Charuwat Taekul, Alejandro A. Valerio, Andrew D. Austin, Hans Klompen and Norman F. Johnson
Molecular phylogeny of telenomine egg parasitoids (Hymenoptera: Platygastridae s.l.: Telenominae): evolution of host shifts and implications for classification
Systematic Entomology, 2014; 39(1):24-35

© 2013 The Authors. Systematic Entomology published by John Wiley & Sons Ltd on behalf of The Royal Entomological Society This is an open access article under the terms of the Creative Commons Attribution License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

Originally published at:
http://doi.org/10.1111/syen.12032

http://creativecommons.org/licenses/by/3.0/

http://hdl.handle.net/2440/95969
Molecular phylogeny of telenomine egg parasitoids (Hymenoptera: Platygastridae s.l.: Telenominae): evolution of host shifts and implications for classification

CHARUWAT TAEKUL, ALEJANDRO A. VALERIO, ANDREW D. AUSTIN, HANS KLOMPEN and NORMAN F. JOHNSON

Abstract. Parasitoid wasps of the subfamily Telenominae (Hymenoptera: Platygastridae, Platygastridae) develop as immatures within the eggs of other insects (Lepidoptera, Hemiptera, Diptera and Neuroptera). Rearing records indicate that individual species are restricted to attack hosts within only one of these four main groups. We conducted a phylogenetic analysis of the group using sequence data from multiple genes (18S, 28S, COI, EF-1α) to assess the pattern of shifts among host groups and to test the monophyly of and relationships among genera and species-groups. Telenominae sensu Masner – that is, including only the nominate tribe Telenomini – is not monophyletic. Representatives of the Psix group of genera (Psix Kozlov & Lê and Paratelenomus Dodd) form a monophyletic group that is sister to Gryon Haliday (Sceletoninae: Gryonini) and are excluded from the subfamily. The remaining telenomines are monophyletic. The genus Phanuromyia Dodd and the crassiclava group of Telenomus Haliday, both recorded as parasitoids of planthopper eggs (Hemiptera: Auchenorrhyncha, Fulgoroidea), form a monophyletic group that is sister to all other telenomines exclusive of the Psix group. Twenty-nine species of the crassiclava and aradi groups of Telenomus are transferred to Phanuromyia as new combinations. Basal elements of the remaining species are all in groups reared from the eggs of true bugs (Heteroptera), primarily the stink bugs (Pentatomoidea) and seed bugs (Lygaeoidea). A shift to parasitism of lepidopteran eggs evolved within a single clade, occurring either one or two times. From this clade a small group of species, the Telenomus tabanivorus group, subsequently shifted to parasitism of egg masses of true flies (Tabanidae and Stratiomyiidae). Aholcus Kieffer and Platytenomus Dodd both belong to the clade of lepidopteran parasitoids and are considered as junior synonyms of Telenomus (new synonymy for Aholcus). The monophyletic status of the two core genera, Telenomus and Trissolcus could not be resolved using these data. The phylogenetic pattern of host shifts suggests comparisons among taxa that may be fruitful in elucidating mechanisms by which parasitoids locate their hosts, the proximate factors that determine the host range, and the changes in these factors that influence host changes.
Introduction

The mechanisms and evolution of host finding among parasitoid insects is an area of great significance given the important role such species play in regulating the populations of their hosts, both naturally and in biological control programmes. Wasps of the superfamily Platygastroidea (Hymenoptera; Fig. 1) are idiobiont endoparasitoids of the eggs of insects and spiders (Scelioninae, Teleasinae and Telenominae) or koinobiont parasitoids of the immature stages of Auchenorrhyncha (Hemiptera), Sternorrhyncha (Hemiptera) or Cecidomyiidae (Diptera) (most Platygastrinae and Sceliotrachelinae). The major groups within the superfamily show a clear pattern of host group specificity (Austin et al., 2005). However, because of the current paucity of well-supported hypotheses of phylogenetic relationship, the sequence of shifts from one host taxon to another is uncertain. The many species of the subfamily Telenominae are particularly diverse in their host relationships, having been recorded from Lepidoptera, Diptera, Neuroptera and the suborders Heteroptera and Auchenorrhyncha within the Hemiptera (summarized in Bin & Johnson, 1982; Kozlov & Kononova, 1983; Johnson, 1984a). Several species of telenomines have been shown to find their egg hosts by cueing in to semiochemicals produced by the adult host, in some cases synergized by chemicals released by the plant upon which the host feeds or oviposits (for example, Colazza et al., 2009; Moraes et al., 2009; Conti et al., 2010; Arakaki et al., 2011; Peñaflor et al., 2011). No telenomine species, to our knowledge, has been reared from hosts from more than one order. Therefore, the change from, for example, Heteroptera to Lepidoptera (or vice versa), seems to be an evolutionarily ‘difficult’ and significant step. Insights into how it takes place may emerge from comparisons of the chemosensory capabilities of closely related species of wasps that attack the two different groups of hosts. The goal of this work is to contribute to a better understanding of relationships among telenomines to facilitate such work and, in the process, test the monophyly of its genera, species groups and the subfamily itself.

The scope of the subfamily Telenominae is the subject of some disagreement. Kozlov (1970) and Kozlov & Kononova (1983) divided it into three tribes: Telenomini, Tiphodytini (with one genus, Tiphodytes Bradley) and Aradophagini (also monobasic, Aradophagus Ashmead). Masner (1972, 1976) and Masner & Huggert (1979) argued that the latter two tribes were misplaced because they possess metasomal laterosternites (absent in Telenomini). They treated Aradophagini and Tiphodytini as members of the subfamily Scelioninae and restricted Telenominae to the nominate tribe. The concept used here follows Masner’s (1976) restricted version. Within the subfamily 51 genus-group and 1057 species-group taxa have been described, of which 20 genera and 904 species are considered to be valid (Johnson, 2013). The bulk of species are placed in one of two genera, either Trissolcus Ashmead (179 valid species) or Telenomus Haliday (652 valid species), both of which are cosmopolitan. Of the remaining genera, 14 have only 1–2 described species. Modestly sized genera remaining are Eumicrosoma Gahan (13 spp.), Paratelenomus Dodd (13 spp.), Phanuromyia Dodd (9 spp.) and Psix Kozlov & Lé (22 spp.). Species of Trissolcus parasitize the eggs of stink bugs and their allies (Kozlov & Kononova, 1983; Johnson 1984a,b, 1987, 1991); Telenomus shares these hosts, but also attacks a wider range of Heteroptera, as well as Auchenorrhyncha, Lepidoptera, Diptera and Neuroptera (Bin & Johnson, 1982; Johnson & Bin, 1982; Kozlov & Kononova, 1983; Johnson, 1984a). The host records for Eumicrosoma are limited to seed bugs (Lygaeoidea: Blissidae) (Gahan, 1913; Ryu & Hirashima, 1989); Paratelenomus to turtle bugs (Pentatomoidea: Platarepidae) (Johnson, 1996); Phanuromyia to auchenorrhynchos Hemiptera (Issidae, Fulgoridae) (Johnson & Musetti, 2003); and Psix to Coreoidea and Pentatomoidea (Johnson & Masner, 1985).

Murphy et al. (2007) included seven representative species of the Telenominae in their molecular phylogeny of the Platygastroidea: one Phanuromyia, two Trissolcus, and four Telenomus. The subfamily emerged as a monophyletic group in their analyses, sister to the genus Gryon Haliday (Scelioninae: Gryonini, three species represented). Phanuromyia grouped together with Telenomus crassiclava at the base of the telenomine clade, and the two Trissolcus species grouped together at the apex. The remaining Telenomus species emerged as a paraphyletic basal grade in the subfamily between these two. Johnson (1985c, 1988a) grouped together the genera Psix, Paratelenomus, Nirupama Nixon and Madigere Johnson as the Psix group of genera. The relationship of this group to the bulk of Telenominae, however, was unclear. Johnson & Musetti (2003) recognized Phanuromyia as a distinct and valid genus, a conclusion subsequently rejected by Mineo (2006). Some progress in understanding relationships at a finer level has been made in the attempts to circumscribe natural groups within Telenomus (Kozlov & Kononova, 1983; Johnson, 1984a) and Trissolcus (Kozlov & Lé, 1976; Kozlov & Kononova, 1983; Johnson, 1984b, 1985a,b).

We report here the results of phylogenetic analyses of an extensive species sampling of Telenominae based on sequence data from four molecular markers: 18S and 28S rDNA, ...
the mitochondrial protein-coding gene COI, and the nuclear protein-coding gene EF-1α F2 copy. Also included in the analyses were a wider range of species of Gryon, designed to test the putative sister-group relationship or, if possible, to test whether Telenominae are more closely related to a subset of Gryon species. The results serve as a framework for interpreting the evolution of host relationships within the subfamily and, in addition, have repercussions for the classification of Telenominae.

Materials and methods

Taxonomic sampling and specimen vouchering

A total of 62 species were sequenced: 51 species for the ingroup Telenominae, and 11 species in the outgroups (File S1). The ingroup comprises representatives of six genera: Telenomus (eight species groups), Trissolcus (three species groups), Psix (three species), Paratelenomus (one species), Phanuromyia (two species) and Eumicrosoma (one species). The generic names Platytelenomus Dodd and Aholcus Kieffer are still recognized as valid by some authors (e.g. Kononova, 2008; Mineo et al., 2011). Representatives of each were also included: Platytelenomus spl and Telenomus dalmanni (Ratzburg), respectively. Other telenomine genera were not sampled because they are fossil taxa (e.g. Sinoprotelenomus Zhang) or are rare, and fresh material was unavailable. Within Trissolcus all three recognized major groups (Johnson, 1984b, 1985a,b) are represented by multiple species: thyanta group by Tr. thyanta Ashmead, Tr. THY1 and Tr. THY2; the flavipes group by Tr. euschiisti (Ashmead), Tr. strabus (Johnson) and Tr. urichi Crawford; and the basilis group by Tr. basilis (Wollaston), Tr. hulensis (Harrington), Tr. latisulcus (Crawford), Tr. BAS1, Tr. BAS2 and Tr. BAS3. The Australian species Tr. oggyes (Dodd) is an unusual species that cannot be placed in one of these species groups. For Telenomus, representatives of eight species groups are included: the floridanus group (parasitoids of Lygaeoidea) by Te. consimilis Ashmead, Te. nystorius Huggert, Te. FLO1, Te. FLO2 and Te. FLO3; the crassichlava group (Auchenorrhyncha) by Te. CRA1, Te. CRA2, Te. CRA3, Te. CRA4 and Te. CRA5; the podisi group (Pentatomoidea) by Te. podisi Ashmead, Te. grenadensis Ashmead and Te. sechellensis Dodd; the larici group (Miridae) by Te. LAR1, Te. LAR2 and Te. LAR3; the phymatae group (Reduviidae) by Te. dolichocerus (Ashmead); the californicus complex (Lepidoptera) by Te. dalmani, Te. CAC1, Te. CAC2, Te. CAC3, Te. CAC4 and Te. CAC5; the tabanivorus group (Diptera) by Te. goniopis Crawford and Te. TAB1; and the longicornis group (with unknown aquatic hosts) by Te. LON1, Te. LON2, Te. LON3 and Te. LON4. Morphological characterization and host relationships of each are from Johnson (1984a). Outgroup taxa include four genera of Scelioninae – six species of Gryon, two species of Scelio Latreille, and one species each of Oxyteleta Kieffer and Probaryconus Kieffer – and one species of Telesinae, Trimorus caraborum (Riley). Material studied was freshly collected from around the world.

After nondestructive DNA extraction the voucher specimens were deposited in the C. A. Triplehorn Insect Collection (OSUC). Supplementary File S1 lists the specimens, voucher information and GenBank accession numbers for markers included in the analyses.

DNA extraction, amplification, and sequencing

Fresh specimens were sorted from bulk material, preserved in 95% ethanol, and maintained in the freezer at −15°C. Nondestructive DNA extraction was performed using the DNeasy extraction protocol (Qiagen, Germantown, MD; cat. num. 69506) as modified by C.D. Zhu and J.S. Noyes (unpublished data): Individual specimens were initially softened in 70% ethanol at room temperature for 1 day. Vortexing in step 2 of the manufacturer’s protocol was modified by mixing the reaction gently and incubating at 55°C for 24 h. The mixture was stored at −20°C for 24 h. The intact specimen was then removed from the tube and prepared for standard mounting. The reaction was incubated for 10 min at 70°C after addition of Buffer AL. Then 200 μL of cold ethanol (96–100%) was added to the supernatant. Finally, in Step 7 the Buffer AE was warmed to 55–70°C before addition.

Four genes were targeted: 18S (positions 398 to 142) and the D2–D3 regions of 28S rDNA (nucleotide positions D2-3365 to D3-4413), the mitochondrial protein-coding gene cytochrome oxidase I (COI), and the F2 copy of the nuclear protein-coding gene elongation factor 1-alpha (EF-1α). Primers for PCR amplification were drawn from Whiting et al. (1997: ai, bi for 18S; D23, D25, F2R for 28S), Gribet et al. (1996: 18S-5R for 18S), Simon et al. (1994: 1F, 1R for COI), Folmer et al. (1994: HCO2198, LCO1490 for COI), Danforth et al. (1999: F2F, Cho10 for EF-1α), Heraty et al. (2011: F2F8, For3, F2R6 for EF-1α) and Simon et al. (2010: F7, F9 for EF-1α). Amplification was carried out via PCR following the protocols of Murphy et al. (2007) and Klompen (2000).

Nested PCR optimizations were employed to amplify the F2 copy of EF-1α. This approach is relatively successful in reducing or eliminating unwanted products, although it dramatically increases sensitivity to contamination. The initial PCR primers were F2F and Cho10 (Danforth et al., 1999) followed by a combination of other primers from Heraty et al. (2011): F2F8 [5′-CAA RTA TGC NTG GGY ATT GGY AAG-3′], F2R6 [5′-TGT WGC GRT GAA GTA TGC AGC NGC-3′], and Simon et al. (2010): EF-7 [5′-AAC AAT GTG ATC GYN ACN GAR CCN CC-3′] and EF-9 [5′-CCN ACN GGB ACH GTT CCR ATA CC-3′]. The choice of second round primers depended on the taxa and product results. Thermocycle conditions of F2F-Chol, F2F8-F2R6 and For3-Chol followed Klompen (2000) except that annealing temperatures of 54°C, 54°C and 58°C were used (respectively). The amplification profile for EF-7 and EF-9 followed Simon et al. (2010). We excluded the F1 copy from the analysis because of the difficulty of amplification and the often incomplete products (as in Heraty et al., 2011). Contaminants were detected using negative controls in all rounds.

© 2013 The Authors. Systematic Entomology published by John Wiley & Sons Ltd on behalf of The Royal Entomological Society. 39, 24–35
of PCR, as well as comparisons of the product sequences with each other and with a range of published arthropod sequences. PCR products were purified either using the QIAquick PCR purification kit (Qiagen) protocol or done prior to sequencing by Beckman Coulter Genomics (Danvers, MA). Products were sequenced in both directions and assembled using Sequencher v4.0 (Gene Codes Corporation, Ann Arbor, MI).

Sequence alignment

Sequences of COI (approximately 857 bp) were aligned initially using MUSCLE (Edgar, 2004) with default settings and then adjusted by eye. The total of 1124 bp of EF-1α were aligned by eye against a reference taxon (Archaeoteleia mellea – GenBank:GQ410731.1) and reported intron positions (Danforth & Ji, 1998; Heraty et al., 2011). Alignments of both protein-coding genes were translated to amino acids to verify the homology using Mesquite (Maddison & Maddison, 2011). Secondary structural alignments were implemented for ribosomal RNA sequences of 18S and 28S for a total of 1783 bp. The by-eye alignment conventions followed Kjer (1995) with slight modifications (Gillespie, 2004). The core alignments follow published secondary structure models of Hymenoptera (as in Gillespie et al., 2005; Murphy et al., 2007; Heraty et al., 2011). Ambiguous homology positions were defined across all taxa based on the structural criteria of Kjer (1995). These ambiguous regions were excluded from the final analyses. The final matrix alignment for analyses contains 3767 bp (COI: 857, 18S: 1003, 28S: 782, EF-1α: 1125). The alignment summary, gene partitions, nucleotide composition and percentage of parsimony-informative sites are presented in online supporting documentation.

Data coding and partitioning

Regier & Zwick (2011) discussed the role of heterogeneity in base composition and associated synonymous nucleotide change in protein-coding genes in reducing the performance of phylogenetic analyses, and they proposed mechanisms to attempt to compensate for this effect. Here we employ three strategies to reduce the effect of synonymous changes. The third codon position (nt3) is typically the source of most synonymous change and, therefore, one option is to eliminate (or completely degenerate) these data from the analysis. Additionally, synonymous changes in the first codon position are possible for sequences coding for arginine and leucine. The DEGEN approach (Regier & Zwick, 2011) degenerates synonymous nucleotide changes in both the first and third codon positions. The Perl code for DEGEN coding was modified to use the invertebrate mitochondrial genetic code for COI. The third strategy is to segregate data from nt3 in a separate partition.

We employed three partitioning schemes: (i) use of a single partition for data from all four markers; (ii) four partitions, separating data from 18S, 28S, COI and EF-1α; and (iii) six partitions for data from 18S, 28S, COI nt1+2, EF-1α nt1+2, COI nt3 and EF-1α nt3. Combining the coding and partitioning strategies resulted in seven datasets for analysis: (1–3) no coding, 1, 4 and 6 partitions; (4–5) nt3 excluded, 1 and 4 partitions; (6–7) DEGEN coding, 1 and 4 partitions.

Phylogenetic analyses

Three phylogenetic estimation strategies were implemented to infer relationships: maximum parsimony, maximum likelihood and Bayesian approaches. Maximum parsimony analyses were conducted in TNT v1.1 (Goloboff et al., 2008). Heuristic tree search algorithms were implemented using the New Technology Search by default setting with some modifications followed by tree bisection-reconnection (TBR) branch swapping. Some parameters were adjusted: using a sectorial search, treating gaps as missing data, tree drifting of four cycles, tree fusing of five rounds and best score hit of 20–25 times. The branch support was estimated using the Parsimony Jackknife (JK; Farris et al., 1996) executed with the setting 36% probability removal, ‘emulate JAC’ resampling, 1000 replications, ‘random addition sequences’ = 1, and ‘hold trees’ = 2 (Freudenstein et al., 2004; Klompen et al., 2007). The parsimony jackknife was selected over the alternative, bootstrapping, because it is less sensitive to missing and invariant characters (Farris et al., 1996; Freudenstein et al., 2004).

Maximum likelihood analyses were executed using RAXML v7.2.6 (Stamatakis, 2006; Stamatakis et al., 2008). By using GTRGAMMA for the model of nucleotide substitution (-m), individual θ-shape parameters, GTR-rates and empirical base frequencies were estimated and optimized for each partition. RAXML analyses were implemented using a rapid bootstrap and search for the best-scoring ML tree in one single program run. Heuristic hill-climbing tree searches were performed generated from 1000 distinct randomized maximum parsimony starting trees and computed for 1000 repetitions. An additional ten runs were made using random starting seeds: all resulted in the same topology and likelihood scores.

Bayesian analyses were performed using the parallel (MPI) version of MrBayes v3.2.1 (Ronquist et al., 2012) and the computations were conducted using the CIPRES Science Gateway (www.phylo.org). The evolutionary models used for each data partition were select by comparison of the Akaike Information Criterion (AICc) as calculated in jModelTest v0.1.1 (Posada, 2008), shown in File S2. The Markov Chain Monte Carlo (MCMC) parameters for combined data were as follows: ngen = 40 000 000 printfreq = 1000 samplefreq = 1000 nchains = 4 savebrlens = yes stoprule = yes stopval = 0.01. For each individual marker analysis, the MCMC was designed as ngen = 20 000 000 printfreq = 1000 samplefreq = 1000 nchains = 4 savebrlens = yes stoprule = yes stopval = 0.01.

Results

Data properties

Total DNA sequences of 1185–3322 bp (mean = 2253.5, median = 2242.5) from the four molecular markers were
successfully obtained for 78 taxa. Amplified PCR products varied in length: COI: 565–681 bp (with one short sequence of 195 bp), 18S: 549–974 bp, 28S: 595–802 bp, EF-1a: 277–1123 bp. Sequence characteristics of the four gene partitions are summarized in File S2. Parsimony analyses of the individual gene partitions showed a consistent amount of homoplasy in the data: 18S (150 trees, 192 steps, CI = 0.21, RI = 0.15); 28S (16 trees, 1025 steps, CI = 0.22, RI = 0.14); COI (80 trees, 3746 steps, CI = 0.16, RI = 0.18); and EF-1a (17 trees, 1873 steps, CI = 0.29, RI = 0.27) (trees not shown).

Relationships

The results are largely congruent among the three inference techniques. The parsimony results are, in general, less well-resolved and less well-supported. The level of support for clades discussed below in different analyses are presented in File S3. Parsimony analyses of the individual gene partitions showed a consistent amount of homoplasy in the data: 18S (150 trees, 192 steps, CI = 0.21, RI = 0.15); 28S (16 trees, 1025 steps, CI = 0.22, RI = 0.14); COI (80 trees, 3746 steps, CI = 0.16, RI = 0.18); and EF-1a (17 trees, 1873 steps, CI = 0.29, RI = 0.27) (trees not shown).

All of the remaining Telenominae species of Telenomus, Trissolcus, Eumicrosoma and Phanuromyia – together form a monophyletic group (bootstrap 85–98%, Bayesian posterior probability 98–100%). The two species of Phanuromyia are monophyletic (jackknife 50–53%, bootstraps 59–89%, Bayesian posterior probabilities 89–99%), and the Telenomus crassiclava species group is monophyletic in most analyses (jackknife 80–82%, bootstraps 68–90%, Bayesian posterior probabilities 84–98%). Phanuromyia is consistently the sister to the cluster of Telenomus crassiclava group species (bootstraps 59–89%, Bayesian posterior probabilities 89–95%). The remaining Telenominae species of Telenomus, Trissolcus, Eumicrosoma and Phanuromyia are monophyletic (bootstraps 54–93%, Bayesian posterior probabilities 84–98%). The species of Telenomus californicus complex, the large group within which the parasitoids of the eggs of Lepidoptera and Diptera are found, group together in all analyses. This group, represented by eight species, is not supported as a distinct clade because of the inclusion of one species of the floridanus group (Telenomus FLO3) and, sometimes, Te. sechellensis Kieffer (of the podisi group; Bayesian analysis, DEGEN coding). Representatives of the tabanivorus group, parasitoids of the eggs of horse flies, emerge as a monophyletic group within the clade including lepidopteran parasitoids (bootstraps 74–87%, Bayesian posterior probabilities 70–99%). The Te. longicornis (bootstraps 59–96%, Bayesian posterior probabilities 99–100%) and Te. laricis (bootstraps 95–100%, Bayesian posterior probabilities 100%) species groups were recovered as monophyletic groups in all coding and partitioning schemes.

Telenomus (Aholcus) dalmannii (Ratzeburg) and Platytelenomus always group with the species of the Te. californicus complex. The position of Eumicrosoma is unstable: it emerged either with the californicus complex (most analyses) or as sister to Trissolcus ogyges Dodd (Bayesian analysis, DEGEN coding, four partitions).

Neither Telenomus nor Trissolcus are resolved as monophyletic groups, even with the exclusion of the crassiclava species group from the former. Representatives of the thyanta group of Trissolcus emerge near the base of the Telenomus + Trissolcus + Eumicrosoma clade, but none of the three species groups of Trissolcus are monophyletic. The species assigned to the podisi and floridanus groups of Telenomus do not group together. The floridanus group is particularly dispersed, with one species, Te. FLO1, consistently placed at the base of the Telenomus + Trissolcus + Eumicrosoma clade (but with low support values).

Discussion

Scope of Telenominae

Telenominae has been one of the best-defined groups within the superfamily Platygastroidea since the subfamily was first formally recognized by C.G. Thomson (1860). It has been characterized by the combination of the following characters: metasoma with wide laterotergites loosely attached to the sterna; the absence of laterosternites on the metasoma; the second metasomal segment is significantly longer than any other; and the number of antennomeres in the female is reduced (10 or 11) in comparison to the male (12). Kozlov’s (1970) inclusion of Tiphodytini and Aradophagini was based solely on the broad laterotergites in these two (then) monobasic tribes, but these species possess laterosternites, and the antenna is 12-segmented in both sexes (Masner, 1972; Masner & Huggert, 1979). Masner’s (1976) restriction of the concept of the subfamily to the nominate tribe restored a clear definition of the taxon.

We believe that the results presented here strongly indicate that the subfamily, even in this restricted definition, is not monophyletic. We did not anticipate this outcome because the genera of the Psix group have the four synapomorphies of Telenominae. However, the position of these genera within the subfamily has been problematic. They all share a distinctive and plausibly apomorphic character: the central keel on the frons forks ventrally to pass on either side of the antennal insertions. Additionally, they all have strongly developed fanlike striae on the frons arising near the anterior mandibular articulation (very rarely present and only weakly developed in a few other telenomines), have a different structure of the metascutellum (the dorsellar lip of Johnson & Masner, 1985), and lack differentiated sublateral setae on the first metasomal.

© 2013 The Authors. Systematic Entomology published by John Wiley & Sons Ltd on behalf of The Royal Entomological Society. 39, 24–35
Fig. 2. Relationships of Telenominae derived from RAxML analysis of combined COI, 18S, 28S and EF-1α sequences with 1000 bootstrap replicates, DEGEN coding, single partition. Bootstrap values above 50% indicated on branches. The three-letter abbreviations for species of Telenomus and Trissolcus indicate the species-group to which it belongs: BAS, basalis; CAC, californicus; CRA, crassiclava; FLA, flavipes; FLO, floridanus; LAR, laricis; LON, longicornis; PHY, phymatius; POD, podisi; THY, thyantae. The position of the branches subtending the Psix group of genera and the Telenominae s.s. are indicated. Hosts of species in unshaded are various terrestrial Heteroptera, primarily Pentatomoidea and Coreoidea. Shaded areas highlight clades that have shifted to new groups of hosts.
tergite. Mikó et al. (2007) discovered that the two groups differ in that the pleural apodeme of the mesothorax is fused with the anterior margin of the speculum in Telenomus and Trissolcus, whereas in Psix and Paratelenomus the apodeme extends to the mesopleural articulation. Finally, the Psix group of genera lack two previously unreported, apparent synapomorphies of the other telenomines. First, the metapostnotum is delimited posteriorly by a deep sulcus that arises anteriorly from the propodeal spiracle and continues posteriorly and medially beneath the metascutellum (see, e.g., figs 18, 48 in Johnson, 1984a). Reexamination of newly collected specimens of Mudigere confirms this character, contradicting the statement in Johnson (1984a). Second, the labrum is large, visible externally, articulated with the external ventral margin of the narrow clypeus (i.e. not on the internal surface within the buccal cavity), and is almost always separated from the clypeus by a distinct suture.

The molecular data not only reaffirm the unity of the Psix group, but indicate that they are more closely related to Gryon than to other telenomines. Thus, the defining characters of the subfamily must be reinterpreted as having evolved independently in the Psix group of genera and in the Telenominae s.s. Most of these features are also found in other lineages within the superfamily. The reduction in number of female antennomeres is also found in the Gryonini genus Maruzza Mineo, and commonly occurs in other parts of the subfamily Scelioninae. Wide laterotergites are characteristic of species of Tiphodytes and Aradophagus, as well as males of Baeus Haliday. The size dominance of the second metasomal segment is also found among some species of Gryon, in more distantly related genera such as Yankara Galloway and Baeus, as well as in the subfamilies Platygastroinae and Sceliotrachelinae. The last remaining character is the loss of laterosternites in the metasoma. This is, we believe, one strong piece of morphological evidence supporting the monophyly of Telenominae sensu Masner because it occurs so rarely elsewhere in Platygastroidea. However, it stands in stark contrast to the molecular evidence we present here.

We conclude that the taxonomic concept of Telenominae must be modified. Two options are available, consistent with our desire that any formally recognized taxon be monophyletic. One is to expand the concept of Telenominae to include the genus Gryon and, by implication, closely related genera. The second is to exclude Psix and Paratelenomus – and by extension the closely related Mudigere and Nirupama – from the subfamily. We prefer the latter option. From a practical point of view, to include Gryon in Telenominae would put us in the position of having no morphological characters by which to recognize the subfamily. Furthermore, the design of this study may be inadequate to support such a conclusion. The analyses lead to the conclusion that the Psix group is more closely related to species of Gryon than to other species of Telenominae, but do not necessarily demonstrate that Gryon, out of all other genera of Platygastroidea, is the group to which it most closely related overall. We therefore prefer the option of a further contraction and refinement of the subfamily Telenominae. In this concept the subfamily can be defined on the basis of the presence of broad laterotergites, absence of laterosternites, sexual heterogeneity in antennomere number, as well as the presence of externally demarcated metapostnotum and labrum. We consider the Psix group of genera to belong to the subfamily Scelioninae.

**Phanuromyia**

Within Telenominae s.s. the data support the merging of Phanuromyia together with the species of the crassiclava group of Telenomus. Phanuromyia languished in obscurity for nearly 90 years after it was originally described (Dodd, 1914a) because it had been defined only on the basis of the exserted ovipositor (a highly variable character even among individuals of the same species) and the type species, P. rufobasalis, had not been studied. Johnson & Musetti (2003) offered additional characters (claval formula, head shape, absence of a hyperoccipital carina, orientation of the malar sulcus and size of the gena) that could be used to recognize and define Phanuromyia, specifically to distinguish it from the crassiclava group. Mineo (2006) subsequently rejected this on the grounds that these characters ‘... are generally used to advocate a species group of either Telenomus or other scelionid wasps.’ We cannot adopt the implication that some characters are *a priori* acceptable to define genera whereas others are only of value in defining a group of species. The critical issues are whether a new taxon is at least plausibly monophyletic and what effect the recognition of the new taxon would have on the monophyly of other recognized genera. Phanuromyia, now in an expanded concept to include the crassiclava group, is demonstrated in these analyses to be a monophyletic group, and its recognition does not result in other existing genera becoming paraphyletic. To follow Mineo’s position of synonymizing Phanuromyia with Telenomus, while maintaining monophyly, would require the inclusion of all of the genus Trissolcus as well as any related genera. We believe that the requirement of monophyly is the only objective criterion by which to choose among alternative classifications. As such, it seems that the best option at this point is to treat Phanuromyia as a valid genus. We could treat the crassiclava group as an independent genus, but with this level of taxon sampling, it seems more prudent to recognize only a single genus.

The Telenomus aradi group of Kozlov & Kononova (1983) is essentially the same as the crassiclava group of Johnson (1984a) and, therefore, should also be subsumed within Phanuromyia. The expansion of the generic concept of Phanuromyia to include species of the crassiclava and aradi groups results in the following generic transfers: *P. affinis* (Kozlov & Kononova) **comb.n.**, *P. amazonica* (Cameron) **comb.n.**, *P. aradi* (Kozlov) **comb.n.**, *P. aspera* (Kozlov & Kononova) **comb.n.**, *P. caucasica* (Kozlov & Kononova) **comb.n.**, *P. clavata* (Kozlov & Kononova) **comb.n.**, *P. corticata* (Kozlov & Kononova) **comb.n.**, *P. crassiclava* (Nixon) **comb.n.**, *P. cyane* (Kozlov & Lé) **comb.n.**, *P. flaviventris* (Kozlov & Kononova) **comb.n.**, *P. impressa* (Ashmead) **comb.n.**, *P. infuscatisipes* (Ashmead) **comb.n.**
P. jugoslavica (Szabó) comb.n., P. longiceps (Kozlov) comb.n., P. longistriata (Kozlov) comb.n., P. longivenetris (Cameron) comb.n., P. macalipennis (Ashmead) comb.n., P. marshakovi (Kozlov & Kononova) comb.n., P. meridiana (Kozlov & Kononova) comb.n., P. minima (Kozlov) comb.n., P. minuscula (Kozlov & Kononova) comb.n., P. nioba (Kozlov & Kononova) comb.n., P. picta (Kozlov) comb.n., P. propinqua (Kozlov & Kononova) comb.n., P. proxima (Kozlov & Kononova) comb.n., P. rubella (Kozlov & Kononova) comb.n., P. sphiogis (Ashmead), comb.n., P. taurus (Johnson) comb.n., and P. tuberculus (Kozlov & Kononova) comb.n. According to Article 59.4 of the International Code of Zoological Nomenclature the replacement names Telenomus russiianus Öz dikmen and T. moldovianus Öz dikmen, proposed for T. impressus Kononova and T. minimus Kozlov respectively by Öz dikmen (2011) for junior homonyms in Telenomus, are abandoned in favour of the original epithets (syn.n, stat.n.).

Most species of Phanuromyia may be morphologically recognized among Telenominae s.s. by the presence of a welldeveloped sternalus on the mesepisternum. Typically this is indicated as a line of deep foveae (the episternal foveae of Johnson, 1984a), but sometimes only as a crease in the sclerite. In this genus the sternalus arises anteriorly near the dorsal apex of the acetabular carina and continues obliquely dorsally toward the mesepisternal pit. The great majority of species may also be recognized as Phanuromyia by the convex frons (frontal depression limited to a small area just dorsal of the antennal insertions), the presence of two parallel dorsoventral lines of setae below the median ocellus, eyes glabrous or with very short setation, second metasomal tergite with shallowly incised reticulate sculpture posterior to the basal longitudinal costae.

Status of Aholcus

Whereas females over 90% of telenomine species have 11-segmented antennae, 70 species of Telenomus have been described in which this number is reduced to 10. Kieffer (1913) created the genus Aholcus for a single species from Kenya, A. monticola Kieffer. The same year, Dodd (1913) erected the genus Neotelenomus for five Australian species. In both cases, the only character differing from Telenomus was the number of female antennomeres. Nixon (1935) synonymized Aholcus with Telenomus, but later reversed his finding and treated it as a valid subgenus (Nixon, 1937). In the latter paper he synonymized Neotelenomus with Telenomus (Aholcus). Since then, Aholcus has been used as a valid name, both as a subgenus and, occasionally, as a genus in 20 publications (including Nixon, 1937) for 31 newly described species. Johnson (1988b) proposed that the same name had been used for two distinct groups of species: one, represented by Dodd’s type species, P. planus, has elongate clavomeres and corresponds to the floridanus group of Telenomus; the second has transverse clavomeres and parasitizes the flattened eggs of grass-feeding Lepidoptera (hence the flattened body). The species used in this study, on the basis of morphology, falls in the latter group. Consistent with Johnson (1988b), this species clusters with the other moth parasitoids of the californicus complex, and not with the species of the floridanus group. Therefore, we reaffirm the conclusion of Johnson (1988b) and consider Platytelenomus Dodd, 1914 to be a junior synonym of Telenomus Haliday (syn.n), 1833 and reject its use as a subgenus.

Status of Platytelenomus

The name Platytelenomus is less commonly encountered than Aholcus, but its history is similar in many respects. The genus was originally described by Dodd (1914b) for an Australian species in which the body is strongly flattened dorsoventrally. Johnson (1988b) proposed that the same name had been used for two distinct groups of species: one, represented by Dodd’s type species, P. planus, has elongate clavomeres and corresponds to the floridanus group of Telenomus; the second has transverse clavomeres and parasitizes the flattened eggs of grass-feeding Lepidoptera (hence the flattened body). The species used in this study, on the basis of morphology, falls in the latter group. Consistent with Johnson (1988b), this species clusters with the other moth parasitoids of the californicus complex, and not with the species of the floridanus group. Therefore, we reaffirm the conclusion of Johnson (1988b) and consider Platytelenomus Dodd, 1914 to be a junior synonym of Telenomus Haliday, 1833.

Position of Eumicrosoma

This name encompasses a small group of species found worldwide that parasitize the eggs of blisssine bugs. The genus is quite distinctively defined by its dorsoventrally depressed body, elongate lanceolate wings, elongate marginal and
The speciose genera: Telenomus and Trissolcus

The genera that today go by the names Telenomus and Trissolcus were first distinguished by C.G. Thomson (1860) as Phanurus Thomson and Telenomus, respectively. Most species were recognized by comparison of the sculpture of the frons: absent in Telenomus, present in Trissolcus; eye setation: long in Telenomus, short or absent in Trissolcus; development of notauli: almost always absent in Telenomus, commonly found in Trissolcus; shape of the second metasomal tergite: longer than wide in Telenomus, wider than long in Trissolcus; and number of female clavomeres: most Telenomus with five or fewer, Trissolcus with six. In general, species of Trissolcus are robust and stout-bodied, whereas many Telenomus species are elongate and gracile. None of these is without exception, but in concert they leave few species that cannot be unambiguously placed under one name or the other. The monophyly of these taxa, however, has never been demonstrated. The morphological diversity among species that has made it so difficult to unambiguously diagnose genera is also reflected in the attempts to delimit groups of species within them. We began this study not with the expectation that either Telenomus or Trissolcus would turn out to be monophyletic, but, rather, with the hope that the relationships among the species groups would provide the scaffold upon which to build a better understanding of the phylogeny of the subfamily.

Our data fail to settle decisively the question of the monophyly of either of the two core genera of the subfamily, Trissolcus and Telenomus. Further, though, none of the three species groups of Trissolcus were recovered in these analyses. At least one of these, the flavipes group, is very homogeneous and distinctive in morphology and, therefore, seems likely on those grounds to be monophyletic. The basal position of two of the species of the thyanta group is consistent with our expectations based on structure – for example, presence of notauli, presence of sublateral setae on the first metasomal tergite, and presence of setae on the posteroventral metapleuron.

We were able to recover some of the expected structure among the Telenomus species represented. Particularly, the placement of the crassiclava group distinctly apart from other Telenomus is significant. Among the remaining groups, the podisi and floridanus groups were not found to be monophyletic. As discussed in the section on Eumicrosoma, species of the floridanus group are scattered throughout the phylogeny. The only Old World species of the podisi group in the study, Te. sechellensis Kieffer, is always more closely related to the californicus complex than with others of its putative group. This result is corroborated by morphological evidence, specifically the same character of the occipital carina described above for Te. monticola. This feature is found in all Old World Telenomus parasitoids of stink bugs as well as in all species of the californicus complex, and it is lacking in all New World members of the podisi group. Consistent with the hypotheses in Johnson (1984a) the species of the californicus complex grouped together in a single clade, from which emerges the tabanivorus species group. These are all united by the structure of the metascutellum (evenly produced, coarsely sculptured dorsally, longitudinally striate ventrally) and the lower course of the occipital carina. The laricis and longicornis groups are also monophyletic, and most species are fairly easily recognized on the basis of morphology. The laricis group is characterized by the greatly enlarged gena and elongate body; those of the longicornis group have a strongly pigmented basal vein on the forewing, distinctive male genitalia with an elongate, spatulate aedeagal lobe, and a distinctively convex mesosoma.

Host relationships

Biological data have been acquired for 16 of the 51 ingroup species (31%). Thus, our assumptions of the host relationships and specificity should be viewed with some caution. Nevertheless, the number of host records for telenomines overall is very substantial. With that caveat in mind, it appears that the combined clade of Gryon, the Psix group, and Telenominae s.s. evolved from an ancestral species that probably parasitized the eggs of terrestrial Heteroptera. Hosts in the Pentatomoidae, Coreoidea, Reduvioidae and Lygaeoidea are shared among these three taxa. Paratelenomus has only been recorded as a parasitoid of turtle bugs (Pentatomoidae: Plataspidae), whereas species of Psix have been reared from a number of species of Pentatomodea as well as Coreoidea. For the basal groups of Telenominae s.s., with the exception of Phanuromyia, the most common families of hosts are the stink bugs and their allies in Pentatomoidea, particularly the Pentatomidae and Scutelleridae. This is true of all three of the species groups of Trissolcus, as well as the Telenomus podisi group (represented by Te. sechellensis, Te. grenadensis and Te. podisi). Species of the floridanus group of Telenomus, parasites of Lygaeidae, also arise at or near the base of the sister group of Phanuromyia. These include Te. nystivos, Te. consimilis and three unidentified species. The single species of Eumicrosoma in this study is from Thailand and its host is as yet unknown.
However, three species of the genus, *E. beneficial Gahan, E. bicolor* (Ashmead) and *E. blissae* (Maki) are all well known as parasitoids of chinch bug eggs (*Blissus, Ischnodemus*: Heteroptera: Lygaeidae). *Gryon* is a very large genus of Scelioninae (331 valid species). A few species have been recorded as having been reared from eggs of Lepidoptera, but the vast majority parasitizes the same groups of Heteroptera as these telenomines (with the exception of Blissiidae).

The parasitism of the eggs of auchenorrhynchous bugs—planthoppers of the families Fulgoridae and Issi-daе are the only hosts so far recorded—appears to have evolved but once, in *Phanuromyia* (interpreted as including the *T. crassiclava* and *aradi* groups). *Telenomus* has often been described as a group composed primarily of parasitoids of the eggs of moths and butterflies (e.g. Masner, 1976). Certainly a good number of species have this biology, and these are commonly encountered. Yet the host relationships of the genus are much more diverse. Parasitism of the eggs of Lepidoptera has only been elsewhere recorded in the Platygastroidea for two species of *Gryon*. Clearly, the shift(s) to this pool of hosts was evolutionarily significant, at least for diversification within *Telenomus*. On the basis of our results this shift occurred once or twice, depending on the interpretation of the position and putative host relationships of *Telenomus* FLO3 within the clade corresponding to the *Te. californicus* complex. Then, from a lepidopteran parasitoid, there was a subsequent shift in the *Te. tabanivorus* group to attack the eggs of some groups of horse flies (Diptera: Tabanidae) and soldier flies (Diptera: Stratiomyiidae). In a separate lineage are the *laricis* group of *Telenomus*; one representative of the *phymatae* group, *Tr. dolichocerus* (Ashmead); and the *longicornis* group of *Telenomus*. The first of these, the *laricis* group, has few host records, but these are limited to the plant bugs (Miridae), including the economically important tarnished plant bug, *Lygus lineolaris* (Palisot de Beauvois). *Telenomus dolichocerus* is a parasitoid of Reduviidae. The hosts of the *longicornis* group are as yet unknown. However, the species are somehow associated with water: they are commonly found alongside ponds and streams and have even been collected in floating emergence traps. Marshall (2006) asserted that one species in this group parasitizes the eggs of *Rhagovelia obesa* Uhler (Heteroptera: Veliidae), but supporting data have not been published, and we have never seen specimens so labelled. Certainly, given the position of the *longicornis* group, in a clade of heteropteran parasitoids, either Gerridae or Veliidae would be good candidates for hosts.

In summary, the sequence of host shifts that best fits the results presented here begins from a ground-plan host of some group of terrestrial Heteroptera. From there, one clade, *Phanuromyia*, moved on to the Auchenorrhyncha. Its sister group went in two different directions, with one subgroup (the *californicus* complex) shifting first to Lepidoptera and then to Diptera, whereas a second group shifted to Miridae (the *laricis* group), Reduviidae and a group of aquatic or semiaquatic insects, probably Heteroptera.

Although we have discussed host relationships in terms of taxonomic groups, obviously the parasitoids are using other cues in order to locate suitable eggs to parasitize. Host names merely serve as proxies for the real sensory modalities involved, such as long- or short-range chemicals (e.g. Colazza et al., 2007, 2009; Peri et al., 2007; Moraes et al., 2009; Salerno et al., 2009), tactile cues (e.g. Laumann et al., 2007), or possibly vision (Laumann et al., 2011). Determination of the breadth of host range and shifts from one set of hosts to another would seem involve either an overlap in the cues directly or indirectly produced by a host species, or a change in the capacity or responses of parasitoids to these stimuli. Research on the proximate mechanisms of host-finding in telenomines has been focused on species that are parasitoids of important pests such as *Trissolcus basalis*, *Telenomus podisi*, *Te. busseolae* (Gahan) and *Te. remus* Nixon. A phylogenetic hypothesis is sorely needed to be able to frame questions about the evolution of host relationships and to appropriately integrate the findings from these several species. The results presented here are a first step in that direction, providing support for some hypotheses and highlighting those areas in need of more intense future work.

**Supporting Information**

Additional Supporting Information may be found in the online version of this article under the DOI reference: 10.1111/syen.12032

**File S1.** List of taxa and sequenced markers for phylogenetic analysis of Telenominae: GenBank accession numbers and identifiers for specimens sequenced.

**File S2.** Alignment summary, gene partitions, nucleotide composition and percentage of parsimony informative sites.

**File S3.** Support values for selected clades across analyses.

**Acknowledgements**

This work could not have been completed without the critical contributions of L. Masner, L. Musetti and J. Cora; thanks also to M. Yoder, R. Burks, S. Hemly and E. Talamas and the journal reviewers. This material is based upon work supported in part by the National Science Foundation under grant No. DEB-0614764. C.T. was supported by a fellowship from the Agricultural Research Development Agency, Ministry of Agriculture and Cooperatives, Thailand.

**References**

Arakaki, N., Yamazawa, H. & Wakamura, S. (2011) The egg parasitoid *Telenomus eucryptidis* (Hymenoptera: Scelionidae) uses sex pheromone released by immobile female tussock moth Orgyia postica (Lepidoptera: Lymantriidae) as kairomone. *Applied Entomology and Zoology, 46*, 195–200.

Austin, A.D., Johnson, N.F. & Dowton, M. (2005) Systematics, evolution, and biology of scelionid and platygastroid wasps. *Annual Review of Entomology, 50*, 553–582.

© 2013 The Authors. *Systematic Entomology* published by John Wiley & Sons Ltd on behalf of The Royal Entomological Society. 39, 24–35
Bin, F. & Johnson, N.F. (1982) Potential of Telenominae in biocontrol with egg parasitoids (Hym., Scelionidae). Les Colloques de l’INRA, 9, 275–287.

Colazza, S., Aguila, G., Peri, E. & Millar, J.G. (2007) The egg parasitoid Trissolcus basalis uses n-nonadecane, a cuticular hydrocarbon from its stink bug host Nezara viridula, to discriminate between female and male hosts. Journal of Chemical Ecology, 33, 1405–1420.

Colazza, S., Lo Bue, M., Lo Giudice, D. & Peri, E. (2009) The response of Trissolcus basalis to footprint contact kairomones from Nezara viridula females is mediated by leaf epicuticular waxes. Naturwissenschaften, 96, 975–981.

Conti, E., Salerno, G., Leonbruni, B., Frati, F. & Bin, F. (2010) Short-range allelochemicals from a plant-herbivore association: a singular case of oviposition-induced synomone for an egg parasitoid. Journal of Experimental Biology, 213, 3911–3919.

Danforth, B.N. & Ji, S. (1998) Elongation factor-1α occurs as two copies in bees: implications for phylogenetic analysis of EF-1α sequences in insects. Annals of the Entomological Society of America, 91, 387–391.

Danforth, B.N., Saquet, H. & Packer, L. (1999) Phylogeny of the bee genus Halicus (Hymenoptera: Halictidae) based on parsimony and likelihood analyses of nuclear EF-1α sequence data. Molecular Phylogenetics and Evolution, 13, 605–618.

Dodd, A.P. (1913) Australian Hymenoptera Proctotrypoida. No. 1. Transactions of the Royal Society of South Australia, 37, 130–181.

Dodd, A.P. (1914a) Further new genera and species of Australian Proctotrypoida. Proceedings of the Royal Society of Queensland, 26, 91–140.

Dodd, A.P. (1914b) A new proctotrypod genus from Australia (Hym.). Entomological News, 25, 126–127.

Edgar, R.C. (2004) MUSCLE: multiple sequence alignment with high accuracy and high throughput. Nucleic Acids Research, 32, 1792–1797.

Farris, J.S., Albert, V.A., Källersjö, M., Lipscomb, D. & Kluge, A.G. (1996) Parsimony jackknifing outperforms neighbor-joining. Cladistics, 12, 99–124.

Folmer, O., Black, M., Hoeh, W., Lutz, R. & Vrijenhoek, R. (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. Molecular Marine Biology and Biotechnology, 3, 294–299.

Freudentstein, J.V., Berg, C., Goldman, D.H., Kores, P.J., Molvray, M. & Chase, M.W. (2004) An expanded plastid DNA phylogeny of Orchidaceae and analysis of jackknife branch support strategy. American Journal of Botany, 91, 149–157.

Gahan, A.B. (1913) New Hymenoptera from North America. Proceedings of the United States National Museum, 46, 431–443.

Gillespie, J.J. (2004) Characterizing regions of ambiguous alignment caused by the expansion and contraction of hairpin stem-loops in ribosomal RNA molecules. Molecular Phylogenetics and Evolution, 33, 936–943.

Gillespie, J.J., Munro, J.B., Heraty, J.M., Yoder, M.J., Owen, A.K. & Carmichael, A.E. (2005) A secondary structural model of the 28S rRNA expansion segments D2 and D3 for chalcidoid wasps (Hymenoptera: Chalcidoidea). Molecular Biology and Evolution, 22, 1593–1608.

Giribet, G., Carranza, S., Bagui, J., Riutort, M. & Ribera, C. (1996) First molecular evidence for the existence of a Tardigrada + Arthropoda clade. Molecular Biology and Evolution, 13, 76–84.

Goloboff, P.A., Farris, J.S. & Nixon, K.C. (2008) TNT, a free program for phylogenetic analysis. Cladistics, 24, 774–786.

Heraty, J., Ronquist, F., Carpenter, J.M. et al. (2011) Evolution of the hymenopteran megaradiation. Molecular Phylogenetics and Evolution, 60, 73–88.

Johnson, N.F. (1984a) Systematics of Nearctic Telenomus: classification and revisions of the podisi and phymatae species groups (Hymenoptera: Scelionidae). Bulletin of the Ohio Biological Survey, 6, 1–113.

Johnson, N.F. (1984b) Revision of the Nearctic species of the Trissolcus flavipes group (Hymenoptera: Scelionidae). Proceedings of the Entomological Society of Washington, 86, 797–807.

Johnson, N.F. (1985a) Revision of the New World species of the thytanea group of Trissolcus (Hymenoptera: Scelionidae). The Canadian Entomologist, 117, 107–112.

Johnson, N.F. (1985b) Systematics of New World Trissolcus (Hymenoptera: Scelionidae): species related to T. basalis. The Canadian Entomologist, 117, 431–445.

Johnson, N.F. (1985c) Phylogenetic relationships of the telenomine genus Niripama (Hymenoptera: Scelionidae). International Journal of Entomology, 27, 369–374.

Johnson, N.F. (1987) Systematics of New World Trissolcus, a genus of pentatomid egg-parasites (Hymenoptera: Scelionidae). Journal of Natural History, 21, 285–304.

Johnson, N.F. (1988a) Madiure, a new genus of Telenominae (Hymenoptera: Scelionidae) related to the Psix-group of genera. Colemania, 5, 25–28.

Johnson, N.F. (1988b) Species of Australian Telenominae (Hymenoptera: Scelionidae) of A.P. Dodd and A. A. Girault. Proceedings of the Entomological Society of Washington, 90, 229–243.

Johnson, N.F. (1991) Revision of Australasian Trissolcus species (Hymenoptera: Scelionidae). Invertebrate Taxonomy, 5, 211–239.

Johnson, N.F. (1996) Revision of world species of Paratelenomus (Hymenoptera: Scelionidae). The Canadian Entomologist, 128, 273–291.

Johnson, N.F. (2013) Hymenoptera Online [WWW document]. URL http://hol.osu.edu/index.html?id=592 [accessed on 17 June 2013].

Johnson, N.F. & Bin, F. (1982) Species of Telenomus (Hym., Scelionidae), parasitoids of stalked eggs of Neuroptera (Chrysopidae and Bertholdiidae). Redia, 65, 189–206.

Johnson, N.F. & Masner, L. (1985) Revision of the genus Psix Kozlov & L (Hymenoptera: Scelionidae). Systematic Entomology, 10, 33–58.

Johnson, N.F. & Musetti, L. (2003) Redefinition of the genus Phaneromyria Dodd (Hymenoptera: Scelionidae). Journal of the New York Entomological Society, 111, 138–144.

Kieffer, J.-J. (1913) Proctotrupidae, Cynipidae et Evaniidae. Voyage de Ch. Alluau et R. Jeannel en Afrique Orientale (1911-1912). Résultats scientifiques. Hyménoptères, 1, 1–35.

Kieffer, J.-J. (1926) Scelionidae. Das Tierreich, Vol. 48. Walter de Gruyter & Co., Berlin.

Kjer, K.M. (1995) Use of rRNA secondary structure in phylogenetic studies to identify homologous positions: an example of alignment and data presentation from the frogs. Molecular Phylogenetics and Evolution, 4, 314–330.

Klompen, H. (2000) A preliminary assessment of the utility of elongation factor-1α in elucidating relationships among basal Mesostigmata. Experimental and Applied Acarology, 24, 805–820.

Klompen, H., Lekveishvili, M. & Black, W.C. IV (2007) Phylogeny of parasitiform mites (Acari) based on rRNA. Molecular Phylogenetics and Evolution, 43, 936–951.

Konovalova, S.V. (2008) New species of Telenominae (Hymenoptera, Scelionidae) from the Palaeartic fauna. Zoologicheskii Zhurnal, 87, 42–48.

Kozlov, M.A. (1970) Supergeneric groupings of Proctotrupoida (Hymenoptera). Entomologicheskoye Obozreniye, 49, 203–226.

Kozlov, M.A. & Konovalova, S.V. (1983) Telenominae of the Fauna of the USSR. Nauka, Leningrad.
