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Basal polyphagan beetles in mid-Cretaceous amber from Myanmar: biogeographic implications and long-term morphological stasis

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The origin and early evolutionary history of polyphagan beetles have been largely based on evidence from the derived and diverse 'core Polyphaga', whereas little is known about the species-poor basal polyphagan lineages, which include Scirtoidea (Clambidae, Decliniidae, Eucinetidae, and Scirtidae) and Derodontidae. Here, we report two new species *Acalyptomerus thayerae* sp. nov. and *Sphaerothorax uenoi* sp. nov., both belonging to extant genera of Clambidae, from mid-Cretaceous Burmese amber. *Acalyptomerus thayerae* has a close affinity to *A. herbertfranzi*, a species currently occurring in Mesoamerica and northern South America. *Sphaerothorax uenoi* is closely related to extant species of *Sphaerothorax*, which are usually collected in forests of *Nothofagus* of Australia, Chile, and New Zealand. The discovery of two Cretaceous species from northern Myanmar indicates that both genera had lengthy evolutionary histories, originated at least by the earliest Cenomanian, and were probably more widespread than at present. Remarkable morphological similarities between fossil and living species suggest that both genera changed little over long periods of geological time. The long-term persistence of similar mesic microhabitats such as leaf litter may account for the 99 Myr morphological stasis in *Acalyptomerus* and *Sphaerothorax*. Additionally, the extinct staphylinoid family Ptismidae is proposed as a new synonym of Clambidae, and its only included species *Ptisma zasukhae* is placed as incertae sedis within Clambidae.

1. Introduction

Beetles (Coleoptera) are the most species-rich group of animals on our planet, exhibiting extraordinary morphological disparity and ecological diversity. Among four extant suborders of beetles, Polyphaga are the largest and most diverse group of beetles [1–3]. Clambidae is a small group of small-sized (0.7–2.0 mm long) polyphagan beetles distributed worldwide, with approximately 150 described species grouped in five extant genera [4]. Clambidae (Scirtoidea) include three extant subfamilies: Acalyptomerinae (*Acalyptomerus*

Crowson), Calyptomerinae (*Calyptomerus* Redtenbacher), and Clambinae (*Clambus* Fischer von Waldheim, *Loricaster* Mulsant and Rey and *Sphaerothorax* Endrödy-Younga) [4]. Among them, *Clambus* is the most species-rich genus, comprising approximately 130 described species with a worldwide distribution. Most clambid adults occur in decaying vegetation, leaf litter, and rotten wood and occasionally fly at dusk [5,6]. Recent molecular-based studies indicate that the superfamilies Scirtoidea + Derodontoidea (excluding Jacobsoniidae) are the basal-most groups of the hyper-diverse Polyphaga, sister to the remaining polyphagan beetles (core Polyphaga) [2,3,7]. By contrast, a comprehensive morphology-based study suggested that Clambidae is the sister group to the remaining polyphagan lineages [1]. Clambid adults are characterized by a combination of the following traits: body minute (usually less than 2 mm long); head usually strongly declined; antennae with 8–10 antennomeres and apical two antennomeres forming a distinct club; procoxal cavities slightly to strongly transverse; metacoxal plates enlarged and legs partly concealed; tri- or tetramerous tarsi; and abdomen with five or six free ventrites [4].

Fossils belonging to Clambidae are sparse. To date, only two fossil species of Clambidae have been formally described, and both are known from amber. Kirejtshuk & Azar [8] described the first and earliest known representative of Clambidae, *Eoclambus rugidorsum* Kirejtshuk and Azar, from Early Cretaceous Lebanese amber. The other clambid fossil species, *Clambus helheimricus* Alekseev, has just recently been reported from Eocene Baltic amber, although *Clambus* has been recorded from Baltic and Bitterfeld ambers for a long time [9]. Here, we report two new species of Clambidae from mid-Cretaceous Burmese amber. The new species, represented by five well-preserved fossils, can be attributed to the extant genera *Acalyptomerus* and *Sphaerothorax*, which have important biogeographic implications.

2. Results

(a) Systematic palaeontology

(i) Insecta

Order Coleoptera Linnaeus, 1758

Family Clambidae Fischer von Waldheim, 1821

Subfamily Acalyptomerinae Crowson, 1979

Genus *Acalyptomerus* Crowson, 1979

(Type species: *Acalyptomerus asiaticus* Crowson, 1979)

Acalyptomerus thayerae Cai and Lawrence sp. nov. (figure 1; electronic supplementary material, figures S1 and S2)

(ii) Diagnosis

The new species can be separated from other members of *Acalyptomerus* by having a remarkable spiny appearance; pronotum with long, stiff, posteriorly directed setae along lateral margins; lateral pronotal margin denticulate, forming long spikes basally; elytron with three regularly arranged rows of long setae on the disc and a row of similar setae along its lateral margin; and lateral elytral denticulation widely spaced, forming long spikes in the posterior third.

(iii) Etymology

The specific epithet is a patronym formed from the surname of Dr Margaret K. Thayer (Field Museum of Natural History,

Chicago), a well-known coleopterist working on rove beetles (Staphylinidae).

(iv) Type material, locality, and age

Holotype: NIGP168027, male. Paratypes: NIGP168028, NIGP168029, sex undetermined; completely preserved adults. All specimens are housed in the Nanjing Institute of Geology and Palaeontology, Chinese Academy of Sciences (CAS), Nanjing, China. Preserved in Burmese amber; absolute age 98.79 ± 0.62 Ma, established by U-Pb (uranium-lead) dating of zircons from the associated matrix of the unprocessed amber [10].

(v) Description

Body (figure 1; electronic supplementary material, figure S1*a*) 1.05–1.15 mm long, oval, convex. Head widest across compound eyes, 0.23 mm wide, slightly declined, finely punctate, and pubescent. Compound eye protuberant, evenly and symmetrically convex. Frontoclypeal sulcus straight. Antennal insertion concealed, close to the eye. Antenna (electronic supplementary material, figure S2*a*) with nine antennomeres; antennomeres 1 and 2 enlarged, much wider than antennomere 3; antennomere 3 elongate, as long as antennomere 9; antennomeres 3–7 successively shortened towards apex; antennomeres 8 and 9 forming a distinct club; antennomere 8 conical; antennomere 9 as wide as antennomere 8, suboval. Pronotum strongly transverse, 0.17 mm long and 0.55 mm wide, trapezoidal; posterior lateral angle scooped; lateral margin (electronic supplementary material, figure S2*b*) denticulate, with six long posterolaterally directed stiff setae, successively shortened apically. Disc pubescent, with anterior median elevation and a pair of posterior median depressions; with four slightly developed longitudinal carinae, two median carinae close to each other. Elytron (electronic supplementary material, figure S1*b*) slightly convex, 0.82 mm long and each 0.29 mm wide, lateral elytral margin visible along the entire length in dorsal view. Elytral marginal serration (electronic supplementary material, figures S1*c*, S2*c,e,f*) more spaced; left elytron (electronic supplementary material, figure S1*b*) with 18 or 19 long posterolaterally directed stiff setae and two much shorter setae at elytral base and apex; right elytron with 19 long stiff setae and two shorter setae at elytral base and apex. Elytral disc (electronic supplementary material, figure S1*a,c*) with three longitudinal setiferous carinae; inner carina incomplete, with 10 or 11 long, erect, posteriorly directed setae; middle carinae complete, with about 15 long, erect, posteriorly directed setae, setae at base shorter; outer carina with 10 or 11 erect posteriorly directed setae, setae at base shorter. Hind wing fully developed. Metaventrite with posteriorly directed setae. Metacoxal plate with about eight long setae. Legs (electronic supplementary material, figure S2*d*) slender and thin; tarsi (electronic supplementary material, figure S2*d,g*) elongate, tarsomere 1 longest, tarsomere 2 much shorter than tarsomere 1, tarsomere 3 slightly longer than tarsomere 2. Abdomen subtriangular; ventrites 1 and 2 each with four small median setae; ventrite 3 with eight large median setae; ventrite 4 with 12 setae along apical margin; ventrite densely setose.

Male: apical abdominal tergite (figure 1*g*) with about 12 small spines at apex; apical abdominal ventrite (figure 1*h*)

Figure 1. *Acalyptomerus thayerae* sp. nov. from mid-Cretaceous Burmese amber. (*a,b,d,e*) Under green epifluorescence; (*c,f*) under normal reflected light; (*g−i*) under transmitted light. (*a,c*) Dorsal view of holotype (NIGP168027). (*b*) Ventral view of NIGP168027. (*d,f*) Dorsal view of paratype (NIGP168028). (*e*) Ventral view of NIGP168028. (*g*) Enlargement of holotype (NIGP168027), showing apical abdominal tergite. (*h*) The same as (*g*), showing apical sternite. (*i*) Z-staked image, showing male internal genitalia, including parameres (pm) and median lobe (ml). (Online version in colour.)

coarsely sculptured, rounded apically; aedeagus (figure 1*i*) trilobate, symmetrical; median lobe lanceolate at the apex.

(vi) Remarks

These specimens are placed in Clambidae based on the following combination of characters: body small; head declined; antennae with nine antennomeres and apical two forming a club; metacoxal plates enlarged and legs partly concealed; and abdomen with five free ventrites [4]. They are referred to the extant genus *Acalyptomerus* based on the following features: (1) cuticular surface of the body rugose, (2) head hypognathous, visible dorsally, (3) compound eyes entire and strongly protuberant, not divided by a canthus,
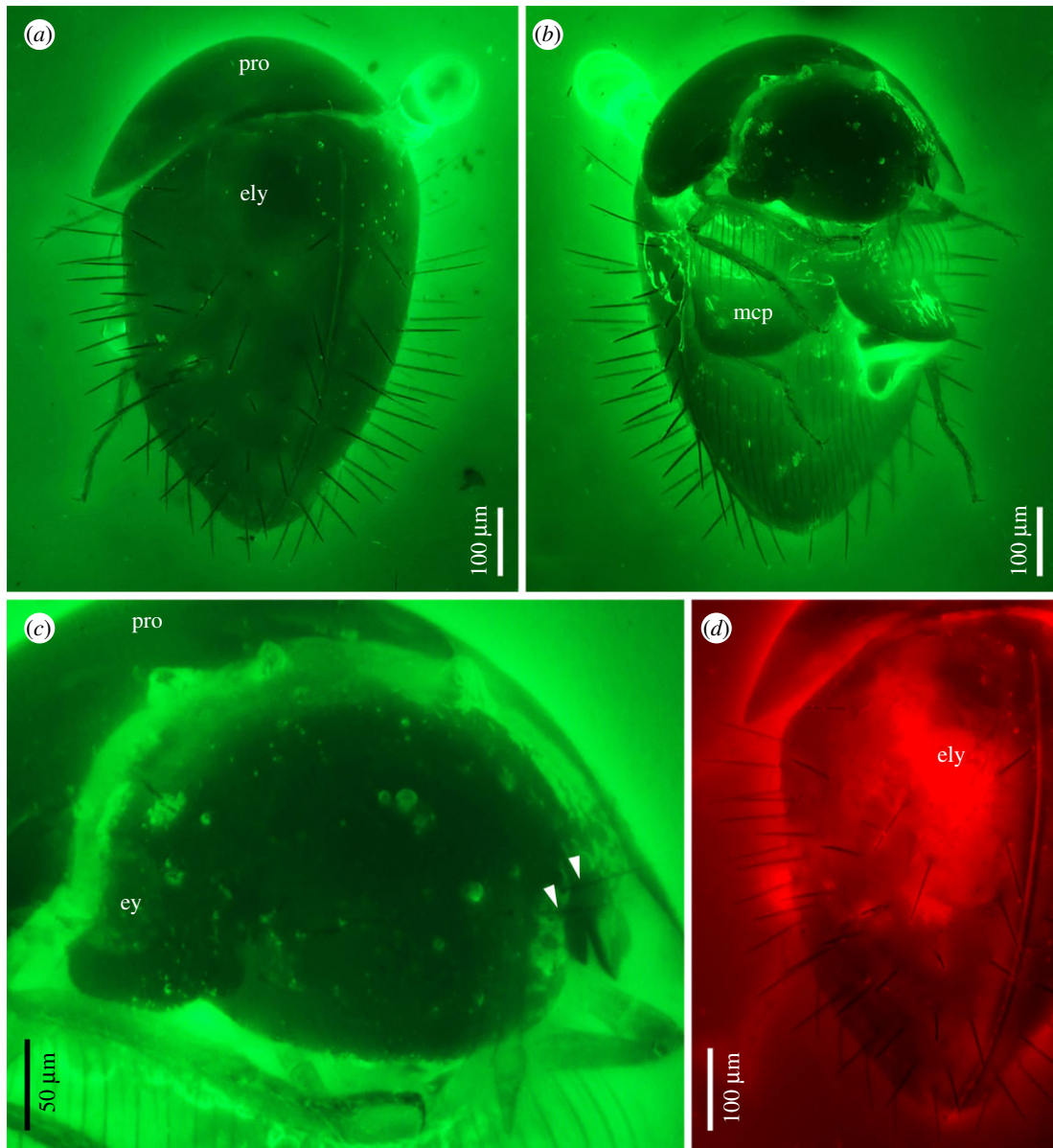
Figure 2. *Sphaerothorax uenoi* sp. nov. from mid-Cretaceous Burmese amber. (*a–c*) Under green epifluorescence; (*d*) under red epifluorescence. (*a*) Dorsal view of holotype (NIGP168930). (*b*) Ventral view of NIGP168930. (*c*) Details of head of holotype, showing unframed compound eyes. (*d*) Left elytron of holotype, showing dense strong setae. ely, elytron; ey, eye; mcp, metacoxal plate; pro, pronotum. (Online version in colour.)

(4) antennal insertions exposed from above, (5) sides of prothorax nearly straight, (6) procoxal cavities strongly transverse, (7) posterior angles of pronotum moderately acute, (8) pronotal disc with depressions, (9) mesoventrite carinate, (10) mesocoxal cavities narrowly separated, and (11) tarsal formula 3–3–3 [4,11]. Among the four extant species of *Acalyptomerus*, *Acalyptomerus thayerae* sp. nov. is probably closely related to *A. herbertfranzi* Endrödy-Younga, a species distributed in Costa Rica, Ecuador, Peru, and Venezuela [12]. They share a similar small body size, widely spaced lateral elytral denticulation, presence of spike-like processes in the posterior third of elytral margin, and presence of three longitudinal rows of semierect or erect long setae on the elytral disc. However, *A. thayerae* can be readily separated from *A. herbertfranzi* by having much stronger setae on the pronotum and elytra, especially on the lateral margins. As *A. herbertfranzi* has been described based on six females [12], males and genitalic morphology remain unknown. It is fortunate that the aedeagus of the holotype of *A. thayerae* is well preserved and clearly visible with transmitted light under a compound microscope. The aedeagus of *A. thayerae*

is of a typical trilobate type, and the median lobe (or penis) is lanceolate at apex, a trait similar to that of *A. africanus* Endrödy-Younga from Kenya & La Réunion [12].

    Subfamily Clambinae Fischer von Waldheim, 1821
    Genus *Sphaerothorax* Endrödy-Younga, 1959
    (Type species: *Clambus tasmani* Blackburn, 1902)
    Sphaerothorax uenoi Cai and Lawrence sp. nov. (figure 2; electronic supplementary material, figure S3)

(vii) Diagnosis

The new species can be separated from other members of *Sphaerothorax* by having the following combination of characters: (1) small body size (ca 0.71 mm long), (2) compound eye less delimited by a lateral lobe of head, and (3) elytron with approximately 40 long, strong, erect setae.

(viii) Etymology

The specific epithet is a patronym formed from the surname of Mr Teruhisa Ueno (Fukuoka, Japan), who kindly donated the paratype for our study.

(ix) Type material, locality, and age

Holotype: NIGP168930, male. Paratype: FMNHINS-3260641, sex undetermined. The holotype is deposited in the Nanjing Institute of Geology and Palaeontology, CAS, Nanjing, China; the paratype is housed in the Integrative Research Center, Field Museum of Natural History, Chicago, USA. Preserved in Burmese amber; earliest Cenomanian [10].

(x) Description

Body (figure 2a,b; electronic supplementary material, figure S3a–c) very small, ca 0.71 mm long (measured from the anterior pronotal margin and elytral apex in dorsal view), brown, strongly convex. Head (figure 2c) large, slightly convex. Clypeus slightly depressed, arcuate, with a few short setae. Frontoclypeal sulcus slightly curved. Lateral lobe of head nearly oblong, lateral margins nearly parallel. Anterior and posterior lateral angles broadly rounded. Compound eye not completely framed by a genal margin, relatively small, about half of its margin not delimited. Integument smooth and shiny, with two erect setae close to antennal insertion. Pronotum more convex than head; anterior lateral angle more rounded than posterior angle. Pronotal disc with at least one pair of small setae on the median part. Elytra (figure 2d) convex, 0.60 mm long and each 0.31 mm wide; with about 40 long and erect setae (electronic supplementary material, figure S3d), more densely set along lateral margin than elytral base. Metaventrite transverse; posterior part horizontal, slightly convex, with row of elongate posteriorly directed setae along the anterior margin of posterior part. Posterior horizontal plate of metaventrite short medially, much shorter along mid-length than lateral length. Metacoxal plates large, with scattered setae, as long as horizontal posterior part of metaventrite. Abdomen 5-segmented, each ventrite with a row of long posteriorly directed setae located near the posterior margin.

(xi) Remarks

The specimens can be attributed to Clambidae by the following combination of characters: body small, conglobate; head strongly declined; metacoxal plates enlarged and legs partly concealed; and abdomen with five free ventrites [4]. They can be placed in the extant genus *Sphaerothorax* based on the following combination of features: (1) head broad, with evenly arcuate clypeus, (2) compound eye partially unframed (synapomorphy), (3) pronotum with rounded lateral margin, (4) abdomen with five visible sternites, and (5) tarsi tetramerous (electronic supplementary material, figure S3e). *Sphaerothorax* comprises 13 extant species, currently occurring in Australia, Chile, and New Zealand [13–15]. The new species *S. uenoi* can be easily separated from its extant counterparts by a much smaller body size (0.71 mm long, from anterior pronotal margin and elytral apex). *Sphaerothorax zealandicus* Endrödy-Younga, with a body length of 0.92–1.00 mm (with head bent), is the smallest known extant species of *Sphaerothorax*, but it is distinctly larger than *S. uenoi*. Another character important for distinguishing the new species from extant species is the number of strong erect setae on the elytra. *Sphaerothorax uenoi* bears 40 long strong erect setae on each elytron, whereas extant ones have weaker and/or fewer erect setae ranging from about 20 (*S. pubiventris* (Lea) from Tasmania) to 35 (*S. tierensis* (Blackburn) from Australia and New Zealand) [13–15].

3. Discussion

(a) Systematic positions of *Acalyptomerus thayerae* and *Sphaerothorax uenoi*

The circumtropical *Acalyptomerus* currently comprises four extant species (*A. africanus*, *A. americanus*, *A. asiaticus* (electronic supplementary material, figure S4), and *A. herbertfranzi*) occurring in Asia, Africa, and Central and South America [4,12]. All extant species except *A. herbertfranzi* possess finely and densely denticulate pronotal and elytral margins, whereas *A. herbertfranzi* and the fossil species *A. thayerae* have widely spaced lateral elytral denticulations, forming long spikes posteriorly, which probably represents a derived character. Compared to *A. herbertfranzi*, *A. thayerae* appears to have several more derived features, including widely spaced lateral pronotal denticulations forming spikes basally, pronotum with a pair of large median depressions, and elytral disc with three longitudinal carinae. As such, the Cretaceous *A. thayerae* is probably a crown species of *Acalyptomerus*, which may be of importance for dating phylogenetic trees in the future.

The species of *Sphaerothorax* include two major evolutionary lineages: one comprises species with a short posterior plate of the metaventrite, and the other plesiomorphy-rich lineage being characterized by a long posterior plate of the metaventrite and symmetrical aedeagi [13,14]. Based on the morphology of the posterior metaventral plate, the fossil *Sphaerothorax uenoi* apparently belongs to the first comparatively derived lineage of *Sphaerothorax*, which currently occurs in Australia and New Zealand, but not in South America [15]. *Sphaerothorax* is characterized by its partially unframed compound eyes, which represent a transitional form from *Acalyptomerus* (compound eyes entire and protuberant) and *Calyptomerus* (compound eyes entire but not protuberant) to the more derived *Clambus* (compound eyes completely divided by a canthus) [4]. In *S. uenoi*, the compound eyes are more unframed or laterally free than those of the extant species, and this is clearly a plesiomorphic feature for the genus. Although it is challenging to determine whether the much smaller body size and denser erect setae on the elytra of *S. uenoi* are derived or not, the Cretaceous species appears to intermingle both ancestral and derived traits. Its exact systematic position within the genus needs further phylogenetic exploration by the extensive sampling of characters of all extant and extinct taxa.

Probably due to their tiny body size (less than 2 mm long), clambids are rarely known as fossils. The previous clambid fossil from the Mesozoic, being important for understanding the origin and early diversification of the family, is represented by one species (*Eoclambus rugidorsum*) from the Early Cretaceous (Barremian) Lebanese amber [8], at least 125 Ma [16]. *Eoclambus rugidorsum* is a member of the more advanced subfamily Clambinae as evidenced by its conglobate body, metaventrite with the sloping anterior part, and tetramerous tarsi. The discovery of a definitive clambid from Lebanese amber represents the oldest record for Clambinae and the entire family. This early date is congruent with recent molecular clock estimates, which indicated an origin of Clambinae (represented by *Clambus* and *Loricaster*) in the Middle/Late Jurassic [2,17]. Therefore, it is not surprising to discover the clambine genus *Sphaerothorax* from the mid-Cretaceous. As *Acalyptomerus* (subfamily Acalyptomerinae) occupies the
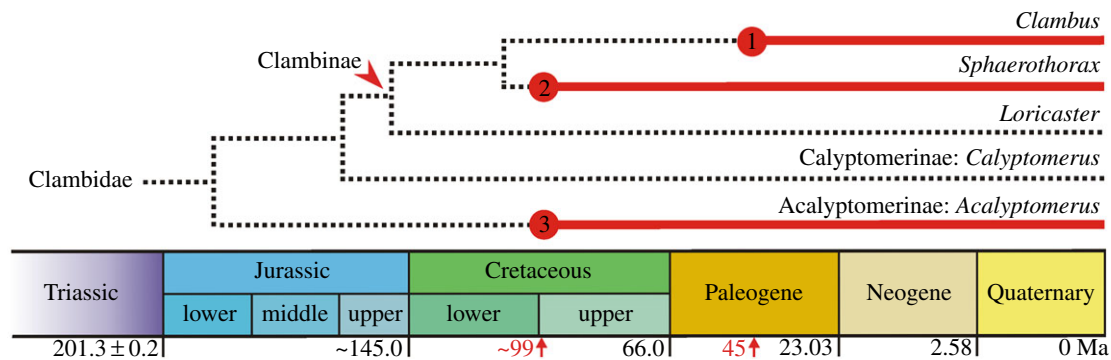
Figure 3. Phylogeny of all extant genera of Clambidae, modified from Leschen [4]. Geological scale after an updated version of the International Commission on Stratigraphy (ICS) International Chronostratigraphic Chart [18]. 1. *Clambus helheimricus* from Eocene Baltic amber [9]; 2. *Sphaerothorax uenoi* sp. nov. from mid-Cretaceous Burmese amber; 3. *Acalyptomerus thayerae* sp. nov. from Burmese amber. (Online version in colour.)

basal-most position among all known Clambidae, it is likely that Acalyptomerinae originated in the Early Jurassic [2] or even in the Late Triassic [3] (figure 3). As clambids are exceedingly small, usually less than 2 mm long, it seems difficult to find Triassic or Jurassic compression fossils of Clambidae, considering the usually poor preservation condition of compressions and a lack of fossiliferous amber from those early periods.

## (b) Systematic position of *Ptisma zasukae* Kirejtshuk and Azar

Kirejtshuk et al. [19] described a new family Ptismidae based on a single species, *Ptisma zasukhae* Kirejtshuk and Azar, from Lower Cretaceous Lebanese amber, and placed the new family in Staphylinoidea. They compared particular characters of the new taxon to a variety of staphylinoid groups, especially those of compact forms such as Ptiliidae and the staphylinid genera *Empelus* LeConte (Empelinae) and *Silphotelus* Broun (Proteininae), as well as to the scirtoid families Eucinetidae and Clambidae, among other groups. They also noted significant differences between their new family and all of those groups, although their characterization of the differences is often incomplete or erroneous. For example, the main 'staphylinoid' character cited for placement in that group (large, transverse, and laterally open mesocoxal cavities with exposed mesotrochantins) is also found (but not noted) in some of the other compared groups such as clambids; clambids were also said to differ in having 'lobed' tarsomeres 1–3 and a short metaventrite, but clambid tarsi are not lobed and the metaventrite can be large as in *Acalyptomerus* Crowson [4]. Although they failed to provide convincing evidence (synapomorphies) linking Ptismidae to any of the staphylinoids and based that placement mainly on superficial similarities or symplesiomorphies (e.g. the mesocoxal cavities), they tentatively placed the family in Staphylinoidea. Anton et al. [20], in a review of clambid morphology, already concluded in a brief note regarding Ptismidae that 'the strongly deflected head fitting with the venter of the anterior thorax, the abrupt two-segmented antennal club, and the broad and large metacoxal plates strongly suggest that the extinct taxon belongs in the family Clambidae', although they did not formally synonymize these families. We agree with this placement and propose that *P. zasukhae* can be undoubtedly placed in the extant family Clambidae by a combination of those diagnostic characters cited by Anton et al. [20] and additional ones, including

10-segmented antennae and tetramerous tarsi [4]. In addition, *P. zasukhae* bears a conglobate body, which represents a more derived feature than that of the basal-most *Acalyptomerus*. *Ptisma* intermingles both plesiomorphic (entire but not protuberant compound eyes as in *Calyptomerus*) and derived (conglobate body, thickened tibial apex with strong spurs, and thick setae) characters. Among all extant clambids, *Ptisma* is most closely related to *Calyptomerus* (Calyptomerinae). In addition to the similar conglobate body and entire eyes, they share a wide and subtriangular metanepisternum and an apical antennomere with columella-like sensilla. *Ptisma* differs from *Calyptomerus* in having tibiae strongly widened apically, with their apices bearing long spurs and strong setae, which is a derived character likely adapted for jumping, as in Eucinetidae [19]. Given the above discussions, *P. zasukhae* is a crown clambid closely related to the representatives of extant *Calyptomerus* and Ptismidae is therefore here formally regarded as a junior synonym of Clambidae (*new synonymy*).

(c) Biogeographic implications

Extant species of *Acalyptomerus* are widespread in tropical areas, including Asia, Africa, and Central and South America. In particular, the Cretaceous *A. thayerae* from Burmese amber has a close affinity to *A. herbertfranzi*, which is restricted to Costa Rica, Ecuador, Peru, and Venezuela [12] (figure 4). A very similar biogeographic distribution pattern can also be found in the myxophagan family Lepiceridae. Lepiceridae includes the sole genus *Lepicerus* Motschulsky with three extant species presently distributed in Mexico, Guatemala, Costa Rica, Panama, Venezuela, and Ecuador [21,22]. The extant species of *Lepicerus* are rarely collected, but they are comparatively common in Burmese amber, with three species belonging to *Lepicerus* formally described [23–25]. Like the lepicerid fossils, our discovery of an *A. herbertfranzi*-like species from the mid-Cretaceous of northern Myanmar suggests a relict distribution of the extant species presently known from Mesoamerica and northern South America. The discovery of *Acalyptomerus* in Burmese amber again suggests that the ancient amber forest was probably tropical [26], which has also been evidenced by the presence of multiple circumtropical arthropod lineages, including velvet worms (Onychophora: Peripatidae) [26,27] and Zoraptera [28–30].

Unlike *Acalyptomerus*, *Sphaerothorax* is currently confined to Australia, Chile, and New Zealand (figure 4). Our discovery of a Southern Hemisphere endemic genus from the
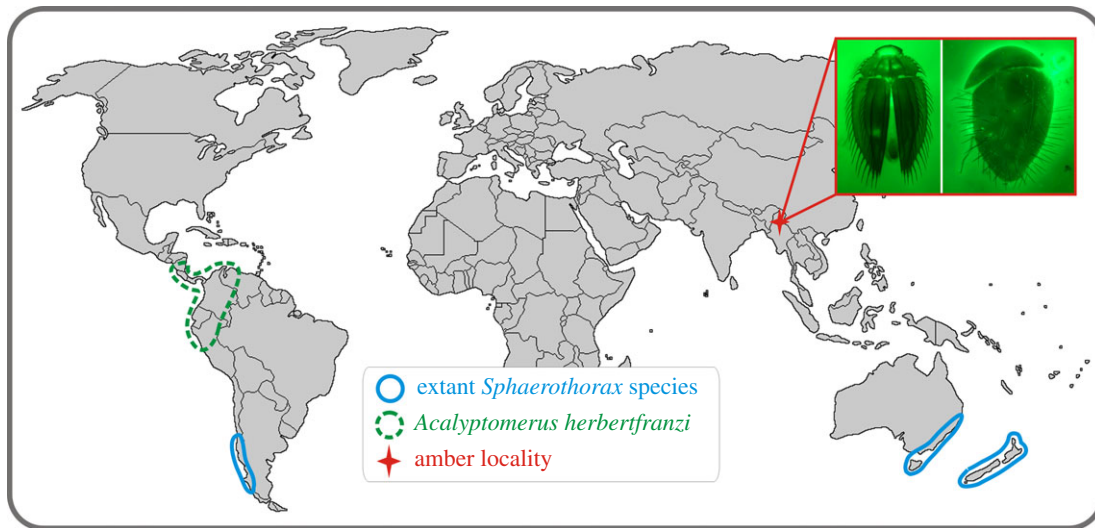
Figure 4. Geographical distribution of *Acalyptomerus thayerae* sp. nov. and *Sphaerothorax uenoi* sp. nov. and their related extant counterparts. (Online version in colour.)

presently northern Myanmar is not surprising, because such a distributional pattern has been exemplified by angiosperms and diverse arthropods from Burmese amber, and more importantly, the Burmese amber forest is suggested to have a Gondwanan origin [31]. Among all known fossil beetles from burmite, a similar pattern can be found in a tribe of Leiodidae (Agyrtodini), which have a disjunct distribution in Australia, Chile, New Zealand, and South Africa [32,33]. In addition, an increasing number of beetle groups from the austral fauna have been recently recorded from Burmese amber, such as the Australia endemic *Omma* of Ommatidae [34], the South America endemic rove beetle (Staphylinidae) subfamily Solieriinae [35], and the New Zealand endemic cucujoid family Cyclaxyridae [36].

Most extant species of *Sphaerothorax* are associated with *Nothofagus* forest, and it is suggested that not only this type of forest but also members of a complex biome associated with them, persisted since the separation of the Gondwanan continents [15]. *Nothofagus* (the southern beeches) is an iconic angiosperm genus with ancient Gondwanan roots reaching back into the Cretaceous [37]. The current transoceanic distribution of the genus received extensive attention and it has been regarded as a key genus in plant biogeography. The biogeographic data supported by both *Nothofagus* itself and associated animals suggests that transoceanic *Nothofagus* distributions probably resulted from early vicariance events and subsequent extinction [15,38]. However, this hypothesis has been challenged by molecular clock estimates, which showed evidence for long-distance dispersal of *Nothofagus* [37]. Although there are no fossils of Nothofagaceae known from Burmese amber, multiple possible *Nothofagus*-associated beetle lineages other than *Sphaerothorax* have been known from this source. The rove beetle subfamily Solieriinae includes a single extant species, *Solierius obscurus* (Solier), from the Valdivian temperate rainforest and *Nothofagus*-dominated forests of southern Chile and Argentina [35]. By contrast, Solieriinae are diverse and abundant in Burmese amber, with three species described and undescribed forms from slightly older Lebanese and Spanish ambers [35]. The New Zealand endemic Cyclaxyridae with one genus and two extant species is primarily associated with *Nothofagus*, feeding on sooty-mould fungi on the tree [39]. Recently, a fossil species belonging to the extant genus *Cyclaxyra* has been documented from Burmese amber [36]. There is no

doubt that these fossils suggest that certain small groups were once more widespread and current species are relicts. However, the origin of the establishment of associations between beetles and *Nothofagus* is difficult to test, but there is evidence indicating that some insects were probably secondarily associated with *Nothofagus*. Like *S. obscurus*, the scorpionfly family Eomeropidae contains only one species, *Notiothauma reedi* MacLachlan, occurring in the *Nothofagus* forests in southern Chile [40]. Eomeropid fossils are diverse, known from the Middle Jurassic to the late Eocene [41,42]. Particularly, the Middle Jurassic *Jurathauma simplex* Zhang *et al.* from northeastern China closely resembles the living species *N. reedi* [42]. Since the oldest undisputed angiosperms date back to the Early Cretaceous [43], the eomeropids apparently have a much longer evolutionary history than trees of Nothofagaceae. Collectively, the associations between insects and the *Nothofagus* forest may not be obligate, and *Nothofagus*-like forests were not necessarily present in the mid-Cretaceous of northern Myanmar.

(d) Long-term morphological stasis

Our discovery of two species belonging to extant genera provides two interesting examples of morphological stability over long-term geological time. The morphological stasis or bradytely in fossil insects is widely explained by the long-term persistence of the types of habitats the animals occupied [44–48]. Among fossil beetles reported from Burmese amber, the largely unchanged mesic microhabitats [49] may account for the 99 Myr morphological stasis [44,50]. In particular, although little is known about the natural history of extant clambids, specimens of *Acalyptomerus* and *Sphaerothorax* are usually collected from leaf litter or on vegetation [13,15]. Thus, similar mesic microhabitats for these clambids such as leaf litter may have persisted at least since the time of the Burmese amber forest. In addition to these clambids, Burmese amber has yielded multiple beetle species belonging to extant genera which changed very little over long evolutionary history. These beetles include the ommatid *Omma* [34], the jacobsoniid genera *Derolathrus* [48] and *Sarothrias* [51], the leiodid genus *Colon* [52] and *Colonellus* (Leiodidae) [53], diverse rove beetles belonging to *Megalopinus* (Megalopsidiinae) [54], *Octavius* (Euaesthetinae) [44] and *Oxyporus* (Oxyporinae) [55,56], the dermestid *Attagenus* [57–59], and

Microborus bark beetles (Curculionidae) [60]. It is obvious that the body plans of many extant beetle lineages have been established and remained almost unchanged from the mid-Cretaceous, some 99 Ma.

Molecular-based phylogenies indicated that Clambidae is among the basal and species-poor polyphagan lineages [2,3]. Clambids from the Mesozoic appear to be comparatively diverse, with at least four genera (two from Lebanese amber and two from Burmese amber). These fossil clambids also suggested a high level of morphological disparity early in the Cretaceous, as exemplified by the potentially jumping *Ptisma zasukhae* and the more spiny *Acalyptomerus thayerae*. Those exhibiting more specialized morphology, such as the Cretaceous *Ptisma* possibly having jumping ability, went extinct, whereas the morphologically unspecialized ecological generalists such as *Acalyptomerus* and *Sphaerothorax* persisted to the present, despite dramatic climatic changes over long geological time.

4. Material and methods

The Burmese amber specimens described here originated from the Hukawng Valley in Tanaing Township, Myitkyina District of Kachin State, Myanmar. The amber pieces were ground and polished in order to make all morphological features accessible for observation. The holotype (NIGP168027) and a paratype (NIGP168028) of *A. thayerae* are mounted between two microscopic coverslips with Canada balsam as the medium. The left side of the body of the other paratype (NIGP168029) is obviously distorted. Observations and photographs were taken using a Zeiss Axio Imager 2 compound microscope with an AxioCam MRc 5 camera attached. The Zeiss Axio Imager 2 microscope was equipped with a mercury lamp and specific filters for DAPI (4′,6-diamidino-2-phenylindole), eGFP (enhanced green fluorescent protein), and rhodamine. Photomicrographs with a green background were taken under the eGFP mode, and those with a red background were under the rhodamine mode. Fluorescence imaging technique is a useful tool to visualize strongly sclerotized inclusions in amber. The paratype of *S. uenoi* was photographed with an Olympus DP26 digital camera mounted on an Olympus BX50 stereomicroscope. Extended depth of field images was then digitally compiled using Zerene Stacker (v. 1.04) and Helicon Focus (version 5.3) software and arranged in Adobe Photoshop CS5.

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