amplified using conventional PCR methods and sequenced (20). We obtained a total of 4926 aligned bp: 1904 bp for 18S, 2505 bp for 28S, and 517 bp for elongation factor 1 F2. We failed to sequence additional genes from the limited amount of DNA extract. The obtained DNA sequences were aligned with the data matrix of Brady *et al.* (20).

The phylogenetic position of *Martialis* was estimated using both ML and BI. ML trees were inferred using Garli v0.951 (30) with a single model of evolution. Bayesian phylogenies were inferred using MrBayes v3.2 (31) with a four-partition model (1st and 2nd codon positions of EF1 α F2, 3rd codon positions