Wing stridulation in a Jurassic katydid (Insecta, Orthoptera) produced low-pitched musical calls to attract females

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Behaviors are challenging to reconstruct for extinct species, particularly the nature and origins of acoustic communication. Here we unravel the song of *Archaboilus musicus* Gu, Engel and Ren sp. nov., a 165 million year old stridulating katydid. From the exceptionally preserved morphology of its stridulatory apparatus in the forewings and phylogenetic comparison with extant species, we reveal that *A. musicus* radiated pure-tone (musical) songs using a resonant mechanism tuned at a frequency of 6.4 kHz. Contrary to previous scenarios, musical songs were an early innovation, preceding the broad-bandwidth songs of extant katydids. Providing an accurate insight into paleoacoustic ecology, the low-frequency musical song of *A. musicus* was well-adapted to communication in the lightly cluttered environment of the mid-Jurassic forest produced by coniferous trees and giant ferns, suggesting that reptilian, amphibian, and mammalian insectivores could have also heard *A. musicus'* song.

call evolution | Tettigoniidae | bushcricket | biomechanics | biological asymmetry

Katydids produce species-specific calling songs that form part of the acoustic ecology of tropical forests (1, 2). The rubbing of a toothed vein on one wing against a plectrum on the other wing results in sound production by stridulation, exploiting resonant (musical) (3, 4) or nonresonant (broadband) (4, 5) biophysical mechanisms, depending on the species. Which of these two mechanisms represents ancestral sound production remains a key question in the evolution of insect acoustic communication (6–8). Here, we reconstruct the song of a katydid fossil with exceptionally well-preserved stridulatory structures, and find that musical singing at low frequencies was already established by the middle Jurassic (165 Ma). We describe this specimen as *Archaboilus musicus*, from the extinct family Haglidae (Orthoptera), a group basal to all extant katydids (9, 10). These findings imply that *A. musicus* was nocturnal, and that its call was adapted for long-range communication in an environment with light clutter (11). A recent paleobotanical reconstruction of the geometry, vegetation density, and biomass of the Jurassic Forest from northwest China (12) reveals an environment populated by coniferous trees (e.g., *Araucaria*) with nearest neighbor distances ranging from 1.5 to 20.3 m, and giant ferns (e.g., *Angiopteris*, *Osmunda*, and *Caniopteris*) occupying the lower layers of the understory forest. This forest architecture indicates a sparse vegetation density that is acoustically compatible with the proposition that the song frequency of *A. musicus* was well adapted to long-distance communication close to the ground. Such a forested environment would also enable long-range acoustic signaling and communication by other animals (e.g., amphibians, reptilians) as well as a variety of arthropod species (13).

Systematic Paleontology

Insecta Linnaeus, 1758; Orthoptera Olivier, 1789; Haglidae Handlirsch, 1906; Cyrtophyllitinae Zeuner, 1935; *Archaboilus* Martynov, 1937.

Archaboilus musicus Gu, Engel and Ren sp. nov.

Holotype. CNU-ORT-NN2009001PC, part and counterpart. The holotype (positive and negative imprints) is a pair of forewings as preserved. The right forewing (RW) is strongly fragmented, nearly half of the wing indiscernible; the left forewing (LW) moderately well preserved, base and apex partially missing (Fig. 1 *A–D*). The holotype is deposited in the Key Laboratory of Insect Evolution and Environmental Changes, Capital Normal University, Beijing, China.

Etymology. The specific epithet derives from Latin "*musicus*," for its tonal call.

Locality and Age. Collected from the Jiulongshan Formation, the late Middle Jurassic (Bathonian-Callovian boundary interval, approximately 165 Ma) of Daohugou Village, Shantou Township, Ningcheng County, Inner Mongolia, China (14, 15).

Diagnosis. Forewings are large (>7 cm), with broad-striped coloration; posterior radius (RP) ramifying earlier than anterior radius (RA); anterior media (MA) not distinctly undulating; the posterior branch of first posterior cubitus (CuPaβ) with broken section at level of the "handle" and directed toward anterior cubitus (CuA) and the anterior branch of first posterior cubitus (CuPaα). Wing venation nomenclature follows that standardized and used elsewhere for Orthoptera (16).

Description and Comparisons. Estimated total wing length is about 72-mm long. The forewing posterior subcosta (ScP) is undulating, with numerous branches; the radius (R) is straight, forking distally; RP is ramifying earlier than RA, and with four preserved branches. The area between the R and MA has a distinct vein separating two sets of parallel cross-veins between the RP and MA. The free section of the media (M) is short; the MA is curved, without distinct undulation; the posterior media (MP) is strongly curved toward the posterior margin in its basal section. The CuA + CuPaα with five branches, "handle" almost straight, reaching the last branch of CuA + CuPaα; the CuPaβ between its base and handle is slightly curved toward the wing
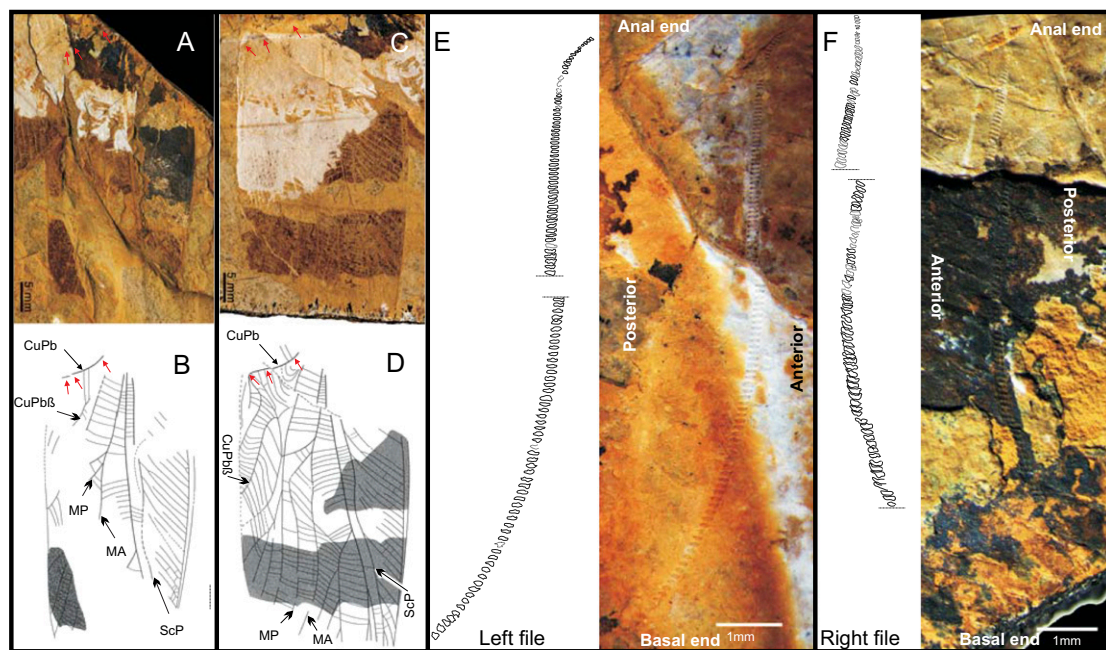
Fig. 1. *A. musicus* sp. nov. (*A–D*) photographs and line drawing of the RW and LW of holotype illustrating wing venation. Red arrows show the location of the stridulatory file (CuPb). (*E*) Detail of the LW file. (*F*) Detail of the RW file. Line drawings illustrate tooth morphology and spacing along the file. Details of tooth morphology are available in Fig. S1.

base, between the handle and posterior wing margin the CuPaβ is posteriorly curved, with a broken section at level of the handle directed toward the CuA + CuPaα. A "fan" reaches the second posterior cubitus (CuPb) at level of the bow of the CuPb. Cross-veins between the CuPb and CuPa are extremely curved in the basal part. Overall, the wing exhibits a disruptive and broad coloration pattern (Fig. 1 *A* and *C*).

In the stridulatory vein teeth developed on the ventral surface only. The LW file is well preserved (approximately 9.34 mm); the RW file is fragmented (approximately 7.81 mm), located on the CuPb (Fig. 1 *E* and *F*). The LW file shows 107 teeth, probably missing only a few in the median area. The RW exhibits 96 well-preserved teeth, missing approximately 11 toward the basal end. Tooth density of both files is approximately 11 mm. Teeth are asymmetrical, with lateral flaps projecting anteriorly, similar to extant Haglidae (Fig. S1).

This species is similar to *Archaboilus shurabicus* in wing venation but differs in the following characters: the forewing has broad-striped coloration, the RP is ramifying earlier, the vein connecting the MA and RP reaches free RP in the middle section, and the CuPaβ has broken section directed to CuA + CuPaα. *A. musicus* also differs from other Cyrtophyllitinae in the following ways: the MA is without distinct undulation, and the R is straight and branching earlier.

Discussion

Because of the lack of well-preserved fossils, the reconstruction of the evolutionary history of acoustic communication in ensiferan insects (crickets and katydids) is usually based on hypothetical phylogenetic hypothesis from extant taxa (7, 13, 17, 18). The best-known katydid fossil is the extinct *Pseudotettigonia amoena*, a tettigoniid from the lowermost Tertiary sediments of Denmark (also known as the "Mo-clay" or Fur Formation, 55 Ma), which preserves the sound-radiating cells of the stridulatory field and exhibits lower bilateral asymmetry in the forewings than extant species (19). *P. amoena* emerged relatively late in ensiferan evolution [as most Tettigoniidae (10)]; hence, it is distantly removed from the earliest calling insects (Fig. 2) and is likely to display derived characters.

A. musicus, here described, belongs to the family Haglidae, a group spanning the Late Triassic to the mid-Jurassic, but becoming extinct by the Early Cretaceous (Fig. 2). *A. musicus*'s closest living relatives are from the families Prophalangopsidae (e.g., *Cyphoderris* spp. and *Tarragoilus* spp.) and Tettigoniidae (katydids).

The fossil of *A. musicus* clearly shows veins of nearly identical dimension on both wings, and stridulatory files with exceptionally well-preserved and elaborate cuticular teeth (Fig. 1). Stridulatory files with such structural details are unknown in the fossil record. Most extant katydids exhibit conspicuous wing asymmetry: the LW is acoustically damped, but has a functional file; the RW exhibits sound-radiating cells, a plectrum, and an atrophied or absent file. Therefore, katydids can only sing using one wing overlap (LW over RW) and, depending on the species, produce either musical or broadband calls (Figs. S2–S4). In contrast, male *Cyphoderris* species exhibit highly symmetric wings with identical files (Fig. S5), can swap wing overlap for singing, and produce only musical calls (20). Given the phylogenetic position of Haglidae (Fig. 2), the wing morphology of *A. musicus* and other hagloids provide direct evidence that extant Tettigonioidea evolved from ancestors with bilaterally symmetrical wings.

A recent molecular phylogeny of Ensifera introduced the notion that musical stridulation is plesiomorphic for extant tettigoniids (21), as Prophalangopsidae (pure-tone species) emerge before Tettigoniidae. Paleontological evidence places Prophalangiosidae as descendants of a subfamily of the extinct, stem-group family Haglidae (9, 10) (Fig. 2). *A. musicus* therefore offers a unique opportunity to infer the origin and nature of sound production in this lineage of singing insects.

Using innovative comparative analysis (22) and current knowledge of the biomechanics of wing stridulation, we reconstructed the main properties of the acoustic signals of these ancient insects. In extant Ensifera, the production of pure-tone calls demands complex biomechanics (e.g., narrow-wing resonances, regularly spaced file teeth). However, wing symmetry and files with increasing tooth spacing constitute the unambiguous signature of wing resonance and pure-tone calls at low frequencies (3, 23–25). Symmetric wings have been shown to work in concert,
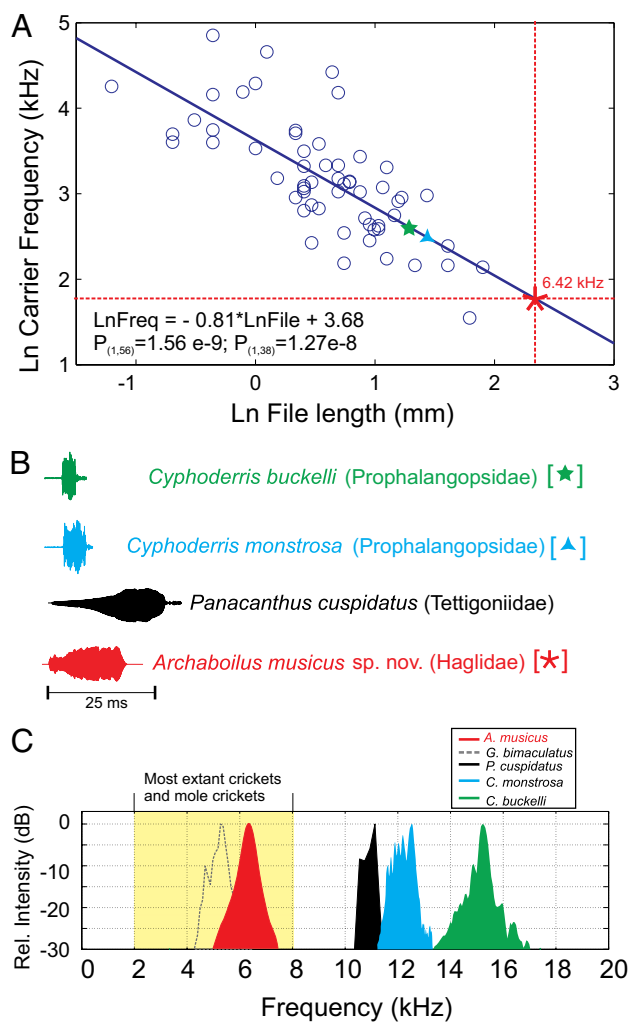
Fig. 2. Current phylogenetic relationship between major Ensifera inferred from molecular and morphological data (9, 10, 44, 45). Audible resonant stridulation has been documented in many extant species of crickets and mole crickets (among the Grylloidea), and in most extant prophalangopsids and many tettigoniids (within Hagloidea). However, cricket wings from the Early Cretaceous of Brazil look identical to those of modern species (6), which suggest musical song production. Gryllids emerged some 30 Ma after haglids, supporting the notion that musical stridulation in *A. musicus* preceded that of crickets. Some lineages of modern crickets evolved high frequency, nonresonant stridulation, and wing asymmetry (46).

■ Evidence of musical stridulation (3,5,7,8,20,25,28)
■ Possibly non-resonant stridulation (19)
■ Evidence of non-resonant stridulation (7)
● Evidence of stridulation, possibly musical (6)
□ Lost of stridulation (17)

optimizing the efficiency of sound radiation for low-frequency tones (23, 25, 26).

During song production, the wings accelerate in a closing scissoring manner, as the plectrum travels across the file (3, 4, 25). In pure-tone species, tooth spacing increases progressively toward the basal end of the file (i.e., in the direction of plectrum

motion during wing closure) (Fig. 3 *A–C*), a file morphology essential to generating a coherent, pure tone (3, 24, 25, 27). This increasing tooth spacing ensures that the rate at which the plectrum skips from one tooth to the next remains constant across the file, thus producing a constant strike rate and a constant carrier frequency (f_c) sustained by wing resonance (3, 27). Every tooth strike should be achieved at the period of the fundamental wing oscillation ($1/f_c$). In contrast, an accelerating closure over equally or irregularly spaced teeth fails to maintain a constant strike rate, and to match wing resonance, losing sound purity (*SI Text: Fossil Deposit and Venation Terminology*, and Figs. S3 and S4) (24). In *A. musicus*, tooth spacing increases linearly toward the basal end of both files (Fig. 3*D*). Such tooth spacing is just as well organized as that of extant species producing pure-tone calls (Fig. 3). Together, this tooth distribution and wing symmetry suggest that the stridulatory files of *A. musicus* were already adapted to producing pure-tone sounds, during the closing of the wings, in the same way as extant species.

Phylogenetic regression models between wing morphology and song features can predict song f_c from file morphology (28). In extant species using pure tones, only the file region with the linear increase in tooth spacing is used for sound production (Fig. 3 *A–C*; see also *SI Text: Fossil Deposit and Venation Terminology*). In *A. musicus*, tooth spacing increases linearly across the files of both wings (Fig. 3*D*), suggesting that the entire file was used to generate sounds.

Fitting the file length of *A. musicus* in the regression model indicates that these ancient Ensifera called with an f_c of approximately 6.4 kHz (Fig. 4*A*). Because tooth impacts take place every $1/f_c$ s, a continuous plectrum sweep over all 100 teeth results in a pulse duration of approximately 16 ms (Fig. 4*B*; see also Movie S1 for reconstructed ancestral song). This pulse duration is comparable to those of present day species using similar f_c (Fig. 4*C*).

In contrast to both extant and Tertiary fossil (19) Tettigoniidae, *A. musicus* possesses symmetric wings. The song of *P. amoena* is estimated to have a broadband structure centered at approximately 7 kHz (19). Therefore, the broadband calls mapped on tettigoniid phylogeny (Fig. 2) descend from ancestors that produced low-frequency musical songs using symmetric wings, like *A. musicus* and its relatives (29).

Fig. 3. Tooth distribution pattern of extant species of Ensifera and *A. musicus*. (*A*) *C. buckelli* with functional files on both wings (as indicated by colors, n = 8 males). (*B*) *Gryllus bimaculatus*, only the right file is measured (n = 20). (*C*) *Panacanthus cuspidatus*, only the LW file is functional and wings are strongly asymmetric (n = 5). Pink areas highlight the file region used for resonant stridulation. (*D*) Tooth distribution pattern in *A. musicus*. Tooth spacing gradually increases toward the basal end of the file. TD, tooth density per distance unit. Error bars represent one SD.

Fig. 4. Reconstruction of *A. musicus'* call features using a mathematical model (28) and biomechanics predicting song f_c and pulse duration from file morphology (Fig. S2). Regression phylogenetically corrected using the phylogenetic least-squares model algorithm (22). (*A*) Prediction of f_c. Empty and black circle data points represent file-length values for 58 species (28). Model was validated by computing f_c for two extant species *C. buckelli* and *C. monstrosa* (Prophalangopsidae) that generate tonal songs with symmetric wings (green and blue symbols). (*B*) Song pulse of extant tonal species (*C. buckelli, C. monstrosa*, and *P. cuspidatus*) and reconstruction of a song pulse produced by *A. musicus*. (*C*) Relative power spectra. *C. buckelli* and *C. monstrosa* produce songs at 13 and 15.5 kHz, respectively (20). *P. cuspidatus* produce calls at 10.8 kHz (7). Yellow highlighted area depicts spectral range used by extant field crickets, such as *G. bimaculatus* (4.7 kHz, line spectrum) (23), notably including the f_c predicted for *A. musicus* (red spectrum). *A. musicus* sound pulse was built taking into account tooth distribution and f_c, the spectrum was calculated on the synthetic pulse.

This mid-Jurassic fossil provides a deeper insight into acoustic communication 165 Ma. As an evolutionary innovation, the emergence of musical stridulation enabled the enhancement of broadcasting power and spatial range of communication (11, 30). The large sizes of both body and wings support the fact that *A. musicus* used low frequencies. However, low frequencies involve large wavelengths, which required large radiators for optimal source-to-medium sound power transfer (26). Hence, in *A. musicus*, the two large, identical, symmetrical wings stimulated at their shared resonant frequency constitute efficient sound radiators to improve this sound power transfer at such low frequencies. Pure-tone signals would have been advantageous then, as they are today,

to improve both signal-to-noise ratio for transmission and detection amid abiotic background noise (rain, wind, water streams). At 3–6 kHz, pure tones are better adapted for long-distance communication close to the ground than higher-pitched signals (30). *A. musicus* males seem to have serenaded females from the ground, a terrestrial niche preference also implied by the leg morphology of other cyrtophyllitines (29).

As extant katydid species using musical calls are nocturnal, *A. musicus* was likely to have been acoustically active at night. Using one frequency, *A. musicus* occupied a private communication channel to attract mates, an indication that the acoustic niche 165 Ma was already teeming with singing arthropods (6) and amphibians (13). In the darkness of the Jurassic forest, *A. musicus* surely experienced the dilemma of singing loud and clear, while simultaneously attempting to avoid nocturnal predators. Remaining quiet during the day, *A. musicus* perhaps managed not to raise the attention of contemporary diurnal *Archaeopteryx*-like reptiles. We cannot rule out the possibility that Jurassic mammals (e.g., *Morganucodon* and *Dryolestes*) might have been predators of these insects, as they were able to hear sounds in the range of frequencies used by *A. musicus* (31, 32).

Conclusion

Our results imply that resonance stridulation was already established by the mid-Jurassic in the tettigonioid lineage, some 100 Ma before the emergence of tettigoniid katydids (Fig. 2). Evidence of this establishment is the presence of symmetric wings with cells and files specialized for sound radiation in *A. musicus*, other Haglidae, and extinct and extant Prophalangopsidae (33–35). Because extant species of prophalangopsids use pure tones and stridulatory structures similar to those of *A. musicus*, we conclude that the elaborate broadband calls (see *SI Text: Fossil Deposit and Venation Terminology*, and Figs. S3 and S4) used by extant tettigoniids are derived from a resonant mechanism that involved the use of symmetric wings and files with increasing tooth spacing.

Because of the small number of fossils of early Tettigoniidae with well-preserved stridulatory structures, it is difficult to assert how widespread pure tones were in relation to broadband calls during the Tertiary. However, unquestionably, the resonant calls developed by haglids in the Jurassic might have been preceded by some other kind of primitive wing stridulation that involved nonelaborate broadband calls (see *SI Text: Fossil Deposit and Venation Terminology* for more information).

These calls might have been used by large species like Titanoptera (Fig. S6), an orthopteran group that diversified in the mid-Triassic (36). The wings of these insects were exceptionally large (approximately 15 cm) and show cells already modified for sound radiation, as well as several veins that might have worked as multiple stridulatory files (6, 36). The excitation of several stridulatory files at the time would have produced various vibrational patterns and frequencies, which ended in a noisy output. Curiously, examples of such heterogeneous stridulatory fields occur in the wings of females of extant *Panoploscelis* spp. (Pseudophyllinae), a neotropical genus. In this group the male sings a broadband call and the female answers with a noisy response (37, 38).

There should be an even more plesiomorphic condition that preceded such heterogeneous stridulatory fields of titanopterans; these could have involved random wing rubbing and friction of some veins, with pegs of irregular distribution and formation without cells specialized to radiate sound. This primitive form of sound production is observed in the defensive startle display of *Mantis religiosa* (39). Adult *M. religiosa* emit defensive sounds produced by abdomino-alary stridulation. This mechanism involves contact between irregularly distributed teeth on several of the longitudinal veins of the metathoracic wings against pegs located on the abdominal pleura. The acoustic output is very noisy and of

relatively low intensity. This type of stridulatory mechanism contrasts with the coherent structures responsible for the production of musical calls described above for many Ensifera, which—as documented herein—were already established by the mid-Jurassic.

Thus, the formation of random teeth across several veins on the forewings, and the associated production of noisy sounds, could have been the start of tegminal stridulation. Orthopteran insects using this form of "nonelaborate stridulation" probably occurred early in the Triassic. A hypothesis of the phylogenetic distribution of such evolutionary events is outlined in Fig. S6.

Methods

Measurements of Stridulatory Files. Analysis of the file morphology was performed on digitized scanning electronic microscopy photographs using the dimension tool of a drawing program (Corel Draw ×4, Corel). For the fossil specimen, measurements were done on photographs with high resolution and magnification (Fig. S2). See *SI Text: Fossil Deposit and Venation Terminology*.

Phylogenetic Least-Squares Model of Prediction of Carrier Frequency. The present analysis used comparative phylogenetic data reported by Montealegre-Z (28) (Table S1), including a phylogenetic tree of 58 species. File length was shown to have a strong effect on f_c after removing the effect of phylogeny using phylogenetically independent contrasts (PIC). However, PIC does not allow determining the *y* intercept because the regression line is forced through the origin. To recover the *y* intercept, we follow the procedure described by Lavin et al. (22).

The raw data (natural log-transformed) for file length vs. f_c used by Montealegre-Z (28) (Table S1), was exported as text files from Mesquite. The phylogenetic tree (28) was converted to variance-covariance matrices using the Mesquite package PDAP (40). The Mesquite tree was then exported to a PDI file. The Matlab program Regressionv2.m (22) was then used to implement linear statistical models via both ordinary (i.e., nonphylogenetic) least squares and phylogenetic generalized least-squares (PGLS) regressions (22, 41). RegressionV2 will request both the character matrix and the PDI file to compute the PGLS. For more details of the method see *SI Text*. In the analysis, file length was chosen as the independent variable and f_c as the dependant.

Calculation of Sound Acoustic parameters in the Fossil. Using the PGLS equation (Table S2), we predicted f_c for the fossil based on file length. The model was validated by computing f_c for two extant species *Cyphoderris buckelli* and *Cyphoderris monstrosa* (Prophalangopsidae) that generate tonal songs with symmetric wings. These two species are the closest living relatives of *A. musicus*. Pulse duration was calculated based on the following parameters and concepts: file length, the predicted f_c, evidence that the insect used the entire file during stridulation, and the fact that the sound produced by this insect was a pure tone. For a pure tone at 6.4 kHz, the duration of a single oscillation is 1/6,400. Because a single tooth strike would produce one oscillation (3, 27, 42, 43), the striking of nearly 100 teeth should last 100/6,400 (i.e., approximately 16 ms). Particulars of the calculation and reconstruction of the sound sequence can be found in *SI Text: Fossil Deposit and Venation Terminology*.

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EVOLUTION

Supporting Information

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Fossil Deposit and Venation Terminology

The specimen described in this contribution is housed in the Key Lab of Insect Evolution and Environmental Changes, Capital Normal University, Beijing, China.

The wing venation nomenclature used in this article follows Béthoux and Nel (1). Corresponding abbreviations are: ScP, posterior subcosta; RA, anterior radius; RP, posterior radius; MA, anterior media; MP, posterior media; CuA, anterior cubitus; CuP, posterior cubitus; CuPaα, the anterior branch of first posterior cubitus; CuPaβ, the posterior branch of first posterior cubitus; and CuPb, the second posterior cubitus (Fig. 1). The term "handle" describes a strong cross-vein appearing as a main vein. The term "Fan" describes the fascicle of cross-veins in the area between the CuPaβ and CuPb, diverging from the point of fusion between the handle with the CuPaβ or from the angle of the CuPaβ (when present), and reaching the CuPb.

SI Methods

Measurements of Stridulatory Files. The stridulatory file of extant species used in the analysis was studied under scanning electronic microscopy (SEM). Specimens mounted on stubs were gold-coated and studied by SEM using a Philips 501B. SEM images were digitized with a Keithley DAS 1202 plug-in card (Keithley Instruments), and the software SEM 1.2 (A. Gebert and G. Preiss, Medical School, Laboratory of Cell and Electron Microscopy, Hannover, Germany). Pictures with high magnification and resolution sufficient to provide accurate measurements of intertooth distances were taken for each specimen (Fig. S1). Therefore, different sequences of pictures of the same file were usually necessary. Analysis of the file morphology was performed on digitized SEM photographs using the dimension tool of a drawing program (Corel Draw ×4; Corel). In preserved specimens, intertooth distances were measured from the edge of the cusp of one tooth to the cusp of the next one. This process is equivalent to measuring the distance between teeth flaps (Fig. S1). Therefore, because the teeth of the stridulatory file preserved in the fossil specimen do not exhibit a clear cusp, intertooth distance was measured across using the teeth flaps preserved in the teeth. Measurements were done in photographs with high resolution and magnification (Fig. S1). For analysis and display, intertooth spaces were plotted against their corresponding tooth number. For example, the distance between tooth number 1 and tooth number 2 was plotted against tooth number 1, the distance between teeth 2 and 3 was plotted against tooth number 2, and so on. Tooth number 1 was the first tooth found in the anal end of the file (Figs. S2–S5).

Phylogenetic Least-Squares Model of Prediction of Carrier Frequency.

Male katydids (Orthoptera:Tettigoniidae) produce mating calls by rubbing the wings together, using specialized structures in their forewings (stridulatory file, scraper, and mirror). The wings open and close in a periodic manner, but sound is produced during the closing phase. A large proportion of katydid species (approximately 66%) reported in the literature produce ultrasonic signals as principal output. Montealegre-Z (2) studied the relationships among body size, generator structures, and the carrier frequency (f_c) in 58 tropical katydid species that use pure-tone signals. He used two different methods: (*i*) Ordinary least square (OLS), for exploring interactions of raw data, and (*ii*) phylogenetically independent contrasts (PIC) to control for phylogeny (3, 4). The phylogenetic hypothesis of the family Tettigoniidae proposed by Naskrecki (5) was used to build the clades of the studied species

and to define independent contrasts. This comparative analysis (2) shows how changes in sound generator form are related to changes in f_c. Anatomical changes of the sound generator that might have been selected via f_c and pulse duration are mirror size, file length, and number of file teeth. Selection for structures of the stridulatory apparatus that enhance wing mechanics via file-teeth and scraper morphology was crucial in the evolution of ultrasonic signals in the family Tettigoniidae.

The present analysis used data reported by Montealegre-Z (2) (Table S1), including the phylogenetic tree. This phylogeny shows 16 soft polytomies, which were resolved by inserting small branch lengths. Branch lengths were generated using the method of Pagel (6), implemented in the program Mesquite. The raw data for acoustic variables and file measurements used by Montealegre-Z (2) (Table S1) (file length vs. f_c) was exported as text files from Mesquite. These variables were shown to have a strong effect on f_c after removing the effect of phylogeny using PIC[2]. However, PIC does not allow determining the *y* intercept, as the regression line is forced through the origin. To recover the *y* intercept, we follow the procedure described by Lavin et al. (7) (see below).

The phylogenetic tree (2) was converted to variance-co-variance matrices using the Mesquite package PDAP (8). The Mesquite tree was then exported to a PDI file. For all statistical analyses, the response variable f_c with file length was natural log-transformed. The Matlab program Regressionv2.m (7) was then used to implement linear statistical models via both OLS and phylogenetic generalized least-squares (PGLS) regressions (7, 9). RegressionV2 will request both the character matrix and the PDI file to compute the PGLS.

The PGLS analysis was done using $n - 2$ degrees-of-freedom (*df*) (i.e., 56), but to produce a more conservative analysis, soft polytomies were taken into account and a second test was performed, reducing the *df* by the number of polytomies, to 38. The interaction of both dependent variables was highly significant using both criteria. For comparing and exploring results, analysis was done using OLS. Similar results were obtained with both methods. Results are shown in Table S2. Results involving PGLS and 56 *df* were used for the discussion in the article.

Correlations of stridulatory structures and carrier frequency in katydids have been done in the past, but these studies did not take into consideration the effect of phylogeny because suitable methods were not available at the time (e.g., refs. 10–13).

Reconstruction of the Sound.

Relevant terminology. Tooth strike: The impact produced the elastic clicking of the plectrum between two file teeth (the catch and release).

Resonant frequency (f_o): The natural frequency of vibration of the forewings.

Carrier frequency (f_c): The fundamental frequency of the calling song.

Pulse: The sound produced by a continuous sweep of the plectrum over all file teeth.

Period: The period of a wave is the time for a particle on a medium to make one complete vibrational cycle. Period is measured in units of time, such as seconds. Period is the reciprocal of frequency (*f*) (i.e., 1/*f*).

Duty cycle: The proportion of the call period spent emitting sound (calculated using syllable durations).

Syllable: Sound produced during a complete cycle of opening and closing movement of the wings.

Discrete pulse: In some species the closing phase is not continuous, as a result the sound produced is not sustained but composed of a series of short decaying pulses; these are referred to herein as discrete pulses.

Construction of the sound pulse. In pure-tone singers a sound pulse is built up of the summation of consecutive tooth strikes (2, 14–16). Because of the resonant properties of the wings, a single tooth strike generates a single wing vibration that lasts longer than the period of the wings own resonant frequency (f_o); therefore, it usually exhibits a long free decay (15, 17). However, the next tooth impact occurs at the correct phase with respect to the previous vibration, usually at the end of the completion of the fundamental cycle (15). Therefore, the duration of a single sound pulse can be mathematically reconstructed if the following parameters are known: the wing vibration produced by a single tooth strike, tooth spacing, wing velocity, and the carrier frequency (f_c). The pattern of vibration can be experimentally obtained by producing clicks of a plectrum against a file tooth, and recording either the resultant sound produced using a sensitive microphone or the associated wing vibration from the radiating wing cells using a laser Doppler vibrometer. Either of these methods shows a pulse rapidly building up in amplitude with gradually decaying oscillations following, and a dominant frequency at the wing f_o (which is similar to f_c of the calling song). In extant species using pure tones between 4 and 6 kHz, such pulses produce oscillations with a free decay of 3–4 ms (17, 18).

As the wing vibration of the fossil is not available, we produced a synthetic pulse with frequency at 6.4 kHz and decay of 4 ms; this is inspired by pulses produced by actuating the wing of *Panacanthus pallicornis*, a katydid species that uses a 5 kHz call that shows both resonant and nonresonant features (17, 19). We then used a Matlab program, which calculates the instantaneous period for every tooth impact based on the real tooth distribution and adds together the synthetic pulse every number of teeth (this program is available from the authors upon request). The basic period at 6.4 kHz is 1/6,400 ms, which constitutes the ideal time for individual tooth strikes. The average wing velocity can be inferred from the duration of the pulse and the file segment containing the functional teeth swept by the plectrum; for *Archaboilus*, all file teeth are functional. For a file of nearly 9 mm in length to be swept over 16 ms (pulse duration), the average wing-closing velocity should be around 560 mm/s. Our model assumes that wing velocity increases proportionally to the file tooth-spacing increments, from low values to a maximum value at the end, with the average velocity value reached toward the last one-quarter of the pulse, as observed in *P. pallicornis* and in *Cyphoderris* (17, 19). For *Archaboilus musicus*, the range of velocities assumed was from approximately 200 to 700 mm/s, reaching and average velocity value of 560 mm/s toward the last one-quarter of the pulse. The basic call structure is shown in Fig. 4*B*. We carried out spectral analysis on this synthetic call to produce the power spectrum of Fig. 4*C*.

Construction of syllable repetition pattern. For crickets and katydids with similar body sizes, the syllable repetition rate can greatly vary. Syllable rate also varies between individuals of the same species, even when measured at the same temperature and locality (20). In extant species, syllable rates and f_c are likely to be under the selection pressure of acoustic hunting predators, such as foliage-gleaning bats (21). For example, neotropical katydid species that are at risk for predation by bats tend to exhibit pure-tone song with low duty cycles and high frequencies (21, 22). Notably, large neotropical species singing at frequencies comparable to *A. musicus* (5–15 kHz) exhibit low duty cycles [e.g., *Panoploscelis specularis* (23), *Pristonotus tuberosus* (21), *Balboana tibialis* (21)]. As an adaptation that reduces acoustic conspicuousness, low duty cycles are less prevalent in the songs of paleotropical katydids than in those of neotropical species. Interestingly, the general trend is that paleotropical species

without bat predation pressures exhibit lower f_c and higher duty cycles (11).

Bats are well documented to have originated in the Early Tertiary (24); therefore, there were no bats at the time *A. musicus* filled the acoustic space of the Jurassic forest with its melodious song at low f_c. *A. musicus* certainly had not developed any antibat adaptations. Thus, although there could have been other acoustic predators, it is likely that *A. musicus* did not have such a low duty cycle as those shown by extant, giant neotropical species (21).

Consequently, for the reconstruction of the call duty cycle, we assumed these animals used the simplest call structure documented, a continuous single call (25, 26), with a relatively high duty cycle of 20–25%. This finding is in the range of that used by *Tympanophyllum arcufolium* (0.6 kHz) and *Chondroderella borneensis* (11.6 kHz), two large extant paleotropical species that do not experience bat predation pressure (11).

The complete structure of the call is considered to be a continuous sequence of syllables without particular grouping. This hypothesized plesiomorphic condition offers a continuous pattern that can be modified with different syllable groupings in the course of evolution (26). A video file with audio is available in Movie S1.

Stridulatory Apparatus in Ensifera with Emphasis on the Tettigoniidae

In this article we briefly discuss different types of stridulatory primary structures, with particular reference to the mechanisms involved in the generation of transient and discrete pulses in a pulse train.

The importance of the systematic distribution of file teeth in the production of pure-tone calls has been discussed in a few articles (15, 17, 18, 27–29). Below we summarize this in a survey of file morphology across different katydid species, which produce pure-tone or broadband songs, as well as some using songs that can be considered intermediate states between pure-tones and broadband. A complete review can be found in Montealegre-Z (18).

For the interaction of the file and scraper during stridulation, species have evolved one of two strategies of tooth contact: either the scraper moves over the file teeth (driven by wing forces) with the same velocity as that of the wing closure, or the scraper moves (at times) independently from wing motion, with higher velocity than the velocity of wing closure. The latter involves wing forces and elastic energy on the scraper itself (30). There are usually four basic modes by which ensiferans, using tegmino-tegminal stridulation, can throw the sound emitter into vibration (18). Two of these modes relevant to the present article are discussed as below.

Mode 1: Resonance Using Sustained Pulses. The number of stimulations per second almost coincides with the natural frequency of the emitter (forewings), so that each stimulus (tooth strike) evokes an oscillation, reinforced by the next stimulation before any decay can set in. This process is called resonance and occurs in the calling song of crickets, prophalangopsids, and in a large number of katydid species. Songs of this type are also termed pure-tone, musical, or tonal songs (Fig. S2).

Characteristics of this mode. In pure-production, every oscillation of the radiating surfaces corresponds to one tooth-impact, thus the resonator must be sharply tuned to vibrate periodically and to emit sinusoidal pulses when stimulated at its own f_o (14). A common feature of the stridulatory file in crickets (and also in some katydids using pure-tone songs) is that the intertooth space increases from the anal to the basal part of the file (Fig. S2). This feature is required to achieve compensation for proportional increments in scraper velocities. These increments in velocity might be the result of applying a constant force during the closing stroke of the wings (29), because there is little chance to

finely adjust force. Thus, there is no need for controlling, behaviorally, scraper velocity from one tooth to the next, which might be energetically more expensive. The scraper flexible region of the species using this mode is normally reinforced ventrally by another layer of cuticle that probably controls how much of this region bends. For all species studied, which use this mechanism, the velocity of the scraper displacement corresponds with the velocity of wing closure (17, 18, 31).

Additionally, some species perfectly adapt the velocity of the scraper to the morphology of the file, but others gradually reduce or increase the velocity of tooth contact (even if intertooth distances increase basally). The latter technique might results in frequency modulation or glissando effect.

Mode 2: Nonresonant Because of the Noisy Response of the Sound Generator. The tegminal emitter has no narrow natural f_o of its own, but produces a damped pulse of something approaching "white" noise at each stimulus (Figs. S3 and S4). The total band of frequencies would thus be distributed over a wide band instead of being concentrated into a single frequency (e.g., some katydid species).

Special features of this mechanism. Nonresonant calls are found in two forms among katydids: (*i*) a continuous pulse with high amplitude modulation (Fig. S3); and (*ii*) a pulse train built of various discrete pulses, each of which might show variable amplitude modulation, and then decays rapidly before the next discrete pulse begins (Fig. S4).

Continuous stimuli and damped resonator. The scraper contacts file teeth in sequence over large portions of the file, just as in the mechanism of resonance explained as mode 1. The difference is that the forewings will respond to the stimuli with a continuous vibration, for which spectral energy is noisy (32) (Fig. S3). The stridulatory file exhibits usually a random arrangement of teeth, but in some species the file might show an increasing random pattern of intertooth-distance increments. The last situation has been reported in species with pure-tone ancestry, which might preserve some of the file arrangement needed for pure-tone production (17, 19). The velocity of scraper displacement should match that of the wing closure, but the output sound is noisy because of the physical properties of the sound generator and the irregular pattern of file teeth. In some species, this situation can be thought of as a transitional stage between pure-tone emission and broadband with transient pulses (e.g., *P. pallicornis*) (19).

Transient pulses and damped resonator. If the wing vibration response to every stimulus (tooth impacts) is broadband, the distribution of teeth in the stridulatory file is not critical any more. Teeth might exhibit more or less constant spacing, so that compensation between increments in wing closing velocity and space will not occur, resulting in a broadband pulse-train song in which discrete pulses decay rapidly (Fig. S4). Every tooth impact will produce a discrete rapid-decaying pulse because of the damped response of the resonator (33, 34); this is common in several species of katydids, and might explain why the calling songs of most katydids are always referred to in the literature as broadband (35–37).

We must highlight the fact that some species of katydids using broadband calls seem to exhibit an increasing tooth-spacing pattern similar to that described for musical singers. Although these insects have a conspicuous wing asymmetry, it is unknown if this file morphology is derived from a pure-tone ancestor or if the observed condition secondarily evolved in the opposite direction (from broadband to pure tones).

Although the phylogeny of the Tettigoniidae is not completely resolved, one can note trends in the evolution of sound-production mechanism. Above we mentioned that the stridulatory file of *P. pallicornis* exhibits features that suggest some evolutionary transition between pure-tone and broadband. *Panacanthus* spp. have descended from a pure-tone ancestor, as implied by the position of *Panacanthus cuspidatus* in the phylogeny (19). *P. cuspidatus* exhibits a stridulatory file with very

proportional increments in tooth spacing (Fig. 3) and produces a high-quality song at approximately 11 kHz. Crownward in the phylogeny is *P. pallicornis*, singing with a relative pure tone at 5 kHz, but the spectrum also shows a broadband between 12 and 30 kHz. The file of this species exhibits increasing tooth spacing, but increments occur with a more irregular pattern than that found in *P. cuspidatus* (17). More crownward in the phylogeny of *Panacanthus*, all species produce broadband calls with infrastructure [i.e., transient pulse and damped resonator (Fig. S4)], and increasing tooth spacing is not observed any more. Because the wings of *P. pallicornis* show high resonance at 5 kHz, as measured with laser Doppler vibrometer (17), it is rational to think that selection for broadband songs in this genus began on the stridulatory file first and then on the wing resonant properties. Even if the wings have sharp resonances, tooth strikes do not occur strictly regularly, therefore other frequencies are also induced in the wing generator, producing the broadband observed. In a given lineage wing resonance might be lost because of the irregular tooth strikes that change the smooth vibrational properties of an "elaborate instrument." Similar tendencies have recently been observed in eneopterine crickets (38, 39).

In summary, the evolutionary transition from resonant to nonresonant mechanisms of stridulation is feasible, and selection might act first on the stridulatory file teeth (tooth spacing, angle of orientation, height, and so forth). Of course there are only a few documented examples where wing resonances and morphology have been completely studied in an evolutionary scenario. More work is needed on numerous other species.

How many teeth of the stridulatory file are used? All extant species (with calls properly documented) known to exhibit wing symmetry (or nearly so) and increasing intertooth distances pattern in their files (e.g., *Cyphoderris* spp, Gryllinae, molecrickets) tend to use pure tones in the audio range (15, 26, 40–45). Most species of field crickets are bound to sing at nearly 5 kHz, with song pulses lasting between 10 and 30 ms (26). *Cyphoderris* spp. use calls between 12 and 15 kHz, but shorter pulses than crickets, lasting 3–7 ms (44, 45). The wing having the file being struck experiences energy inputs at different locations as the scraper moves across the file, which should result in variable phases of vibrations at specific wing locations. This situation is different for the scraper wing, which has one and only one point of energy input, the scraper active edge; thus, this wing should vibrate with constant phase at all loci during a file-scraper sweep (18). Therefore, for higher frequencies (short periods) it seems to be harder to achieve the proper phase and synchronization of both wings (the essence of the escapement mechanism) (31, 46). Thus, *Cyphoderris* spp., singing three-times higher than crickets, are bound to stridulate using only a small file segment, even when the file is as large as that of large field crickets (3–5 mm) (18, 45). Hence, energy inputs will occur in a small region, and the phase between both wings will be achieved more straightforwardly than if the whole file was used (18). It has been demonstrated that the vibration of the file correlates with the form of the pulse envelope. The stridulatory file in crickets, *Cyphoderris*, and katydids usually vibrates with higher amplitude from the center toward the basal end and with very low amplitude at the basal end (15, 18, 43); therefore, a pulse that has a large build-up and large or moderate decay will suggest that the insect is using most of the file for stridulation, as the build-up and decaying portions are the result of low-amplitude vibration at the file ends (15). The small file segment used by *Cyphoderris* spp. for stridulation occurs in the central part of the file, which causes the pulses not to have a pronounced build-up and decaying parts (Fig. 4C and Fig. S5D). Because the file of *A. musicus* exhibits a linear continuous and gradual increment on intertooth distances, we suggest most of the file is being used for stridulation, and therefore the pulse must have shown a gradual build-up and a moderate decay. The synthetic pulse we produced shows these features (Fig. 4C).

SI Discussion

We conclude that the broadband calls used by extant tettigoniids are derived from a resonant mechanism that involved the use of symmetric wings and files with increasing tooth spacing. This analysis results from comparison of the sound output and morphology of the stridulatory apparatus in extant and extinct Haglidae and Prophalangopsidae species, as these groups are plesiomorphic to the Tettigoniidae. Wing symmetry, and pure-tone production at low frequency was already established in Haglids some 160 Ma, quite before the emergence of Tettigoniidae.

The resonant calls developed by haglids in the Jurassic might have been preceded by some other kind of primitive wing stridulation that involved nonelaborate broadband calls. By non-elaborate, we imply that the sound did not have infrastructure (i.e., did not have transient pulses or pulse trains, as that shown by many extant tettigoniids, which we call elaborate broadband). Such nonelaborate broadband calls might have been used by large species like Titanoptera, a orthopteran group that diversified in the mid-Triassic (47). The wings of these insects are exceptionally large (approximately 15 cm) and show heterogynous fields modified for sound radiation (cells and multiple files) (47, 48). Due to this anatomy, we hypothesize that the sound produced by titanopterans could have been noisy because several stridulatory files excited at the same time would have produced various vibrational patterns and frequencies. Heterogynous stridulatory fields are possible, and examples occur in the wings of females of extant species of *Panoploscelis* (Pseudophyllinae) from the Amazon. In this group, the male sings a broadband call and the female answers with a very noisy response (23, 49).

Such heterogynous fields of Titanopterans should have evolved from even less specialized structures. These structures could have involved random wing rubbing and friction of some veins with pegs of irregular distribution and formation. This primitive form of sound production is observed in the defensive startle display of *Mantis religiosa* (50). In adult *M. religiosa* this display entails both visual and acoustic components, the latter being abdomino-alary stridulation. Defensive sounds are generated via an abdomino-alary mechanism involving contact between irregularly distributed teeth found on several of the longitudinal veins of the metathoracic wings and pegs located on the abdominal pleura. The acoustic output of *M. religiosa* is very noisy and of relatively low intensity, with significant energy observed between 4 and 12 kHz, extending to the ultrasonic range at nearly 24 kHz. In several species of Phaneropterinae where the female stridulates in response to a calling male, the female wings exhibits curved spines on the dorsal surface of the anal area of the right tegmen (51). Besides *Panoploscelis* spp., females of other Pseudophyllinae katydids have stridulatory apparatus. Females of *Pterophylla camellifolia* have on the undersurface of the anal area of the left tegmen a network of veins bearing many short ridges (51). The sound produced by phaneropterinae katydids using this kind of stridulatory structuresis is usually broadband (36). This type of stridulatory mechanism is in contrast to the coherent structures responsible for the production of resonant calls described above for many Ensifera, which were already established by the mid-Jurassic. A hypothetical scenario is shown in Fig. S6.

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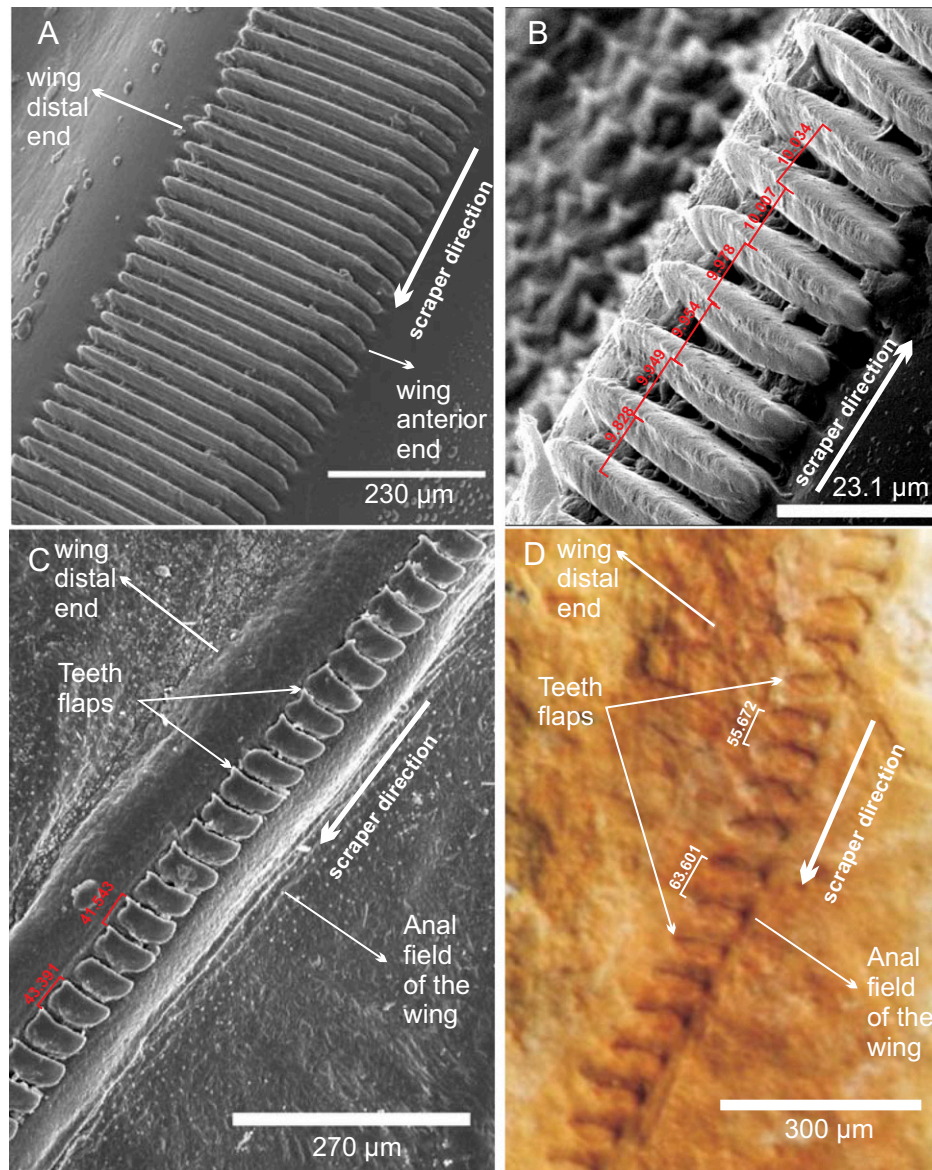
Fig. S1. File morphology of three different species of Ensifera, showing the method used to measure intertooth distances. (*A*) SEM picture of the file of (Tettigoniidae), a species used in the comparative analysis and phylogeny. *P. pallicornis*. (*B*) SEM picture of the file of *Daedalellus nigrofastigium* (Tettigoniidae), one of the species used in the comparative and phylogenetic analyses. (*C*) SEM picture of the left file of *Cyphoderris buckelli*. (*D*) Left file morphology of *A. musicus* (positive print seen dorsally). Note how the file of *A. musicus* is more similar to the file of *Cyphoderris* spp. than to the typical katydid file (Fig. S3). *C* and *D* highlight the tooth asymmetry imposed by wing flaps oriented toward the posterior wing end, in *Cyphoderris* and the fossil. Measurements in fresh material involve high magnification and high resolution pictures taken laterally from the file teeth (as that shown in *A*). The dorsal view of *Cyphoderris* file shown in *C* is used to depict the wing flaps.

Fig. S2. File morphology and sound quality in *Artiotonus artius*. (*A*) SEM picture of the file and tooth distribution. (*B*) Sound pulse. (*C*) High resolution of a segment of the pulse in *B*. (*D*) Power spectrum of a sound pulse. *Q* represents the average of five specimens.

Fig. S3. File morphology and sound quality in *Listroscelis* sp. (*A*) SEM picture of the file and tooth distribution. (*B*) Sound pulse. (*C*) High resolution of a segment of the pulse in *B*. (*D*) Power spectrum of a sound pulse.
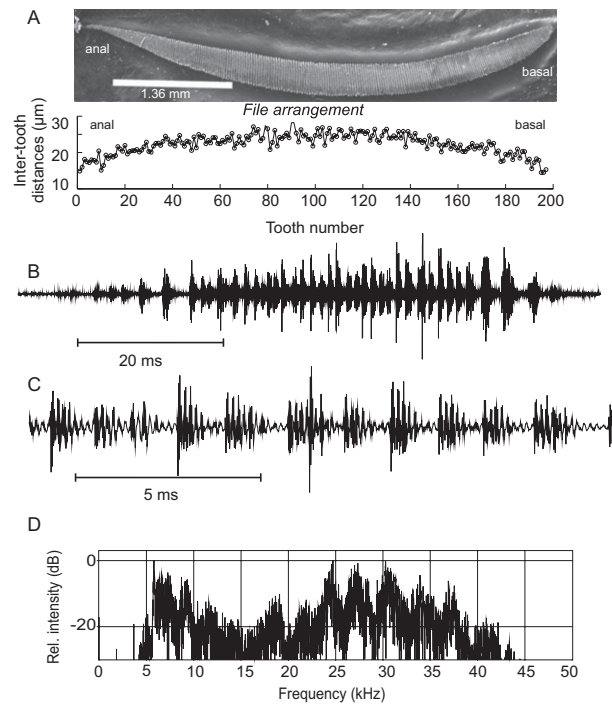
Fig. S4. File morphology and sound quality in *Panacanthus gibbosus*. (*A*) SEM picture of the file and tooth distribution. (*B*) Sound made of transient pulses. (*C*) High resolution of a segment of the pulse in *B*. (*D*) Power spectrum of a sound pulse.

Fig. S5. File morphology and sound quality in *Cyphoderris monstrosa*. (*A*) SEM picture of the left file and tooth distribution. (*B*) SEM picture of the right file morphology and respective tooth distribution. (*C*) A series of sound pulses. (*D*) High resolution of the segment enclosed in the dashed box in *C*. (*E*) Power spectrum of the sound pulse shown in *C*.

Fig. S6. Hypothetical evolutionary scenario for the evolution of stridulation in Ensifera.

Table S1. Anatomical and acoustic data used in the comparative analysis

| Species	f_c kHz	SD	F_L (mm)	SD	n	Source
Panacanthus pallicornis	4.7	0.2	6.0	0.2	5	(1)
Promeca Sumatran	8.5	0.0	6.7	0.0	4	(2)
Copiphora rhinoceros	8.7	0.0	3.8	0.2	3	(3)
Promeca perakana	8.7	0.0	5.0	0.0	1	(2)
Acanthacara sp.	8.9	0.6	2.1	0.0	2	(4)
Typophyllum mortuifolium	9.4	0.0	3.0	0.0	2	(5)
Panacanthus cuspidatus	10.9	0.3	5.0	0.0	2	(6)
Neoconocephalus stigmaticus	11.3	0.1	1.6	0.2	5	(7)
Chondroderella borneensis	11.6	0.0	2.6	0.0	1	(2)
nr. *Sphyrometopa* sp.	12.7	0.9	2.1	0.1	1	(7)
Aemasia sp.	13.2	1.1	2.7	0.0	1	(8)
Championica walkeri	13.3	0.2	2.8	0.3	5	(9)
Macrochiton macromelos	13.8	0.3	2.8	0.2	5	(9)
Typophyllum bolivary	14.0	0.0	2.6	0.1	5	(5)
Ischnomela gracilis	15.1	0.0	2.5	0.0	2	(7)
Copiphora brevirostris	15.6	0.0	3.2	0.1	3	(10)
Copiphora cf gracilis	16.5	0.3	1.5	0.0	2	(7)
Trichotettix pilosula	16.9	0.6	1.7	0.0	2	(9)
Mystron sp.2	17.6	0.3	1.6	0.0	1	(8)
Typophyllum sp. 1	18.4	0.5	3.3	0.0	1	(8)
Pemba cochleata	19.2	0.1	1.4	0.0	1	(8)
Typophyllum egregium	19.2	0.9	3.4	0.0	1	(7)
Typophyllum zingara	19.7	0.0	4.2	0.0	1	(9)
Typophyllum cf trapeziforme	20.5	0.0	2.4	0.0	1	(8)
Copiphora gracilis	20.6	0.7	2.0	0.0	2	(7)
Typophyllum sp. 205	20.6	0.0	1.5	0.0	1	(7)
Diacanthodis granosa	21.4	0.0	1.5	0.0	1	(7)
Eubliastes aethiops	21.6	0.8	2.9	0.0	2	(11)
Teleutias akratonus	22.0	0.0	1.5	0.0	2	(9)
Docidocercus gausodontus	22.5	0.0	2.1	0.0	7	(9)
Docidocercus gigliotosi	23.0	0.0	2.2	0.0	3	(10)
Leurophyllum modestus	23.0	1.4	1.6	0.0	2	(12)
Typophyllum trapeziforme	23.2	0.0	2.2	0.0	2	(5)
Docidocercus chlorops	24.0	0.0	2.0	0.0	1	(5)
Teleutias fasciatus	24.1	0.9	1.2	0.0	19	(9)
Eubliastes chlorodyction	27.3	2.5	3.0	0.0	1	(9)
Graminofolium sp.	27.7	0.0	1.5	0.1	2	(7)
Scopiorinus carinulatus	28.0	0.0	1.8	0.1	3	(5)
Scopiorinus mucronatus	28.0	0.0	2.0	0.0	1	(5)
Ancistrocercus circumdatus	31.0	0.0	2.4	0.1	3	(5)
Daedalellus nigrofastigium	33.0	3.1	1.5	0.1	3	(7)
nr. *Cowellia* sp.	34.1	0.8	1.0	0.0	2	(7)
Artiotonus tinae	36.0	0.0	1.7	0.0	1	(13)
Uchuca amacayaca	36.6	0.0	0.7	0.0	2	(14)
nr. *Paranelytra* sp.	36.7	1.3	0.5	0.0	1	(7)
Kawanaphila yarraga	40.3	6.0	0.5	0.1	3	(15)
Artiotonus artius	40.7	0.1	1.4	0.0	2	(13)
Teleutias castaneus	42.1	0.0	1.4	0.0	1	(9)
Uchuca halticos	42.3	0.0	0.7	0.0	7	(14)
Phlugis sp.2	47.6	0.0	0.6	0.0	1	(7)
Eschatoceras sp.	64.0	0.0	0.7	0.0	1	(7)
Myopophyllum sp.	65.5	7.7	2.0	0.0	1	(7)
Haenschiella ecuadorica	66.0	0.0	0.9	0.0	1	(10)
Kawanaphila mirla	70.5	8.3	0.3	0.1	7	(15)
Drepanoxiphus angustelaminatus	73.0	0.0	1.0	0.0	3	(10)
Mypophyllum speciosum	83.3	4.8	1.9	0.1	4	(10)
Haenschiella sp.	105.5	1.7	1.1	0.0	1	(10)
Arachnoscelis sp.	129.0	0.0	0.53	0.2	2	(11)

f_c, Carrier frequency; F_L, file length; n, number of samples. Table modified after Montealegre-Z (7).

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Table S2. ANOVA summary of the PGLM and OLS analysis of carrier frequency on file length

File vs. Fc	Coefficient	SE	t for H0	F for H0	P value (df = 56)	P value (df = 38)
PGLM						
Intercep	3.682	0.122	30.221	91.332	1.56E-09	1.27E-08
Slope	−0.81	0.113	−7.214	52.041	2.31E-13	1.19E-11
OLS						
Intercep	3.630	0.0831	43.664	190.656	0	0
Slope	−0.794	0.0973	−8.167	66.703	4.11E-11	6.88E-10
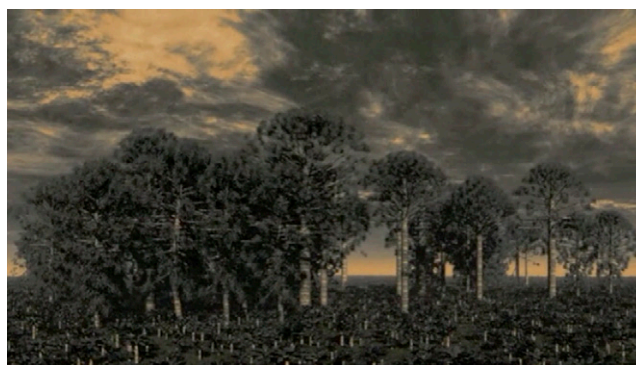
Movie S1. At dusk, *A. musicus* sings in a Jurassic forest of Northwest China. The 3D reconstruction of the forest is taken and modified from Hinz et al. (1). The forest grew under humid conditions, probably close to the banks of an anastomosing river and consisted primarily of conifers, in particular Araucariaceae, and ferns. The song was processed to take into account slight echoes produced by the lightly cluttered environment of coniferous trees (e.g., *Araucaria*), and giant ferns (e.g., *Angiopteris*, *Osmunda*).

[Movie S1](#)

1. Hinz JK, Smith I, Pfretzschner H-U, Wings O, Sun G (2010) A high-resolution three-dimensional reconstruction of a fossil forest (Upper Jurassic Shishugou Formation, Junggar Basin, Northwest China). . *Palaeobiodivers Palaeoenviron* 90:215–240.