

Regularities and irregularities in periodical cicada evolution

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Periodical cicadas are one of those creatures that everyone in eastern North America knows, if only periodically. The seemingly magical invasion every 17 or 13 y of millions upon millions of black, red-eyed, vaguely demonic-looking insects that fill the air with their incessant droning is, indeed, not something that one can easily ignore. These insects even inspired Bob Dylan, in his song “Day of the Locusts,” to proclaim that the cicadas were singing just for him, which seems at odds with current evolutionary theory. In PNAS, Sota et al. (1) make a much more meaningful contribution to our understanding of the evolution of the genus *Magicicada*.

Evolutionary interest in periodical cicadas is driven by their singular life history. The basic life cycle, with a long nymphal stage that feeds underground on the roots of trees and then emerges into a brief adult stage, which mates and oviposits, is not in itself unusual. But they have not only the longest total life cycle of any insect, but two alternative long cycles of 13 and 17 y. The fact that 13 and 17 are prime numbers has not escaped the attention of evolutionary theorists (the literature of *Magicicada* may be unique in encompassing both pest management and mathematical journals; see for example, ref. 2). However, the peculiarities do not stop there. Each species is organized into several developmentally synchronized “broods” (15 total across all species), each with its own characteristic years of adult emergence and geographic range. However, here is the real kicker: almost all broods contain three morphologically distinct, rarely interbreeding forms, called Decula, Cassini, and Decim, all of which have life cycles synchronized with each other. Note that I intentionally used the nonspecific term “forms” in the preceding sentence. This is because species concepts for these insects have been fluid. The morphologically distinct Decula, Cassini, and Decim are species under any conceivable concept, but current opinion (3) is to consider these as species groups and set the species category one level lower, treating the morphologically identical and genetically very similar 17- and 13-y populations in each

species group as operational species (e.g., 17-y *Magicicada septendecula* and 13-y *Magicicada tredecula* of group Decula) for a total of six species. (The symmetry of this system was upset by the recent identification of *Magicicada neotredecim*, about which more later.) All broods of each of these species have occasionally been proposed as species.

Sota et al. (1) have now uniquely cast a great deal of light on the *Magicicada* species problem by providing a phylogeny that includes all species groups, species, and broods. However, in doing so they also add a whole new level of complexity, consisting of significant geographic population structure within each species group, based originally on mitochondrial sequences and now reinforced by nuclear sequences. The pattern in Decim (4) consists of four geographic clades: eastern, middle, and western in the northern half of eastern North America, and the B clade in the southern half. However, the remarkable part of this division is that this phylogeographic pattern is now seen to be repeated in the Cassini and Decula species groups, with the exception that they lack the B clade. The capstone to this parallel population structure is the inclusion across the three species groups of the same clades in a given brood.

If you are confused at this point, you are not alone; I had to ponder all this for quite some time before I could begin writing this commentary. To assist in explaining the fresh insights of Sota et al. (1), I combined elements of their figures 1 and 2 (figures 1 and 2 in ref. 1) and made some simplifications (see the legend to Fig. 1 for species name abbreviations). To understand Fig. 1, imagine that you are in New Jersey later this very year and visit a local wood, aswarm with millions, if not billions, of cicadas. You pick one from your shirt and study it carefully. By going to www.magicicada.org on your cell phone you discover that only brood II is emerging in this area this year. Using the instant DNA sequencing application of your cell phone that will no doubt soon be available, you obtain a mitochondrial sequence that matches sequences from a

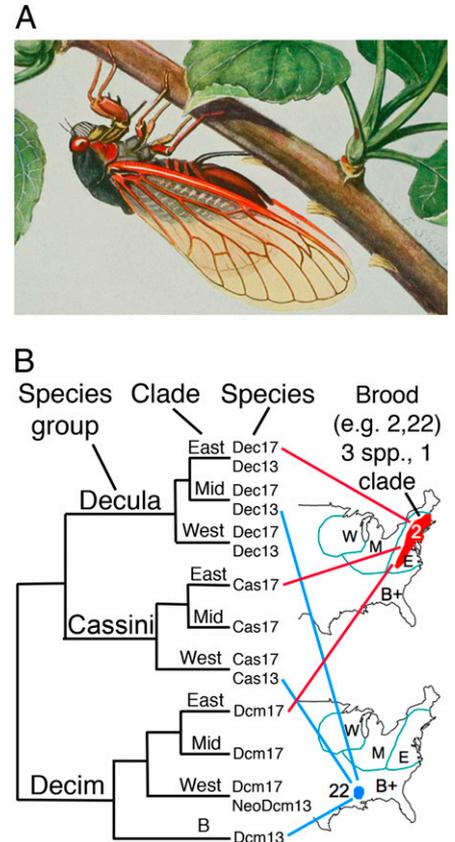


Fig. 1. (A) Female of the 17-y periodical cicada *M. septendecim* laying eggs in a twig. Image reproduced from ref. 9. (B) Simplified schematic of the phylogeny of *Magicicada* showing relationships of species groups, geographic clades, species, and broods. Branch lengths not to scale. Species are abbreviated by denoting Decula, Cassini, and Decim as Dec, Cas, and Dcm, respectively, and replacing the prefixes Septen and Tredec with the suffixes 17 and 13, respectively. Thus, *M. septendecim* is abbreviated Dcm17. *M. neotredecim* is abbreviated NeoDcm13. See text for full explanation. B+ means that the Decim B clade plus other clades of Decula and Cassini occur in this area.

17-y, eastern mitochondrial clade containing *Magicicada cassini* (Cas17 in Fig. 1). Traveling further down the tree in Fig. 1 tells us it is from the species group Cassini. We then sample a few more cicadas, and find not only other *M. cassini* sequences, but

Author contributions: S.H.B. wrote the paper.

The author declares no conflict of interest.

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also *M. septendecim* (Dcm17) and, rarely, *M. septendecula* (Dec17), both of which, surprisingly enough, also belong to the eastern clade of their respective species groups.

This congruence of geographic clades across species is one of the revelations of Sota et al. (1) and suggests parallel evolution of the species constituting these clades. However, the congruence does not hold up in all cases. As shown in Fig. 1, the 13-y brood XXII is made up of *M. tredecula* (Dec13) from a middle clade, *M. tredecassini* (Cas13) from a western clade, and *M. tredecim* (Dcm13) from the unique southern clade B of this species. Even more indicative of independent evolution of the members of some broods is the seventh and most recently described species, *M. neotredecim*. As discussed at length in earlier key work initiated by core *Magicicada* researcher Chris Simon [see Cooley et al. (5)], the northwesternmost broods of what was originally considered to be *M. tredecim* actually constitute the parapatric *M. neotredecim*, which shows reproductive character displacement in the narrow region of overlap. The unique nuclear sequences in Sota et al. (1) confirm the previous mitochondrial evidence that the 13-y *M. neotredecim* arose very recently from a population of the 17-y *M. septendecim*, not the 13-y *M. tredecim*, underlining the importance of life-cycle shifts in *Magicicada* evolution. As can be seen in Fig. 1, the occurrence of both life cycles in several clades of all three species groups indicates much evolutionary shifting of life cycles.

Although Sota et al. (1) have brought much clarification to our current understanding of *Magicicada* evolution, there remains a need for even deeper sampling of individuals and genes. What I have called “clades” are referred to as “haplotype groups” by Sota et al., because the mitochondrial trees (figure S1 in ref. 1) contain a mixture of true clades (e.g., the eastern and middle clades of Decim and Cassini) and paraphyletic assemblages (e.g., the western clades of these two species groups). In the case of the Decula group, there is a striking lack of variation, with only four haplotypes among 151 individuals sequenced. In addition, as Sota et al. point out, several broods contain mitochondrial haplotypes from different, although often adjacent, geographic clades; substantially larger sample sizes will be needed to completely understand the origin of this haplotype sharing.

Overall, Sota et al.’s (1) results bear on some important questions for speciation research. The first concerns the possibly conflicting roles that ecology can play during adaptation and speciation. As has been proposed since at least Marlatt (6), predation pressure may be the key force that drives both the length and synchronization of *Magicicada* life cycles. Emerging in very large numbers likely satiates predator populations, increasing the probability of individual cicada survival. This strategy is enhanced by having a very long life cycle, so that predators are kept at low population densities for all but emergence years. The icing on the cake may be the evolution of prime number generation lengths, which mathematical models have shown can inhibit the synchronization of shorter predator life cycles with those of periodical cicadas (1, 7). The initial evolution of the characteristic periodical cicada life history thus represented the emergence of an evolutionary novelty.

However, species arising later in the periodical cicada lineage could well have been constrained by the success of the initial species. A species that “goes off on its own” into a new geographic area might encounter greater predation pressure than if it stayed in the same area—and retained the same life-cycle periodicity—as the initial species. Adding more species to a brood, and thus even more individuals to satiate predators, could be advantageous to all. This is almost certainly the explanation for the geographic cooccurrence of a Decim, a Cassini, and a Decula species in almost all broods, which could perhaps be interpreted as a kind of cospeciation.

Divergence times estimated from the molecular clock indicate that divergences did not, in fact, occur simultaneously (figure 1 in ref. 1). The date of the most recent common ancestor, which can be taken as an estimate of when a species originated, differs among the three species groups. The most recent

common ancestor of Decim is estimated from the genetic data to be at 530,000 years ago (kya); Cassini is more recent at 320 kya, and Decula is most recent at 230 kya. Unfortunately, although these dates strongly suggest sequential speciation, the 95% highest-probability density intervals of the divergence time estimates overlap fairly broadly, which means that we will not have a complete understanding of this possibility without much more extensive—probably genomic—data.

Sota et al. (1) conclude that the “repeated shifts between the two prime-numbered life cycles in all three *Magicicada* species groups suggest a common genetic basis that evolved prior to the origin of the species groups,” or in other words, that there is ancestral variation for life-cycle length. Identifying and locating such genes or gene regions would certainly carry us much further toward understanding *Magicicada* evolution. Consider what has been learned from perhaps the most significant ancestral polymorphisms known at present, the lateral plate-armor polymorphism of the three-spined stickleback *Gasterosteus aculeatus*. Ancestral marine and derived freshwater populations differ in plate armor largely because of variation in the *Ectodysplasin* gene, at which the freshwater low armor-plate allele invariably consists of the same 16-kb block of sequence (8). The presence of the low plate-number allele at low frequencies in all marine populations worldwide contributes greatly to solving the mystery of the repeated, extremely rapid, parallel evolution of freshwater sticklebacks. Might we expect a similar step forward in understanding of *Magicicada* evolution to be fueled by the discovery of a similarly ancient, widespread polymorphism for life-cycle variation? Quite likely. However, somehow, given the peculiarities of these insects, we shouldn’t be too astonished if some totally unanticipated basis for life-cycle evolution is found.

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