Phylogenomic insight into the radiation of oak-associated gall wasps

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Background

Gall wasps (Hymenoptera: Cynipoidea: Cynipidae) constitute one of the largest radiation of gall-inducing arthropods (Ronquist et al. 2015). They are known mainly from the temperate areas of the Northern Hemisphere. Gall wasps include both true gall formers and phytophagous inquilines. Inquilines live inside the galls induced by gall wasps or sometimes other insects on related sets of host plants (Ronquist 1994). Species of Fagaceae, from genera *Lithocarpus*, *Castanea*, *Castanopsis*, *Chrysolepis*, and especially the diverse lineages of oaks (*Quercus*) are the most frequently attacked hosts (e.g. Csóka et al. 2005).

According to the recently established gall wasp classification (Hearn et al. in press), Cynipidae includes 10 distinct tribes out of which four contain inquilines: Synergini, Ceroptresini, Diastrophini and Rhoophilini (Ronquist et al. 2015, Lobato-Vila et al. 2022). The remaining tribes are gall inducers including oak gall wasps (Cynipini), the largest radiation within Cynipidae (e.g. Pénzes et al. 2018, Lobato-Vila & Pujade-Villar 2021). Oak gall wasps are associated mainly with oaks and their galls are the most frequently used hosts of inquilines from Synergini and Ceroptresini.

Even if the main lineages were long established (Ronquist et al. 2015; Lobato-Vila et al. 2022), their interrelationships were poorly understood until recently. In this way, the interpretation of their evolution, including their life history transitions were hard to understand. In the last years two phylogenomic studies (Blaimer et al. 2020, Hearn et al. in press) filled many gaps and provided sometimes surprising answers for old questions. Conclusions deduced from phylogenetic reconstructions are based on ultraconserved elements (UCEs; Blaimer et al. 2020, Hearn et al. in press) and protein-coding sequences from genome and transcriptome assemblies (Hearn et al. in press). These studies provided very similar conclusions, the main division of Cynipidae has been clarified in most cases. As suggested also by earlier studies (e.g. Ronquist et al. 1995), it is strongly supported that the inquiline tribe Ceroptresini is related to the oak gall wasps more closely than to the Synergini lineages. These results reinforced the complexity of evolution of cynipid life histories, like the shifts among inquilines and gall inducers. Necessarily, these large scale studies involved only a few species from each main lineages including Synergini.

Focusing on Synergini, earlier studies established that *Rhoophilus loewi* is the closest extant relative (van Noort et al. 2008, Rhoophilini tribe has been established recently in Lobato-Vila et al. 2022). In Hearn et al. (in press) Synergini has been emerged as the sister group of the afrotropical lineage with high support. The latter clade includes Rhoophilini and Qwaqwaiini tribes from host plants belong to the families Anacardiaceae and Salicaceae, respectively.

To date, Synergini encompasses six genera: *Agastoroxenia* Nieves-Aldrey & Medianero, 2010 (1 sp.); *Lithosaphonecrus* Tang, Melika & Bozsó, 2013 (= *Lithonecrus* Nieves-Aldrey & Butterill, 2014) (11 spp.); *Saphonecrus* Dalla-Torre & Kieffer, 1910 (about 40 spp.); *Synergus* Hartig, 1840 (about 130 spp.); *Synophrus* Hartig, 1843 (7 spp.); and *Ufo* Melika & Pujade-Villar, 2005 (5 spp.) (references are given in Lobato-Vila et al. 2022). Synergini are mainly distributed in the Nearctic and Palaearctic areas, but is known from the Neotropical and the Oriental regions, too. The single species of *Agastoroxenia* is known only from the Neotropical region (Nieves-Aldrey & Medianero 2010), *Lithosaphonecrus* and *Ufo* are distributed in the Eastern Paleartic, *Lithosaphonecrus* is also known from the Oriental and Australasia realms (e.g. Abe et al. 2014, Bozsó et al. 2015, Ide et al. 2020, Melika et al. 2005, Pujade-Villar et al. 2020, Yang et al. 2019). In contrast, *Synoprus* is known exclusively from the Western Palaearctic (Melika 2006, Pénzes et al. 2009). *Saphonecrus* is distributed throughout the entire Palaearctic, its presence in the New World was suggested but it is repeatedly questioned (Bozsó et al. 2015, Lobato-Vila et al. 2021, Pénzes et al. 2012, Schwéger et al. 2015b). *Synergus*, the most species-rich genus of Synergini has widespread distribution, it is common in the entire Paleartic and Nearctic, and it is known from the Oriental and Neotropical regions (e,g. Lobato-Vila and Pujade-Villar 2021, Pénzes et al. 2012, Schwéger et al. 2015a). At present all known Synergini species' in the Nearctic are classified as *Synergus*.

In general, our knowledge about inquilines is very limited despite their commonness (Sanver and Hawkins 2000), a few studies addressed factors that affect their diversity and evolution (Abrahamson et al. 2003). Diversification of specialist parasitic lineages depends on the evolution of their hosts (e.g. Hamerlinck et al. 2016, Stireman et al. 2005). For phytophagous insects, emergence of reproductive isolation may start with switching to a new host plant and some evidence suggests that this may cascade to the next trophic levels (e.g. Stireman et al. 2006, Ward et al. in press). Thus, we may expect that the patterns of host plant and gall trait (the extended phenotype of gall inducer) diversification may account for the inquiline variation in some degree and coincidence between phylogenies may inform as about host shifts. Frequent host plant change among inquilines has been suggested many times by mapping host plants to inquiline phylogenies (e.g. Pénzes et al. 2012), but it has been explored recently more rigorously in the Nearctic (Ward et al. 2020, Ward et al. inpress).

Oak phylogeny has been established recently, we follow Denk et al. (2021), Hipp et al. (2018) and Manos and Hipp (2021). Considering the Western Paleartic diversity of oak-associated assemblages, there are about 150 gall wasps associated with about 33 endemic oak species from both subgenera (*Quercus* subg. *Quercus* and *Quercus* subg. *Cerris*) that host to > 30 inquiline cynipid wasp species. Nearctic is far more species-rich, with more than 150 species of oak, all from *Quercus* subg. *Quercus* that are host to more than 700 oak gall wasp species (Melika et al. 2021). Study of diversity in host associations has a long history in Western Palearctic, a recent exploration for the Nearctic is presented by Ward et al. (2022b). Furthermore, using an UCE-based phylogeny, Ward et al. (2022a) concluded that oak gall wasps have shifted to new host tree species and organs numerous times and these shifts correlate with lineage divergence. Palearctic Cynipini seemed to be more conservative (Stone et al. 2009), but it is proposed being the consequence of the older divergence of oak lineages (if consider *Quercus* and *Cerris* division). Transitions between Nearctic and Paleartic regions are also demonstrated, but the origin of Cynipini remains largely unclear (discussed further below). Using the same methodology, Ward et al. (in press) revealed patterns of host shifts and diversification for oak associated inquilines from the Nearctic. Frequent shifts in Synergini are demonstrated by many earlier studies in Palearctic (e.g. Bozsó et al. 2015, Pénzes et al. 2012), but results are much clear by mapping inquiline phylogenetic tree to the stable phylogeny of their host inducers (Cynipini) and host plants (*Quercus*) as given in the Nearctic.

The phylogenomic studies of Blaimer et al. (2020) and Hearn et al. (in press) indicated the possibility of mixing of Palearctic and Nearctic lineages of inquilines. For the Nearctic, Ward et al. (in press) mentioned the monophyly of Nearctic *Synergus* noting that this may not be the case considering the Holarctic (as suggested earlier by Lobato-Vila et al 2022, Pénzes et al. 2012 and Ward et al 2020). In the UCE-based *Synergus* tree, clades and their relationships are almost always highly supported. Lower support (91%) was obtained for the first divergence, for the branch connecting *Synergus laeviventris* to the rest of the tree. But they involved only one Western Palearctic sample, one *Synophrus* from Blaimer et al. (2020), within the outgroup. Considering the divergence order on the tree of Blaimer et al. (2020), the split of *Synergus laeviventris* was the first within Synergini, all others including *Synophrus* comes later. Our aim was to attempt to clarify this divergence pattern further on the level of Holartic and add more data to the Nearctic diversity recently started to be explored.

Samples

Detailed sampling was carried out in Hungary between 2018 and 2023 with average frequency of two or three weeks. Regions were selected on the basis of our earlier experiences (e.g. Ács et al. 2010, Pénzes et al. 2009, 2012): Sopron-Kőszeg, Pécs, Bakony-Balaton felvidék, Vértes, Zemplén, Aggtelek. New highly infected regions were also found in the south part of Hungary. Further aspects for sampling site selection included the presence of forests that are highly dominated by one of the *Quercus* subgenera (*Quercus* subg. *Cerris* represented by *Q. cerris* exclusively here, and *Quercus* subg. *Quercus* represented by *Q. robur*, *Q. petraea* and *Q. pubescens*). In each year, we had also sampling periods that focused on a given species or genus, especially we searched for *Synophrus* and frequent hosts of *Saphonecrus*. Unfortunately, no *Saphonecrus* was found at all. Collected galls were put into sealed plastic bags separately in the field and taken to the laboratory. They were stored at room temperature and checked daily to capture the emerged adults. Following taxonomic identification, specimens were stored in 96% ethanol at -20 °C for lab work. Wasps were identified by George Melika. Myriads of galls were collected but parasitoids emerged most frequently from them.

Nearctic *Synergus* samples, identified to genus level, were provided by George Melika from their earlier collections. Wasps were laboratory reared from fresh galls collected in different localities of California (CA), Florida (FL), Arizona (AZ) and British Columbia (BC) between 2008 and 2009. Sampling sites cover all the three regions identified by Hipp et al. (2018) that constituted by different assemblages of North American oaks. We selected 98 *Synergus* specimens for further processing from diverse set of host plants (Supplement 1).

Finally, both dry and ethanol preserved samples were obtained from Juli Pujade-Villar, we used them for the UCE analysis.

Sanger sequencing and phylogenetics based on two locus

One advantage of using the traditional molecular phylogenetic approach is the large amount of available raw data. We can incorporate them to our analyses. There are two parallel projects for exploring and understanding Holarctic diversity (considered here as Palearctic, Nearctic and Neotropical regions, the latter two referred as New World) with our contribution. One of them has been finished with publication (Lobato-Vila et al. 2022; the DNA lab work was done in South-Korea, not by us). We refer to this paper as LV2022 for simplicity. Only the main relevant phylogenetic conclusions are discussed here. Another project is finished recently, it is under publication, so detailed below.

The main methodological difference between the two studies is that in LV2022 4 loci were used (the mitochondrial *cox1* and *cytb*, and the nuclear *28S D2* and *28S D3*, so we have probably two independent units but higher number of characters). The analysis presented here is based on one mitochondrial (*cox1*) and one nuclear (*28S D2*) loci, so studies overlap in this way. We analyzed 28S secondary structure to increase the strength the phylogenetic signal, but these results finally dropped (because we had to involve subjective decisions during the assembly, that affected the estimation of the evolutionary distance). These results are not detailed here. Further difference between the two studies is the different taxon sampling strategy. LV2022 tried to cover the full known geographic distribution of Synergini, e.g. emphasized the Eastern Palearctic species, too. The present study is much more Nearctic oriented, nevertheless sequences from LV2022 are also involved. Furthermore, LV2022 discuss taxonomic details (e.g. a new tribe, Rhoophilini has been established), while the present study focuses on the sequence-based diversity pattern without taxonomic rigor for the

Nearctic. The reason for the latter is the taxonomic uncertainties (see Ward et al. 2020, 2022b and in press).

Methods

Total genomic DNA was extracted from legs from adult specimens, either following the chelex extraction method presented in Nicholls et al. (2010) or by using Quick-DNA Miniprep Plus Kit (Zymo Research) following protocols provided by the manufacturer. A fragment of the mitochondrial cytochrome c oxidase I gene (*cox1*) and nuclear 28S ribosomal array covering part of the D2 expansion segment (*28S D2*) was amplified following the same protocol as in Bozsó et al. (2015). PCR products were cleaned using GeneJET PCR Purification Kit (Thermo Scientific) and sequenced directly by BaseClear B. V. (Leiden, The Netherlands). *28S D2* regions were sequenced in both directions, while *cox1* in forward or both directions. Chromatograms were investigated and sequences were assembled using the Staden package v.2.0 (Bonfield et al. 1995).

We downloaded overlapping Synergini sequences from different studies (Suppl. 2) and added them to our data set. New World samples from Lobato-Vila et al. (2022) and Ward et al. (2020) were included, too. *Cox1* sequences were aligned using ClustalW v.1.83 (Thompson et al. 1994) while *28S D2* were aligned using MAFFT v.7.475 with X-INS-i option (Hofacker et al. 2002; Katoh and Toh 2008; Tabei et al. 2008). Separate gene trees were inferred for each dataset in order to test congruence (results not discussed, see Supplement 3 for an example using the widely used barcoding region of 250 taxa).

Next, the alignments were combined and analyzed together. First, four data blocks of the aligned sequences were defined: Three for the separate *cox1* codon positions and one for the *28S D2* segment. To find the best-fit partitioning scheme and models of evolution, we used PartitionFinder2 v.2.1.1 with the suggested parameters for small dataset (Lanfear et al. 2016, Guindon et al. 2010). Branch lengths were set to be linked. According to the proposed models, finally we accepted four data partitions, where separate GTR+G+I models were applied for *cox1* 1st and 2nd codon positions and *28S D2* sets, and GTR+G model for the set of *cox1* 3rd codon positions. The final full alignment consisted of 1279 positions for 190 specimens.

Phylogenetic analyses were carried out in maximum likelihood (ML) and Bayesian frameworks. Maximum likelihood analysis was done using RaxML-NG v.1.0.0 (Kozlov et al. 2019) as a web service at Vital IT (https://raxml-ng.vital-it.ch/). We used scaled branch linkage model and tree searches were performed using 10 random and 10 parsimony-based starting trees. Branch support values are computed on the basis of MRE-based "bootstopping" (Pattengale et al. 2010) run up to 200 replicates with cutoff threshold 0.03. Bayesian reconstruction was carried out using MrBayes v.3.2.6 (Ronquist et al. 2012) performing two independent searches with four chains run for 15 million generations, sampling every 1000 generations and 40% considered as burn-in and dropped. Parallel runs started from random trees. We used linked branch length model. Diagnostics statistics provided by the software and plots indicated convergence. *Rhoophilus loewi*, a member of the sister group of Synergini was involved as outgroup in all analyses. Tree manipulations were performed using iTOL v.6 (Letunic et al. 2021) and FigTree v.1.4.4 (http://tree.bio.ed.ac.uk/software/figtree/).

Results – Holarctic phylogeny

The best-scoring ML tree and 50% majority rule consensus tree from the Bayesian analysis of combine data set is presented in the Supplement 4 and 5. Collapsing clades on these combined trees when branch support values < 0.6 for maximum likelihood or posteriori probabilities < 0.85 for Bayesian tree, 12 clades remained. Trees show minimal information about their interrelationships (Fig. 1).

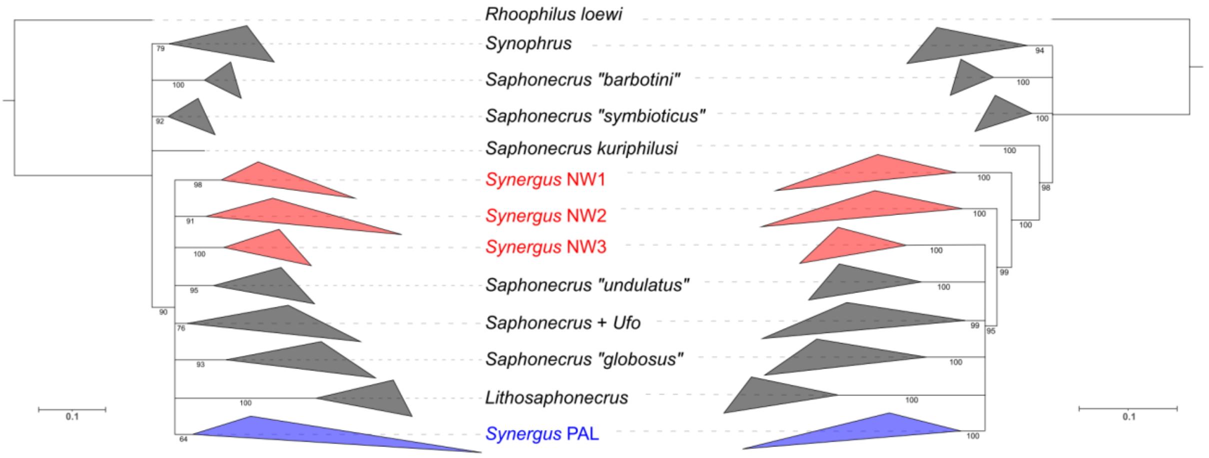

Figure 1. The collapsed ML (left) and Bayesian (right) phylogenetic tree with branch support values. Original trees are given in the in the Supplement 4 and 5. Trees are beased on the combined data set. Trees are rooted to *Rhoophilus loewi*. *Synergus* clades are colored. NW stands for New World.

Clearly, neither the New World (classified as *Synergus*) nor the Palearctic Synergini is monophyletic. *Lithosaphonecrus*, 3 groups of Palearctic *Saphonecrus* including *Ufo* and the monophyletic Palearctic *Synergus* (we refer the group of these 5 clades as PAL2) group together with 3 New World *Synergus* clades in the maximum likelihood analyses. As it was suggested by a few earlier studies (Lobato-Vila et al. 2022, Pénzes et al. 2012, Ward et al. 2020), the New World *Synergus* form separate clades, there is no overlap with the Palearctic *Synergus*. These 3 New World clades were first presented in LV2022, we follow their notation (clades NW1, NW2 and NW3). In contrast to the ML tree, the Bayesian reconstruction supports further structuring (discussed below).

Similarly to the Holarctic *Synergus*, *Saphonecrus* is not monophyletic, mentioned also in many earlier studies (e.g. Lobato-Vila et al. 2022; Pénzes et al. 2009). Some groups are closer to *Synophrus*, while others to the Paleartic *Synergus* and *Lithosaphonecrus*. Considering the Palearctic groups, some trends can be recognized (Suppl. 4 and 5; Fig. 2 in Lobato-Vila et al. 2022). The *Saphonecrus undulatus* group is associated with *Quercus* subg. *Cerris* sect. *Cerris* and known to date from the Western Palearctic exclusively. The *Saphonecrus globosus* group is associated with *Quercus* subg. *Cerris* sect. *Cyclobalanopsis* and known from the Eastern Palearctic and the Oriental regions. The large *Saphonecrus* + *Ufo* clade comprises many supported subgroups associated with *Lithocarpus* or *Cyclobalanopsis*. *Ufo* is the sister group of a *Cyclobalanopsis* associated lineage and known only from sect. *Cerris*. Members of the *Saphonecrus* + *Ufo* clade are known from the Eastern Palearctic and the Oriental regions. The ML tree of LV2022 suggests that these 3 *Saphonecrus* clades with *Ufo* together may form a monophyletic group but with very low support value (35% bootstrap support). No subg. *Quercus* association is known from this lineage. *Lithosaphonecrus* is known only from *Lithocarpus* host plant, it is distributed in the Eastern Palearctic and Oriental regions. *Lithosaphonecrus* is emerged in LV2022 as the sister group of Palearctic *Synergus* with low support (44% bootstrap and 0.86 posteriori probability). In contrast to *Saphonecrus* in PAL2 group, host plants of the Palearctic *Synergus* includes both subgenera of *Quercus* (but most frequently subg. *Quercus*) and it is known also from *Castanea* and *Castanopsis*. It is widespread on the Palearctic (e.g. Melika 2006).

The remaining 4 Palearctic clades (one of them is represented by a single sequence) form a monophyletic group in LV2022 (support values: ML 67%, Bayes: 0.99 posteriori probability). We denote this clade as PAL1. It includes the monophyletic *Synophrus* known from *Quercus* subg. *Cerris* sect. *Cerris* in the Western Palearctic. *Saphonecrus kuriphilusi* is described recently from *Castanea* host plant from Greece (Western Palearctic; Melika et al. 2018), and two other small *Saphonecrus* lineages. One of the latter includes the *Quercus* subg. *Quercus* associated *Saphonecrus* species', to date, one from the Western (*Saphonecrus connatus*) and one from the Eastern (*Saphonecrus symbioticus*) Palearctic. The other *Saphonecrus* clade (denoted as 'barbotini') includes *Quercus* subg. *Cerris* sect. Cerris and sect. *Ilex* associated species' from the Western Palearctic. The similarity of the latter group to *Synophrus* is already established (references given in Lobato-Vila et al 2022) and now recovered as monophyletic in LV2022. *Saphonecrus kuriphilusi* is the sister group of all other PAL1 lineages in LV2022 with low support values (ML: 40%; Bayes: 0.66), but Fig. 1 shows it as the sister group of PAL2+New World clades in the Bayesian analysis.

Results – Nearctic diversity

Considering the Nearctic clades, results of LW2022 suggested that NW1 can be the sister group of all other Synergini lineages, that is the latter group is monophyletic. Nevertheless, the support was very low (ML: 41%; Bayes: 0.56). Present analyses did not support this hypothesis (Fig. 1). Note that phylogenomic analyses with very limited Synergini sampling showed also a basal Nearctic lineage represented by *Synergus laeviventris* (as mentioned above, Blaimer et al. 2020), but this species belong to the NW3 clade (Suppl. 4 and 5). We return later to the basal position as this may have important consequence for the evolution of Synergini inquilines. Considering further divergence from the tree of LV2022, PAL1 is the sister of all others (support values: ML: 63; Bayes: 0.93), followed by the split of NW2 (it is the sister group of the remaining lineages, support values: ML: 40%; Bayes: 0.93). Bayesian tree in Fig. 1 suggests the NW1 then NW2 then NW3 lineages for the New World, where NW3 forms a supported (posteriori probability 0.95) but unresolved clade with the PAL2 group.

Host associations and regional distribution of New World clades are presented on a simplified (pruned and collapsed) tree schematically to increase the visibility (Fig. 2). It is derived from the combined maximum likelihood tree (Suppl. 4).

Diversity of gall associated arthropods in the Nearctic were recently explored using mass rearing (Ward et al. 2022b) as mentioned above. Considering Nearctic *Synergus*, evidences are provided for many general aspects, some of them well known from the Western Palearctic, for example: multiple species of *Synergus* can be associated with the same gall type (Askew 1961, Pénzes et al. 2012); there gall types that have probably no known *Synergus* associates; *Ceroptres* may occurs together with *Synergus*. They found that the association with gall inducer clades is unevenly distributed, there are large inducer clades (e.g. including *Neuroterus*) that associates rarely to *Synergus*. We found the same for *Neuroterus* besides the continuous sampling. They also concluded that New World *Synergus* is most frequently associated to the younger gall inducer clades suggesting a more recent radiation of inquilines but without Palearctic data (discussed below). Considering in another context, association to large oak sections was presented and lower frequency of Lobatae association was concluded. The high frequency of host switching was discussed in a recent paper (Ward et al. in press).

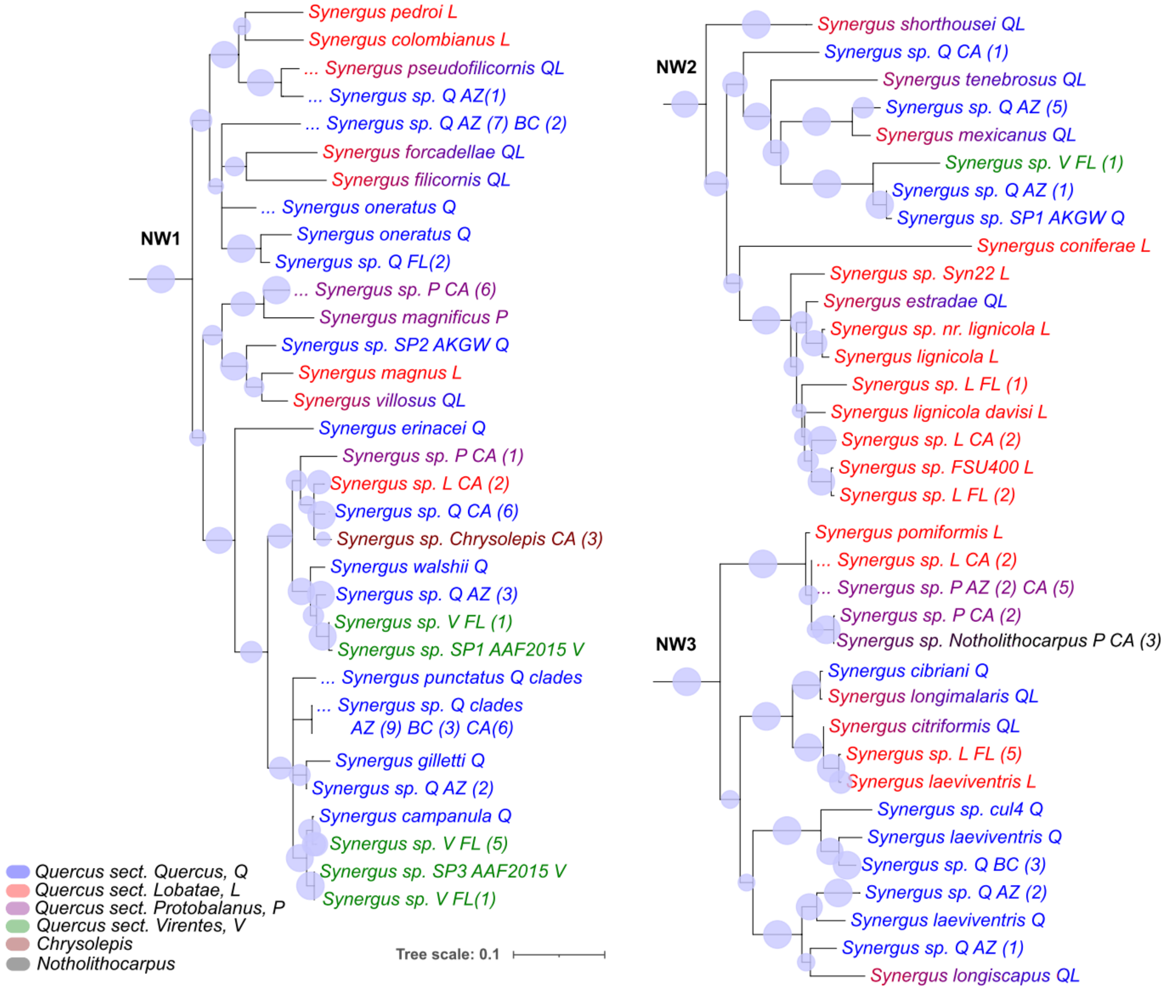

Figure 2. Schematic representation of the New World Synergus clades with host associations. It is simplified from the combined ML tree (Suppl. 4). Regions of collection of the new samples are given together with the sample size in brackets (Suppl. 1). Colors correspond to host plant lineages (color gradient represents associations with more than one clades). Bubble size is proportional to the branch support.

These results coincide with many of our findings. Fig 2 shows many suggested host switching events. For example *Lobatae* associations are present in all there New World clades, actually more than one lineage can be recognized in each group. Accepting the presence of 3 independent New World clades, for example ancestor of NW1 is probably associated with section *Quercus* rather than *Lobatae*, so host switching is proposed for Lobatae here, more than one times. Note that this could be tested with ancestral state reconstruction, but presented uncertainty in the inquiline phylogeny makes quantitative predictions to be very dubious. There are also different sect. *Protobalanus* and sect. *Virentes* clades in different NW lineages, suggesting host switching for these host plant sections, too.

Transition may occur between host plant genera. This phylogenetic pattern is well known in the Paleartic, for example between *Quercus* and *Lithocarpus* (Pénzes et al. 2012). There are 3 samples collected from *Chrysolepis*. They form one clade embedded in a Californian clade with unusually diverse associations within NW1. Furthermore this divergence seems to be derived, corresponding to a more recent transition. We have also a single sample from a gall from *Notholithocarpus*. Their sequence coincided exactly with a sample associated with sect. *Protobalanus* host plant (NW3, California).

Together with other examples from Fig. 2, our results highlight the importance of the South-West region of the Neartic for the differentiation of inquilines, even if the diversification is probably a secondary process. In this region the oak diversity is high, too (e.g. Manos and Hipp 2021). It could be a good place for further studies, to obtain deeper insight into the nature of host plant switching of inquilines that is often the first step of speciation of herbivour insects (e.g. Ward et al. in press).

UCE-based phylogenomics

Recent studies in the Nearctic demonstrated that UCEs provides enough phylogenetic resolution on species level, too. For Cynipini, study of Ward et al. (2022a) involved Paleartic samples and provided information on Holarctic level. In contrast, study on *Synergus* (Ward e al. in press) involved Nearctic sample exclusively. Unfortunately the data of Ward et al. (in press) is not available until now, so we cannot incorporate them into the analyses presented below. All other available Synergini UCE sequences (published before 2024) are involved into our study.

We planned starting genomic lab work, but it was unsuccessful. So finally we involved external service provider for this step besides the Illumina sequencing. DNA samples were sent and raw sequences were obtained. All subsequent bioinformatic and phylogenetic analyses were done by us. UCE-based results are under publication, so they are detailed here.

Samples

We selected 14 specimens for sequencing UCEs and their flanking regions (Supplement 6). The primary aim of sample selection was to maximize the coverage of our two locus tree. All main lineages (Fig. 1; Fig. 2 in LV2022) are represented, except the *Saphonecrus 'barbotini'* lineage. Second aim was to test the procedure. Besides the selection from the previous studies, we involved two dry (actually pinned) museum specimens (*Saphonecrus undulatus* and *Ufo cerroneuroteri*) that are characterized by highly degraded and low amount of extracted DNA (Supplement 7). On the other hand, we involved one fresh sample (*Synergus umbraculus*) to compare efficiency to the published genome assembly. We refer to our sample as *Synergus umbraculus2* below.

Methods

Besides the two dry materials, we used frozen samples collected between 2005 and 2021 (Suppl. 6). Total genomic DNA was extracted from legs from adult specimens or from the whole body by using Quick-DNA Miniprep Plus Kit (Zymo Research) following the protocol provided by the manufacturer. Our quality check showed considerable differences between the samples (not shown). Then dried DNA samples were sent to Daciel Arbor Biosciences (Ann Arbor, MI, USA) for enrichment and targeted sequencing. MyBaits UCE Hymenoptera 2.5Kv2P bait set was used (Branstetter et al. 2017) in order to overlap with previous studies on cynipids (Blaimer et al. 2020, Ward et al. 2022), that is we targeted the same set of loci. The summary of the sequencing is given in Supplement 7.

Then we followed the Phyluce v.1.7.1 pipeline (Faircloth 2016) to process the UCE loci. First, adapter contaminations and low quality regions were trimmed using Illumiprocessor (Faircloth 2013), based on Trimmomatic (Bolger et al. 2014). We then assembled de novo contigs using SPAdes v.3.15.4 (Prjibelski et al. 2020) with single-cell, careful and automatic coverage cutoff options. We kept contigs with unique UCE loci exclusively. Cleaning was performed with the appropriate scripts of Phyluce using the default settings.

We also downloaded available Synergini UCE and genomic data (detailed in Supplement 6). First, raw demultiplexed sequences for 5 species were downloaded from Blaimer et al. (2020), that included *Rhoophilus loewi*. Sequences were processed on the same way as our new ones and reassembled using SPAdes. Second, assembled contigs of one Nearctic *Synergus* sample (identified to genus level, referred here as *Synergus sp3*) was obtained from Ward et al. (2022). We also extracted UCE loci from four published genomes (4 genomes from Bunnefeld et al. 2018 and Gobbo et al. 2020) following the recommendations given in Phyluce manual (Faircloth 2016) using the sequences of bait set.

Next, we aligned sequence data for each locus using Mafft v.7.475 (Katoh and Standley 2013) and internally trimmed the alignment with Gblocks v.0.91b (Castresana 2000) using the relevant Phyluce scripts with their default settings. Finally we collected loci where alignment data were available for at least 18 taxa (75%) out of 24. Extracted loci statistics are given in Supplement 7. Our final data matrix consisted of 357213 aligned positions from 794 loci for 24 taxa.

In the next step, we partitioned the data matrix with the Sliding-Window Site Characteristics algorithm (SWSC-EN, Tagliacollo and Lanfear 2018) and PartitionFinder2 v.2.1.2 (Lanfear et al. 2017) using the relaxed hierarchical clustering algorithm (rclusterf option) to combine subsets and assigned GTR+G model of evolution to each subset. We then generated maximum likelihood trees with IQ-TREE2 v.2.0.7 (Chernomor et al. 2016, Nguyen et al. 2015, Minh et al. 2020), where branch support was calculated using 1000 ultrafast bootstraps (Hoang et al. 2018) with option resampling partitions and then sites within resampled partitions ('genesite', Gadagkar et al. 2005). The analysis was repeated using ModelFinder (Kalyaanamoorthy et al. 2017) with the rcluster algorithm for finding a suitable model within the IQ-TREE framework. We also used RaxML-NG v.1.2.0 (Kozlov et al. 2019) on the SWSC-EN partitions with GTR+G models, where tree searches were performed using 10 random and 10 parsimony-based starting trees. Branch support values are computed on the basis of MRE-based 'bootstopping' (Pattengale et al 2010) run up to 1000 replicates with cutoff threshold 0.01. Scaled branch linkage model were applied for all analyses. Gene trees were also generated for each locus using IQ-TREE with the models suggested by ModelFinder. We then calculated gene (gCF) and site concordance factors (sCF) from the gene trees with IQ-TREE in order to assess the branch support from gene trees (Minh et al. 2020). Finally we used ASTRAL v.5.7.8 (Zhang et al. 2018) to estimate a species tree from the gene trees under the multispecies coalescence model, where *Synergus umbraculus* individuals were forced to be monophyletic. Phylogenetic trees were rooted using *Rhoophilus loewi* as outgroup, as above.

Analyses were repeated using different settings, varying the alignment trimming methods, taxon complete sets (60, 70 and 90%) and partitioning scheme for phylogenetic reconstruction. Tree topologies were robust, except one difference highlighted below. We interpret the ML tree obtained using IQ-TREE with ModelFinder and SWSC-EN on the 75% complete data set. The latter was selected to maximize the number of taxa in the analyses even if this choice results in lower support values (especially expressed by the gCF support values, detailed below).

Results

The recovered number of loci exceeds 1000 for 70% of the samples, including the museum specimens (Supplement 7). This value is around 600 for genomes. There are two problematic samples with < 300 loci: *Synergus laeviventris* (from Blaimer el al. 2020; even if the recovered loci number is higher in our Spades assembly comparing to the original study) and *Synergus citriformis*. Nevertheless, repeating the analyses without these two taxa the topology remained the same. So we decided to keep them as both unfortunately belong the same group (NW3).

Figure 3 presents the maximum likelihood phylogenetic tree rooted to *Rhoophilus*. The analysis delineated the five groups we used before (PAL1, PAL2, NW1, NW2 and NW3) without any conflict where representatives were present. We do not have sequence for the *Synergus 'barbotini'* group (PAL1), all others includes 1-7 samples. All five groups are supported, bootstrap values are 100% and gCF is at least 39.3%. Considering their interrelationships, bootstrap values are high for most of the internal branches (in contrast to the earlier analyses) representing the common ancestor of two lineages. Nevertheless, they are very short. This is a typical pattern for fast subsequent radiations (for a given locus, there are small number of character substitutions within a short time interval). If this assumption holds, we might have issues with incomplete lineage sorting that result in frequent conflict among gene trees as demonstrated by the relatively low gCF values. This pattern suggests uncertainties in the species tree estimation from gene trees. Another issue with similar effect (low gCF values) may come from the varying quality of data (because low quality alignments are produced with many missing data), but we can largely rule out this possibility (see below).

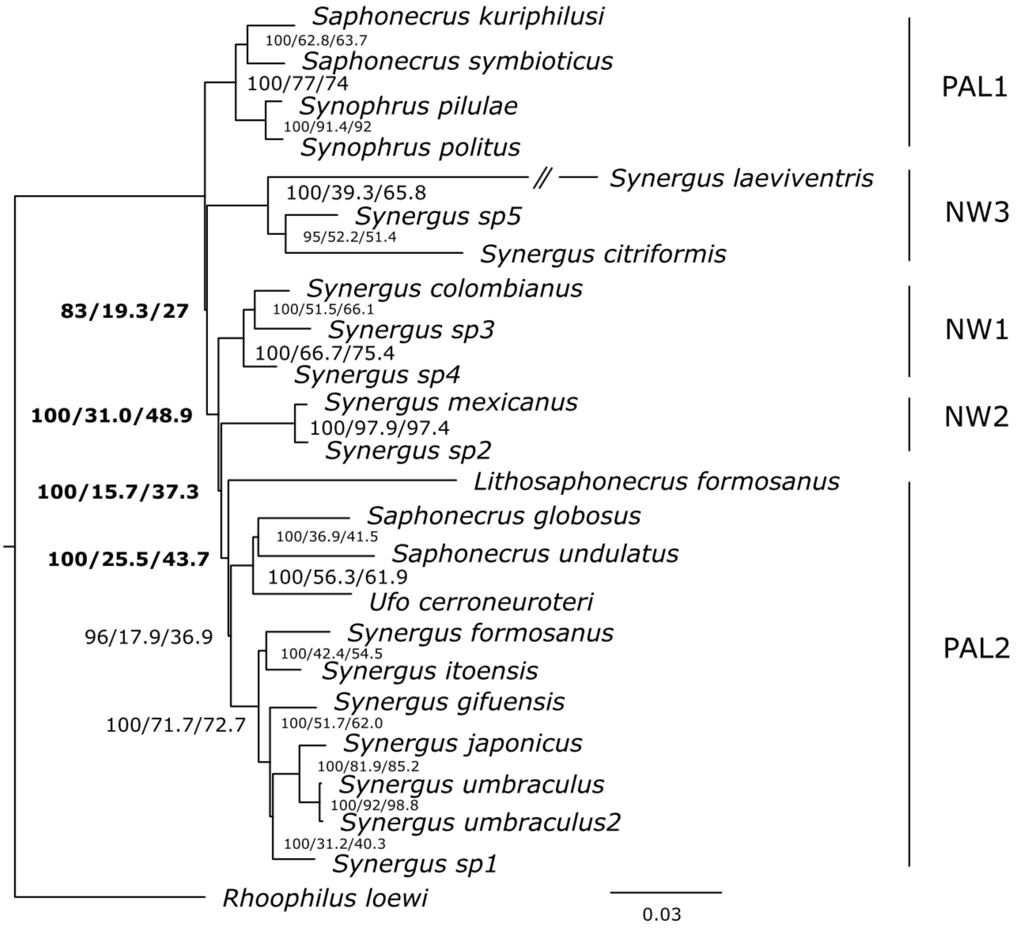

Figure 3. The UCE-based maximum likelihood phylogenetic tree. Branch support values are given as bootstrap/gCF/sCF. Clade notations correspond to Fig. 1 and support of the split of main lineages (for the internal branches) is shown in bold.

The tree represents divergence order PAL1, NW3, NW1, NW2 and PAL2. For example PAL1 emerged as the sister group of the clade of all others and NW2 is the sister group of PAL2. Neither the Palearctic nor the New Word are monophyletic. This order is supported by all analyses with nearly 100% bootstrap support with a single exception. In Fig 3, the support for NW3 as the sister group of all others except PAL1 is weaker (bootstrap 83%, gCF 19.3). Considering different model selection schemes and search methods (using e.g. RaxML, results not presented) this trend remains. The multispecies coalescent analysis even suggests sister group relationship between PAL1 and NW3 (with probability 0.5, but this analysis may be affected by the low quality data). Even we cannot exclude the possibility of NW3 as the sister group of all others (Blaimer et al. 2020 represented by *Synergus laeviventris*). *Synergus laeviventris* is the sister group of all other Nearctic *Synergus* with lower support in the UCE-based tree of Ward et al. (inpress), as mentioned above. The placement of PAL1 and NW3 must be clarified in the future.

Considering the divergence within the Palearctic clades, *Saphnecrus kuriphilusi* emerged as the sister of *Saphnocrus symbioticus* and *Synophrus* is monophyletic as before. PAL1 has *Castanea*, *Quercus* subg, *Quercus* and *Cerris* host plant associations. Within PAL2, the support values are sometimes lower, but the topology is the same in different analyses. *Lithosaphonecrus* is emerged as the sister of all other groups within PAL2. *Saphonecrus* is monophyletic within PAL2 and monophyletic together with *Ufo*. These together represents the sister group of the Palearctic *Synergus*. Palearctic *Synergus* is recovered as monophyletic with high support, but not monophyletic together with the Nearctic *Synergus* samples. So, we have two *Saphonecrus* clades, one in each Palearctic groups, so it is not monophyletic. Nevertheless, taxon sampling is very limited for general conclusions for the Holarctic pattern, but some support are given for the interrelationships of main the main lineages.

Lower gCF values are interpreted as signs of fast radiation (resulting in topology uncertainties), but we have to rule out the possibility that the conclusions are affected by the varying sequence quality. First, low quality samples groups on the same way in all analyses as in the earlier studies. They are always placed to NW3 with high support. Second, similar effect of data quality were present in reconstruction of Blaimer et al. (2020), too. Nevertheless, Hearn et al. (in press) obtained similar topology using a completely different approach. Besides that the latter is strong support for the usefulness of UCE-based phylogeny and it can be used for dry museum specimens, too.

Conclusion

Using two locus based phylogeny, large clades are delineated with high support but the support is week for their interrelationships. In this way, with a limited set of samples, these interrelationships were investigated further using an UCE-based phylogenomic approach. On the basis of the divergence pattern we obtained (Fig. 3) the following hypothesis for the biogeographic history of Synergini inquilines can be proposed. Oak subgenus *Cerris* splits from subgenus *Quercus* an estimated 50 Ma (Hipp et al. 2018). *Cerris* subgenus is entirely Paleartic while subg. *Quercus* primarily Nearctic, with one dominant clade in the Palearctic. The origin of the latter clade (roburoid) is estimated to < 15 Ma with clear origin in the Nearctic (Manos and Hipp 2021). The diversification of Cynipini is estimated at about 50 Ma, phylogenies of extant lineages suggest fast parallel radiation with oaks (Blaimer et al. 2020). The ancestral split between *Cerris* and *Quercus* associated gall wasps is suggested with the origin of Cynipini in the Palearctic (Ward et al. 2022a, it was also proposed by Stone et al. 2009), although questions still remain. That time no members of subg. *Quercus* were present in the Palearctic and most basal oak gall wasps are associated with *Cerris* and *Castanea* host plants in the Palearctic. Nevertheless, Nearctic taxa emerged as basal in other analyses (e.g. Nicholls et al. 2017; host plant section is *Protobalanus*) and this lineage is missing from the phylogenomic studies.

Considering the established framework of Fagaceae and Cynipini, we can map our inquiline phylogeny, too. The lineage of Synergini together with Rhoophilini are somewhat older than Cynipini, with an estimated age of about 89–95 Ma (Blaimer et al. 2020), while divergence of Synergini is estimated at age of 45–48 Ma and the split of *Synophrus* around 38 Ma (Fig. 2 in Blaimer et al. 2020). If we accept the basal split of PAL1, the pattern is similar to Cynipini: Palearctic origin with *Cerris* (*Synoprus*, *Saphonecrus barbotini*) or *Castanea* host plant association can be suspected. But we cannot rule out the New World origin (NW3 with *Synergus laeviventris*). Note also that the basal lineage of NW3 is associated with *Protobalanus* and *Lobatae*, with a single sample on *Notholithocarpus* (Fig 2). Under the latter hypothesis, transition to Palearctic must be happened followed by subsequent radiation on *Cerris* oak lineages. *Quercus* association in PAL1 (*Saphonecrus symbioticus*) is assumed to be secondary (host shift from *Castanea* or *Cerris*) on the basis of our data set.

In contrast to PAL1, PAL2 has clear origin in the New Word, transition to the Palearctic is assumed as sister groups are Nearctic on subg. *Quercus*. Radiation of *Saphonecrus* and *Ufo* on *Cerris* host plant is secondary in this group, similarly to *Lithosaphonecrus* on *Lithocarpus* following host shift. With weak support at present, we propose that the origin of Palearctic *Synergus* might be deduced to a further host shift to *Quercus* oaks from *Cerris*. A few *Synergus* species is associated with *Cerris* (or both *Cerris* and *Quercus*) and sister group relationship with *Saphonecrus* in *Cerris* support somewhat this hypothesis. The age of split of Nearctic and Palearctic lineages (represented by *Synergus sp1* and *sp2*) in Blaimer et al. (2020) is at about 30 Ma, older than the supposed arrival of roburoid ancestor to the Palearctic. It can be imagined that the radiation of Palearctic *Synergus* is associated with the spread of roburoids and started by an assumed *Cerris* to *Quercus* host plant shift. Clearly, more research are required to test this hypothesis.

Further results: Considering Nearctic lineages as independent units, we demonstrated some aspects of the evolution of host plant association. Results support related conclusions of Ward et al. (in press). We also highlighted the more recent radiation on the large oak diversity in California. We also provided support for the non-monophyly of Holartic *Synergus*, we propose for taxonomist that all four clades should be established as different genera. *Saphonecrus* is established as two clades with high support, one of them includes *Synophrus*, the other one includes *Ufo*.

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Theses (supervised by Zsolt Pénzes):

Nagy Gábor (2023) Társbérlő tölgy gubacsdarazsak (Hymenoptera, Cynipidae, Synergini) filogenomikája (MSc) / Phylogenomics of inquiline oak gall wasps (Hymenoptera, Cynipidae, Synergini)

Laub Hajnalka (2021) Gubacsdarazsak (Hymenoptera, Cynipidae) eredete és rendszerezése (BSc) / Origin and sytematics of gall wasps (Hymenoptera, Cynipidae)

Supplement 1. Nearctic samples with sample ID (number of sequenced individuals with the same sequence), host plant (*Q*. stands for *Quercus* subg. *Quercus*) with subsection or clade name, collection locality with rearing code and cynipid host. Data are under publication.

Sample	D2	COI	Host plant	Oak section	Oak clade	Region	Host cynipid
52	52	52	*Chrysolepis chrysophylla minor*	*Chrysolepis*		CA 1255	unknown bud gall spCAb5
53	53	53	*Chrysolepis chrysophylla*	*Chrysolepis*		CA 1211	*Dryocosmus castanopsidis*
54 (2)	106	106	*Notholithocarpus densiflorus*	*Notholithocarpus*		CA 1288	unknown leaf gall spCAl15
55	55	55	*Chrysolepis chrysophylla*	*Chrysolepis*		CA 1208	*Dryocosmus castanopsidis*
65	65	65	*Q. geminata*	*Virentes*		FL 119	*Callirhytis qbatatoides*
66	66	66	*Q. geminata*	*Virentes*		FL 180	*Disholcaspis quercusvirens*
67	67	66	*Q. geminata*	*Virentes*		FL 214	*Disholcaspis quercusvirens*
68	66	68	*Q. geminata*	*Virentes*		FL 388	*Disholcaspis quercussuccinipes*
69	66	66	*Q. geminata*	*Virentes*		FL 389	*Disholcaspis quercussuccinipes*
70	66	70	*Q. geminata*	*Virentes*		FL 585	*Belonocnema quercusvirens*
71	71	71	*Q. geminata*	*Virentes*		FL 585	*Belonocnema quercusvirens*
72	72	72	*Q. chapmani*	*Quercus*	*Stellatae*	FL 740	*Disholcaspis quercusomnivora*
73	73	73	*Q. chapmani*	*Quercus*	*Stellatae*	FL 751	*Sphaeroteras carolina*
74	74	74	*Q. palustris*	*Lobatae*	*Palustres*	FL 1022	*Callirhytis punctata*
75	75	75	*Q. virginiana*	*Virentes*		FL 1027	*Belonocnema kinseyi*
76	76	76	*Q. laurifolia*	*Lobatae*	*Phellos*	FL 1028	*Callirhytis quercusclavigera*
77	77	77	*Q. myrtifolia*	*Lobatae*	*Phellos*	FL 1029	*Amphibolips sp.*
78	77	78	*Q. laurifolia*	*Lobatae*	*Phellos*	FL 1040	*Andricus femoratus*
79	77	79	*Q. myrtifolia*	*Lobatae*	*Phellos*	FL 1059	*Dryocosmus quercusnotha*
80	80	80	*Q. myrtifolia*	*Lobatae*	*Phellos*	FL 1071	*Callirhytis quercusmedullae*
81	81	MT124886	*Q. myrtifolia*	*Lobatae*	*Phellos*	FL 1080	*Amphibolips murata*
82	82	82	*Q. garryana*	*Quercus*	*Dumosae*	BC 153	*Disholcaspis mellifica*
83	82	82	*Q. garryana*	*Quercus*	*Dumosae*	BC 185	*Disholcaspis mellifica*
84 (2)	84	84	*Q. garryana*	*Quercus*	*Dumosae*	BC 267	*Besbicus mirabilis*
85	84	84	*Q. garryana*	*Quercus*	*Dumosae*	BC 287	*Besbicus mirabilis*
86	86	86	*Q. garryana*	*Quercus*	*Dumosae*	BC 358	unknown bud gall spBCb1
87	86	86	*Q. garryana*	*Quercus*	*Dumosae*	BC 378	unknown bud gall spBCb1
88	86	88	*Q. garryana*	*Quercus*	*Dumosae*	BC 409	unknown bud gall spBCb2
91	91	91	*Q. douglasii*	*Quercus*	*Dumosae*	CA 12	*Andricus gigas*
92	92	114	*Q. lobata*	*Quercus*	*Dumosae*	CA 29	*Andricus kingi*
93 (2)	106	93	*Q. vaccinifolia*	*Protobalanus*		CA 36	*Heteroecus flavens*
94	94	94	*Q. lobata*	*Quercus*	*Dumosae*	CA 67	*Disholcaspis eldoradensis*
95	94	95	*Q. berberidifolia*	*Quercus*	*Dumosae*	CA 79	*Disholcaspis prehensa*
96	96	96	*Q. vaccinifolia*	*Protobalanus*		CA 290	*Andricus reniformis*
97	96	96	*Q. vaccinifolia*	*Protobalanus*		CA 295	*Andricus reniformis*
98	106	98	*Q. wislizenii*	*Lobatae*	*Agrifoliae*	CA 301	*Callirhytis quercuspomiformis*
99	106	99	*Q. wislizenii*	*Lobatae*	*Agrifoliae*	CA 343	*Callirhytis quercuspomiformis*
100	84	100	*Q. berberidifolia*	*Quercus*	*Dumosae*	CA 358	*Disholcaspis plumbella*
103 (2)	96	103	*Q. vaccinifolia*	*Protobalanus*		CA 442	*Disholcaspis chrysolepidis*
104	55	104	*Q. lobata*	*Quercus*	*Dumosae*	CA 500	*Antron douglasii*
105	105	105	*Q. wislizenii*	*Lobatae*	*Agrifoliae*	CA 567	*Callirhytis perdens*
106	106	106	*Q. vaccinifolia*	*Protobalanus*		CA 640	*Heteroecus pacificus*
107	71	107	*Q. vaccinifolia*	*Protobalanus*		CA 692	*Heteroecus pacificus*
108 (2)	106	108	*Q. chrysolepis*	*Protobalanus*		CA 731	*Heteroecus sanctaeclarae*
109	55	104	*Q. douglasii*	*Quercus*	*Dumosae*	CA 757	*Antron quercusechinus*
110	55	114	*Q. lobata*	*Quercus*	*Dumosae*	CA 901	*Sphaeroteras trimaculosum*
111	55	114	*Q. lobata*	*Quercus*	*Dumosae*	CA 908	*Andricus confertus*
112	112	112	*Q. wislizenii*	*Lobatae*	*Agrifoliae*	CA 931	*Dryocosmus minisculus*
113 (2)	113	113	*Q. chrysolepis*	*Protobalanus*		CA 963	*Andricus lasius*
114	55	114	*Q. lobata*	*Quercus*	*Dumosae*	CA 999	*Andricus fullawayi*
115	115	115	*Q. lobata*	*Quercus*	*Dumosae*	CA 1027	*Andricus wiltzae*
116	106	116	*Q. chrysolepis*	*Protobalanus*		CA 1100	*Heteroecus dasydactyli*
117	84	100	*Q. durata*	*Quercus*	*Dumosae*	CA 1122	*Disholcaspis plumbella*
151	151	151	*Q. arizonica*	*Quercus*	*Leucomexicana*	AZ 7	*Atrusca bella*
152	152	152	*Q. arizonica*	*Quercus*	*Leucomexicana*	AZ 13	*Atrusca aggregata*
153	71	153	*Q. turbinella*	*Quercus*	*Leucomexicana*	AZ 140	*Andricus reticulatus*
154	154	154	*Q. rugosa*	*Quercus*	*Leucomexicana*	AZ 229	*Trichoteras tubifaciens*
155	155	155	*Q. arizonica*	*Quercus*	*Leucomexicana*	AZ 242	*Dros amphora*
156	156	156	*Q. arizonica*	*Quercus*	*Leucomexicana*	AZ 262	*Xanthoteras pulchripenne*
157	71	157	*Q. arizonica*	*Quercus*	*Leucomexicana*	AZ 262	*Xanthoteras pulchripenne*
158	151	158	*Q. turbinella*	*Quercus*	*Leucomexicana*	AZ 316	*Andricus tecturnarum*
159	159	159	*Q. turbinella*	*Quercus*	*Leucomexicana*	AZ 405	*Andricus tecturnarum*
160	151	154	*Q. arizonica*	*Quercus*	*Leucomexicana*	AZ 569	*Andricus tecturnarum*
161	152	161	*Q. turbinella*	*Quercus*	*Leucomexicana*	AZ 686	*Disholcaspis sulcata*
162	162	162	*Q. oblongifolia*	*Quercus*	*Leucomexicana*	AZ 764	*Disholcaspis sulcata*
163	71	163	*Q. oblongifolia*	*Quercus*	*Leucomexicana*	AZ 782	*Disholcaspis sulcata*
164	164	164	*Q. oblongifolia*	*Quercus*	*Leucomexicana*	AZ 758	*Disholcaspis sulcata*
165 (2)	165	165	*Q. chrysolepis*	*Protobalanus*		AZ 961	*Heteroecus melanoderma*
166	166	166	*Q. rugosa*	*Quercus*	*Leucomexicana*	AZ 973	*Andricus rhizoxenus*
167	167	167	*Q. rugosa*	*Quercus*	*Leucomexicana*	AZ 990	*Andricus rhizoxenus*
169	169	169	*Q. gambelii*	*Quercus*	*Dumosae*	AZ 1167	*Callirhytis frequens*
170	170	170	*Q. oblongifolia*	*Quercus*	*Leucomexicana*	AZ 1294	*Andricus wheeleri*
171 (2)	171	171	*Q. rugosa*	*Quercus*	*Leucomexicana*	AZ 1304	*Andricus wheeleri*
172	172	172	*Q. arizonica*	*Quercus*	*Leucomexicana*	AZ 1370	*Disholcaspis spissa*
173	152	173	*Q. turbinella*	*Quercus*	*Leucomexicana*	AZ 1447	*Disholcaspis spissa*
174	174	174	*Q. arizonica*	*Quercus*	*Leucomexicana*	AZ 1564	*Disholcaspis rubens*
175	164	175	*Q. oblongifolia*	*Quercus*	*Leucomexicana*	AZ 1586	*Disholcaspis rubens*
176 (2)	176	176	*Q. turbinella*	*Quercus*	*Leucomexicana*	AZ 1602	*Disholcaspis rubens*
177	177	177	*Q. oblongifolia*	*Quercus*	*Leucomexicana*	AZ 1817	*Disholcaspis edura*
178	151	178	*Q. gambelii*	*Quercus*	*Dumosae*	AZ 1871	*Disholcaspis rubens*
180	84	100B	*Q. berberidifolia*	*Quercus*	*Dumosae*	CA 358	*Disholcaspis plumbella*
181	105	105B	*Q. wislizenii*	*Lobatae*	*Agrifoliae*	CA 567	*Callirhytis perdens*
182	106	107B	*Q. vaccinifolia*	*Protobalanus*		CA 692	*Heteroecus pacificus*
183	71	112	*Q. wislizenii*	*Lobatae*	*Agrifoliae*	CA 931	*Dryocosmus minisculus*
184	106	116B	*Q. chrysolepis*	*Protobalanus*		CA 1100	*Heteroecus dasydactyli*
185	151B	151	*Q. arizonica*	*Quercus*	*Leucomexicana*	AZ 7	*Atrusca bella*
186	164	177B	*Q. oblongifolia*	*Quercus*	*Leucomexicana*	AZ 1817	*Disholcaspis edura*
187		178B	*Q. gambelii*	*Quercus*	*Dumosae*	AZ 1871	*Disholcaspis rubens*
188	77	MT124886	*Q. myrtifolia*	*Lobatae*	*Phellos*	FL 1080	*Amphibolips murata*

Supplement 2. Samples involved from previous studies, with GenBank accession numbers, host plant (genus/subgenus, section/subsection), distribution and publication reference.

Species/Lineage	**28S**	**CoxI**	**Host plant**	**Host plant sect.**	**Locality**	**Reference**
Rhoophilus loewi	EF487123	EF486876	*Rhus sp.*		Ethiopian	Ács et al 2010
Synophrus olivieri	EF583959	EF579727	*Quercus/Cerris*	*Cerris*	Palearctic	Pénzes et al 2009
Synophrus pilulae	EF583958	EF579716	*Quercus/Cerris*	*Cerris*	Palearctic	Pénzes et al 2009
Synophrus politus	EF583954	EF579710	*Quercus/Cerris*	*Cerris*	Palearctic	Pénzes et al 2009
Ufo cerroneuroteri	JX468367	JX468358	*Quercus/Cerris*	*Cerris*	Palearctic	Melika et al 2012
Ufo nipponicus	JX468367	JX468359	*Quercus/Cerris*	*Cerris*	Palearctic	Melika et al 2012
Lithosaphonecrus dakengi	KC899801	KC899797	*Lithocarpus*		Palearctic	Bozsó et al 2015
Lithosaphonecrus formosanus	KC899802	KC899798	*Lithocarpus*		Palearctic	Bozsó et al 2015
Lithosaphonecrus huisuni	KC899799	KC899795	*Lithocarpus*		Palearctic	Bozsó et al 2015
Saphonecrus barbotini	EF487124	EF486877	*Quercus/Cerris*	*Ilex*	Palearctic	Ács et al 2010
Saphonecrus chinensis	KF532097	KF532109	*Lithocarpus*		Palearctic	Bozsó et al 2014
Saphonecrus connatus	EF487125	EF486878	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Saphonecrus emarginatus	MH178393	MG882010	*Lithocarpus*		Palearctic	Yang et al 2020
Saphonecrus gilvus "TWTI12"	JX468369	JX468364	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Melika et al 2012
Saphonecrus globosus	KF532094	KF532107	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Bozsó et al 2014
Saphonecrus haimi	EF487126	EF486879	*Quercus/Cerris*	*Cerris*	Palearctic	Ács et al 2010
Saphonecrus kuriphilusi	MF353488	MF353487	*Castanea*		Palearctic	Melika et al 2018
Saphonecrus lithocarpii	KF532098	KF532110	*Lithocarpus*		Palearctic	Bozsó et al 2014
Saphonecrus longinuxi "JP02"	JX468368	JX468362	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Bozsó et al 2014
Saphonecrus lusitanicus	EF487131	EF486881	*Quercus/Cerris*	*Ilex*	Palearctic	Ács et al 2010
Saphonecrus morii	KF532092	KF532116	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Bozsó et al 2014
Saphonecrus nantoui	KF532095	KF532108	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Bozsó et al 2014
Saphonecrus nichollsi	KF532100	KF532113	*Lithocarpus*		Palearctic	Bozsó et al 2014
Saphonecrus pachylomai	KF532102	KF532115	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Bozsó et al 2014
Saphonecrus saliciniai	KF532091	KF532121	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Bozsó et al 2014
Saphonecrus shanzhukui	KF532093	KF532106	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Bozsó et al 2014
Saphonecrus shirakashii	JX468370	JX468365	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Melika et al 2012
Saphonecrus shirokashicola	JX468371	JX468366	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Bozsó et al 2014
Saphonecrus symbioticus	KF532103	KF532117	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Bozsó et al 2014
Saphonecrus taitungi	KF532099	KF532111	*Lithocarpus*		Palearctic	Bozsó et al 2014
Saphonecrus undulatus	EF487133	EF486883	*Quercus/Cerris*	*Cerris*	Palearctic	Ács et al 2010
Synergus abei	KR270535	KR270552	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Bozsó et al 2014
Synergus acsi	EF487134	EF486884	*Quercus/Cerris*	*Cerris*	Palearctic	Ács et al 2010
Synergus belizinellus	KR270536	KR270554	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Schwéger et al 2015
Synergus campanula		MT124810	*Quercus/Quercus*	*Quercus/Prinoideae+Stellatae+Albae*	Nearctic	Ward et al 2020
Synergus castaneus	KC533839	KC533848	*Castanea*		Palearctic	Bernardo et al 2013
Synergus chinensis	EF487140	EF486890	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus cibriani	MW298735	MW274098	*Quercus/Quercus*	*Quercus*	Nearctic	Lobato-Vila et al 2022
Synergus citriformis	MW298736	MW274099	*Quercus/Quercus*	*Quercus+Lobatae*	New World	Lobato-Vila et al 2022
Synergus clandestinus	EF487141	EF486891	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus colombianus	MW298737	MW274100	*Quercus/Quercus*	*Lobatae*	Neotropical	Lobato-Vila et al 2022
Synergus coniferae		MT124905	*Quercus/Quercus*	*Lobatae/Phellos*	Nearctic	Ward et al 2020
Synergus consobrinus	EF487190	EF486955	*Quercus/Cerris*	*Cerris*	Palearctic	Ács et al 2010
Synergus crassicornis	EF487147	EF486898	*Quercus/Cerris*	*Ilex*	Palearctic	Ács et al 2010
Synergus diaphanus	EF487177	EF486939	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus erinacei		MT124785	*Quercus/Quercus*	*Quercus/Prinoideae+Albae*	Nearctic	Ward et al 2020
Synergus estradae	MW298741	MW274104	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus filicornis	MW298742	MW274105	*Quercus/Quercus*	*Quercus+Lobatae*	New World	Lobato-Vila et al 2022
Synergus forcadellae	MW298743	MW274106	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus formosanus	KR270532	KR270547	*Quercus/Cerris*	*Cerris*	Palearctic	Schwéger et al 2015
Synergus gifuensis	LC272570	LC272567	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ide et al 2018
Synergus gilletti	MW298744	MW274107	*Quercus/Quercus*	*Quercus*	Nearctic	Lobato-Vila et al 2022
Synergus incrassatus	EF487165	EF486925	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus ishikarii	KR270533	KR270549	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Schwéger et al 2015
Synergus itoensis	LC272574	LC272566	*Quercus/Cerris*	*Cyclobalanopsis*	Palearctic	Ide et al 2018
Synergus japonicus	EF487167	KR270560	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Schwéger et al 2015
Synergus kawakamii	KR270539		*Castanopsis*		Palearctic	Schwéger et al 2015
Synergus khazani	KR270537	KR270557	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Schwéger et al 2015
Synergus laeviventris		MT124886	*Quercus/Quercus*	*Lobatae/Coccineae+Palustres*	Nearctic	Ward et al 2020
Synergus laeviventris		MT124881	*Quercus/Quercus*	*Quercus/Albae*	Nearctic	Ward et al 2020
Synergus laeviventris		MT124912	*Quercus/Quercus*	*Quercus/Albae+Prinoideae*	Nearctic	Ward et al 2020
Synergus lignicola		MT124851	*Quercus/Quercus*	*Lobatae/Coccineae+Palustres*	Nearctic	Ward et al 2020
Synergus lignicola davisi	MW298740	MW274103	*Quercus/Quercus*	*Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus longimalaris	MW298748	MW274111	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus longiscapus	MW298749	MW274112	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus magnificus	MW298750	MW274113	*Quercus/Quercus*	Protobalanus	Nearctic	Lobato-Vila et al 2022
Synergus magnus		MT124918	*Quercus/Quercus*	*Lobatae/Coccineae+Phellos*	Nearctic	Ward et al 2020
Synergus mexicanus	MW298751	MW274114	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus mikoi	EF487169	EF486928	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus nr. lignicola		MT124910	*Quercus/Quercus*	*Lobatae/Coccineae*	Nearctic	Ward et al 2020
Synergus oneratus		MT124815	*Quercus/Quercus*	*Quercus/Prinoideae+Stellatae+Albae*	Nearctic	Ward et al 2020
Synergus oneratus		MT124858	*Quercus/Quercus*	*Quercus/Stellatae+Albae*	Nearctic	Ward et al 2020
Synergus oneratus		MT124846	*Quercus/Quercus*	*Quercus/Stellatae+Prinoideae*	Nearctic	Ward et al 2020
Synergus palmirae		EF486947	*Quercus/Cerris*	*Cerris*	Palearctic	Ács et al 2010
Synergus pedroi	MW298753	MW274116	*Quercus/Quercus*	*Lobatae*	Neotropical	Lobato-Vila et al 2022
Synergus physocerus	EF487184	EF486949	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus plagiotrochi	EF487188	EF486953	*Quercus/Cerris*	*Ilex*	Palearctic	Ács et al 2010
Synergus pomiformis	MW298754	MW274117	*Quercus/Quercus*	*Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus pseudofilicornis	MW298756	MW274119	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus pseudofilicornis2	MW298757	MW274120	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus punctatus		MT124849	*Quercus/Quercus*	*Quercus/Stellatae+Albae+Prinoideae*	Nearctic	Ward et al 2020
Synergus punctatus		MT124877	*Quercus/Quercus*	*Quercus/Prinoideae+Stellatae+Albae*	Nearctic	Ward et al 2020
Synergus shorthousei	MW298759	MW274122	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus sp. cul2	MW298738	MW274101	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus sp. cul4	MW298739	MW274102	*Quercus/Quercus*	*Quercus*	Nearctic	Lobato-Vila et al 2022
Synergus sp. FSU400	DQ012611	DQ012653	*Quercus/Quercus*	*Lobatae*	Nearctic	Ronquist et al 2015
Synergus sp. MOTU1	HM574143	HM574147	*Quercus/Quercus*	*Quercus*	Palearctic	Kaartinen et al 2010
Synergus sp. MOTU3	HM574146	HM574155	*Quercus/Quercus*	*Quercus*	Palearctic	Kaartinen et al 2010

Synergus sp. MOTU4	HM574133	HM574169	*Quercus/Quercus*	*Quercus*	Palearctic	Kaartinen et al 2010
Synergus sp. MOTU5	HM574137	HM574197	*Quercus/Quercus*	*Quercus*	Palearctic	Kaartinen et al 2010
Synergus sp. MOTU6	HM574140	HM574222	*Quercus and Cerris*		Palearctic	Kaartinen et al 2010
Synergus sp. SP1 "pallipes"		EF486946	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. SP1 AAF-2015 B		KR108702	*Quercus/Quercus*	*Virentes*	Nearctic	Forbes et al 2016
Synergus sp. SP1 AKGW		MT124893	*Quercus/Quercus*	*Quercus/Prinoideae*	Nearctic	Ward et al 2020
Synergus sp. SP1 TI-2017	LC272575	LC272562	*Quercus/Cerris*	*Cerris*	Palearctic	Ide et al 2018
Synergus sp. SP10 "variabilis"	EF487219	EF486967	*Quercus/Cerris*	*Cerris*	Palearctic	Ács et al 2010
Synergus sp. SP12 "apicalis/tibialis"		EF486888	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. SP13 "apicalis/tibialis"	EF487138	EF486889	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. SP14 "hayneanus/umbraculus"	EF487216	EF486965	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. sp144	KR270538	KR270559	*Quercus/Cerris*	*Cerris*	Palearctic	Schwéger et al 2015
Synergus sp. SP2 "pallicornis"	EF487171	EF486931	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. SP2 AAF-2015		KR108714	*Quercus/Quercus*	*Virentes*	Nearctic	Forbes et al 2016
Synergus sp. SP2 AAF-2015 B		KR108712	*Quercus/Quercus*	*Virentes*	Nearctic	Forbes et al 2016
Synergus sp. SP2 AAF-2015 C		KR108727	*Quercus/Quercus*	*Virentes*	Nearctic	Forbes et al 2016
Synergus sp. SP2 AKGW		MT124876	*Quercus/Quercus*	*Quercus/Prinoideae*	*Nearctic*	Ward et al 2020
Synergus sp. SP2 TI-2017	LC272571	LC272561	*Quercus/Cerris*	*Ilex*	Palearctic	Ide et al 2018
Synergus sp. SP3 "pallipes"	EF487182	EF486944	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. SP3 AAF-2015		KR108724	*Quercus/Quercus*	Virentes	Nearctic	Forbes et al 2016
Synergus sp. SP3 AKGW		MT124856	*Quercus/Quercus*	*Quercus/Stellatae+Albae+Prinoideae*	Nearctic	Ward et al 2020
Synergus sp. SP4 "pallipes"	EF487180	EF486942	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. SP4 TI-2017	LC272572	LC272563	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ide et al 2018
Synergus sp. SP4 TI-2017 B		LC272568	*Quercus/Cerris*	*Cerris*	Palearctic	Ide et al 2018
Synergus sp. SP6 "umbraculus"	EF487196	EF486961	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. SP7 "umbraculus"	EF487195	EF486960	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. SP8 "hayneanus"	EF487162	EF486916	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. SP9 "hayneanus/reinhardi"	EF487163	EF486919	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010
Synergus sp. Syn20	MW298727	MW274089	*Quercus/Quercus*	*Quercus*	Nearctic	Lobato-Vila et al 2022
Synergus sp. Syn21	MW298726	MW274091	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus sp. Syn22	MW298728	MW274090	*Quercus/Quercus*	*Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus sp. ymorph1	MW298763	MW274126	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus sp. ymorph2	MW298764	MW274127	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus symbioticus	KR270530	KR270541	*Quercus/Quercus*	*Roburoids*	Palearctic	Schwéger et al 2015
Synergus tenebrosus	MW298761	MW274124	*Quercus/Quercus*	*Quercus+Lobatae*	Nearctic	Lobato-Vila et al 2022
Synergus thaumacerus	EF487192	EF486957	*Quercus/Cerris*	*Cerris*	Palearctic	Ács et al 2010
Synergus villosus		MT124931	*Quercus/Quercus*	*Quercus+Lobatae (Prinoideae+Coccineae)*	Nearctic	Ward et al 2020
Synergus walshii		MT124801	*Quercus/Quercus*	*Quercus/Prinoideae+Stellatae+Albae*	Nearctic	Ward et al 2020
Synergus xiaolongmeni	EF487220	EF486968	*Quercus/Quercus*	*Quercus/Roburoids*	Palearctic	Ács et al 2010

Supplement 3. The *coxI* maximum likelihood gene tree (658 bp) based on 250 sequences from Synergini. The identifiers following the name of the species are the GenBank accession numbers (Suppl. 2). Number identifiers denotes the new Nearctic samples (Suppl. 1). Tree was constructed using RaxML-NG v.1.0.0 on the unpartitioned data set using GTR+FO+I+G model and default search arguments.

Supplement 4. The maximum likelihood tree of combined *cox1* and *28S D2* data. Host plant and locality are also shown. Sample data are given in Suppl. 1 and 2. Host plant notations: A: *Castanea*; C: *Quercus* subg. *Cerris*; S: *Lithocarpus*; N: *Notholithocarpus*; Y: *Chrysolepis*; For *Quercus* subg. *Quercus*, Q: sect. *Quercus*; L: *Lobatae*; V: *Virentes*; P: *Protobalanus*. For subg. *Quercus* further host plant divisions are given where it is known following Manos and Hipp (2021). Localities: PAL: Paleartic; NA: Nearctic; NT: Neotropical; AZ: Arizona; BC: British Columbia; FL: Florida; CA: California. Tree is broken at 'x'.

Supplement 5. The Bayesian majority rule tree of combined *cox1* and *28S D2* data. Host plants and localities are also shown. Sample data are given in Suppl. 1 and 2. Host plant and locality notations are given in Supplement 4. Tree is broken at ‘x’.

Supplement 6. Samples involved in the UCE-based phylogenetics (upper table) and sequence references for the downloaded data. New samples sequenced in this study are in bold. n.d. stands for no data. Genome in Method column means that UCE loci were extracted from the partly assembled genome, while UCE sequencing refers to the method we used.

Species (sample)	**Host plant clade**	**Locality**	**Reference**
Lithosaphonecrus formosanus	*Lithocarpus*	Taiwan, 2008-2012	this study
Rhoophilus loewi	known from Rhus sp.	South-Africa, 2000	Blaimer et al 2020
Saphonecrus symbioticus	*Quercus/Quercus*	South Korea, 2018	this study
Saphonecrus globosus	*Cerris/Cyclobalanopsis*	Taiwan, 2008-2009	this study
Saphonecrus kuriphilusi	*Castanea*	Greece, 2016	this study
Saphonecrus undulatus	*Cerris/Cerris*	Hungary, 2000 (dry)	this study
Synergus citriformis	*Quercus/Lobatae*	Mexico, 2012	this study
Synergus colombianus	*Quercus/Lobatae*	Colombia, 2019	this study
Synergus formosanus	*Cerris/Cerris*	Taiwan, 2008-2012	this study
Synergus gifuensis	Quercus/Quercus	Japan, 2016	Gobbo et al 2020
Synergus itoensis	Cerris/Cyclobalanopsis	Japan, 2016	Gobbo et al 2020
Synergus japonicus	known from Quercus/Quercus	known from Eastern Palearctic	Bunnefeld et al 2018
Synergus laeviventris	known from Quercus/Quercus & Lobatae	USA, 2015	Blaimer et al 2020
Synergus mexicanus	*known from Quercus/Quercus & Lobatae*	Mexico, 2019	this study
Synergus sp1	n.d.	Sweden, 2012	Blaimer et al 2020
Synergus sp2	n.d.	Arizona, USA, 2015	Blaimer et al 2020
Synergus sp3 (syn_117-1-2)	n.d.	Iowa, USA	Ward et al 2022
Synergus sp4 (spl100)	*Quercus/Quercus*	California, USA, 2008-2009	this study
Synergus sp5 (spl173)	*Quercus/Quercus*	Arizona, USA, 2008-2009	this study
Synergus umbraculus	known from Quercus/Quercus	Western Palearctic	Bunnefeld et al 2018
Synergus umbraculus (2)	*Quercus/Quercus*	Hungary, 2021	this study
Synophrus pilulae	known from Cerris	Hungary, 2007	Blaimer et al 2020
Synophrus politus	*Cerris/Cerris*	Hungary, 2005	this study
Ufo cerroneuroteri	*Cerris/Cerris*	Taiwan, 2008 (dry)	this study

Species (sample)	**Method**	**Genbank accession number**
Rhoophilus loewi	UCE sequencing	SRX8883047
Synergus gifuensis	Genome	GCA_904066015.1
Synergus itoensis	Genome	GCA_904066005.1
Synergus japonicus	Genome	GCA_900474275.1
Synergus laeviventris	UCE sequencing	SRX8883054
Synergus sp1	UCE sequencing	SRX8883055
Synergus sp2	UCE sequencing	SRX8883057
Synergus sp3 (syn_117-1-2)	UCE sequencing	Dryad, https://doi.org/10.5061/dryad.37pvmcvn6
Synergus umbraculus	Genome	GCA_900474325.1
Synophrus pilulae	UCE sequencing	SRX8883060

Supplement 7. Result of UCE sequencing, assembly and UCE extraction. Upper table: sequencing, the amount of DNA extracted in ng, number of sequence reads from one direction and sequence Q30 quality check. Lower table: number and length statistics of contigs. The latter column (# of UCE loci found) gives the number of loci finally identified.

Species (sample)	Total gDNA mass (ng)	Raw reads (R1)	% Bases >=Q30
Lithosaphonecrus formosanus	99	16 155 731	87.32
Saphonecrus symbioticus	87	11 679 910	88.74
Saphonecrus globosus	11.05	8 246 958	88.33
Saphonecrus kuriphilusi	13.65	17 442 716	89.37
Saphonecrus undulatus	12.8	12 313 330	90.77
Synergus citriformis	low. n.a.	221 740	87.46
Synergus colombianus	118.5	10 421 735	86.77
Synergus formosanus	76.5	15 864 000	86.41
Synergus mexicanus	94.5	9 995 987	89.11
Synergus sp4	152	11 283 716	86.95
Synergus sp5	47	16 237 913	87.13
Synergus umbraculus2	32.05	19 196 856	86.27
Synophrus politus	1280	14 898 193	86.71
Ufo cerroneuroteri	5.7	12 692 007	91.17

Samples	Contigs	Total bp	Mean length	95 CI length	Min. length	Max length	Median length	Contigs >1kb	# of UCE loci found
Lithosaphonecrus formosanus	136101	61131625	449.1637	1.2048	55	7937	293	12391	1879
Rhoophilus loewi	111904	31152380	278.3849	0.5224	56	7858	247	845	1581
Saphonecrus globosus	186077	65548579	352.2659	2.4123	51	148610	264	7295	1953
Saphonecrus kuriphilusi	207451	100339491	483.6780	1.0567	54	20844	333	18453	1903
Saphonecrus symbioticus	104990	44718276	425.9289	1.0634	47	9297	295	6596	1991
Saphonecrus undulatus	42910	13182187	307.2055	0.7959	56	5306	265	283	1889
Synergus citriformis	6489	2618929	403.5952	3.6516	56	3536	277	314	184
Synergus colombianus	108127	44801022	414.3370	1.2894	52	15354	275	8265	1957
Synergus formosanus	72398	35707290	493.2082	1.7194	56	14994	300	8346	1919
Synergus gifuensis	608	315600	519.0789	1.3903	348	589	520	0	608
Synergus itoensis	623	322973	518.4157	1.4163	305	589	520	0	623
Synergus japonicus	616	319810	519.1721	1.3239	322	589	520	0	616
Synergus laeviventris	17740	3799650	214.1855	0.8709	56	3243	221	23	287
Synergus mexicanus	81288	36667400	451.0801	1.3781	56	11532	296	6735	1963
Synergus sp1	184256	51936055	281.8690	0.5154	54	15146	250	2113	1767
Synergus sp2	265926	63161961	237.5171	0.4804	56	11777	216	3262	1767
Synergus sp3	1633	502860	307.9363	2.2621	78	988	285	0	888
Synergus sp4	110071	48834221	443.6611	1.3372	55	15323	281	9688	1921
Synergus sp5	80799	41062611	508.2069	1.6413	55	8903	316	9501	1936
Synergus umbraculus	619	322105	520.3635	1.3016	334	589	521	0	619
Synergus umbraculus2	164326	78475754	477.5614	1.2080	54	12888	309	16112	1862
Synophrus pilulae	93582	23773246	254.0365	0.6790	52	9905	228	877	1571
Synophrus politus	128357	58372976	454.7705	1.2411	55	9687	292	11483	1929
Ufo cerroneuroteri	41955	12633819	301.1279	0.7683	56	6883	264	196	1842