

## Digital supplementary material to

KLIMES, P., BOROVANSKA, M., PLOWMAN, N.S. & LEPONCE, M. 2017: How common is trophobiosis with hoppers (Hemiptera: Auchenorrhyncha) inside ant nests (Hymenoptera: Formicidae)? Novel interactions from New Guinea and a worldwide overview. – Myrmecological News 26: 31-45.

Tab. S1: List of all localities and plot sizes sampled for the *Pseudolasius* and their trophobionts in New Guinea (for the list of the occupied sites and trees see Tab. 1); a.s.l. = above sea level; GPS = geographic coordinates (latitude, longitude) as gathered using a global positioning system.

Elevation (m a.s.l.)	GPS	Total area sampled (ha)	Area per plot (ha)	N of plots	N of trees with <i>Pseudolasius</i> nests	N of trees with plant-hoppers	Hand collecting method	Source
10	-5.1382, 145.75367	0.28	0.28	1	0	0	Ant mosaic study in trees	M. Leponce, unpubl.
100	-5.2254, 145.0808	0.56	0.28	2	0	0	Ant mosaic study in trees	M. Leponce, unpubl.
200	-5.2317, 145.1788	0.50	0.50	1	4	2	Tree felling	KLIMES & al. (2015)
200	-5.7398, 145.3297	0.56	0.28	2	0	0	Ant mosaic study in trees	M. Leponce, unpubl.
700	-5.7319, 145.2521	0.56	0.28	2	1	1	Ant mosaic study in trees	M. Leponce, unpubl.
900	-5.72167, 145.2700	0.20	0.20	1	8	4	Tree felling	N.S. Plowman, unpubl.
1200	-5.7208, 145.2694	0.56	0.28	2	2	1	Ant mosaic study in trees	M. Leponce, unpubl.
1700	-5.7592, 145.2356	0.56	0.28	2	0	0	Ant mosaic study in trees	M. Leponce, unpubl.
2200	-5.7589, 145.1860	0.28	0.28	1	0	0	Ant mosaic study in trees	M. Leponce, unpubl.
2700	-5.8152, 145.1564	0.28	0.28	1	0	0	Ant mosaic study in trees	M. Leponce, unpubl.

Tab. S2: Results of comparisons between observed and random phylogenetic mean pairwise distance (mpd) between families containing trophobioses using the Auchenorrhyncha cladogram (Fig. 6, see methods section for details). No trophobiosis is statistically clustered across the cladogram (i.e., phylogenetically clustered  $p < 0.025$ , or overdispersed  $p > 0.975$ ); mpd.obs = observed mpd, mpd.rand.mean = mean random mpd, mpd.rand.sd = standard deviation of the random mpd, mpd.obs.rank = rank of observed vs. random mpd, mpd.obs.z = standardized effect size of observed vs. random mpd, mpd.obs.p = p-value (quantile) of observed vs. random mpd.

Dataset (presence / absence)	N of taxa (lineages)	mpd. obs	mpd. rand. mean	mpd. rand. sd	mpd. obs. rank	mpd. obs. z	mpd. obs. p	N of runs
All trophobioses (sensu lato)	13	9.56	10.21	0.65	157.00	-1.00	0.16	999
All trophobioses inside ant structures	7	10.00	10.20	1.15	401.00	-0.17	0.40	999
Obligate trophobioses inside ant nests	5	9.40	10.18	1.56	278.50	-0.50	0.28	999

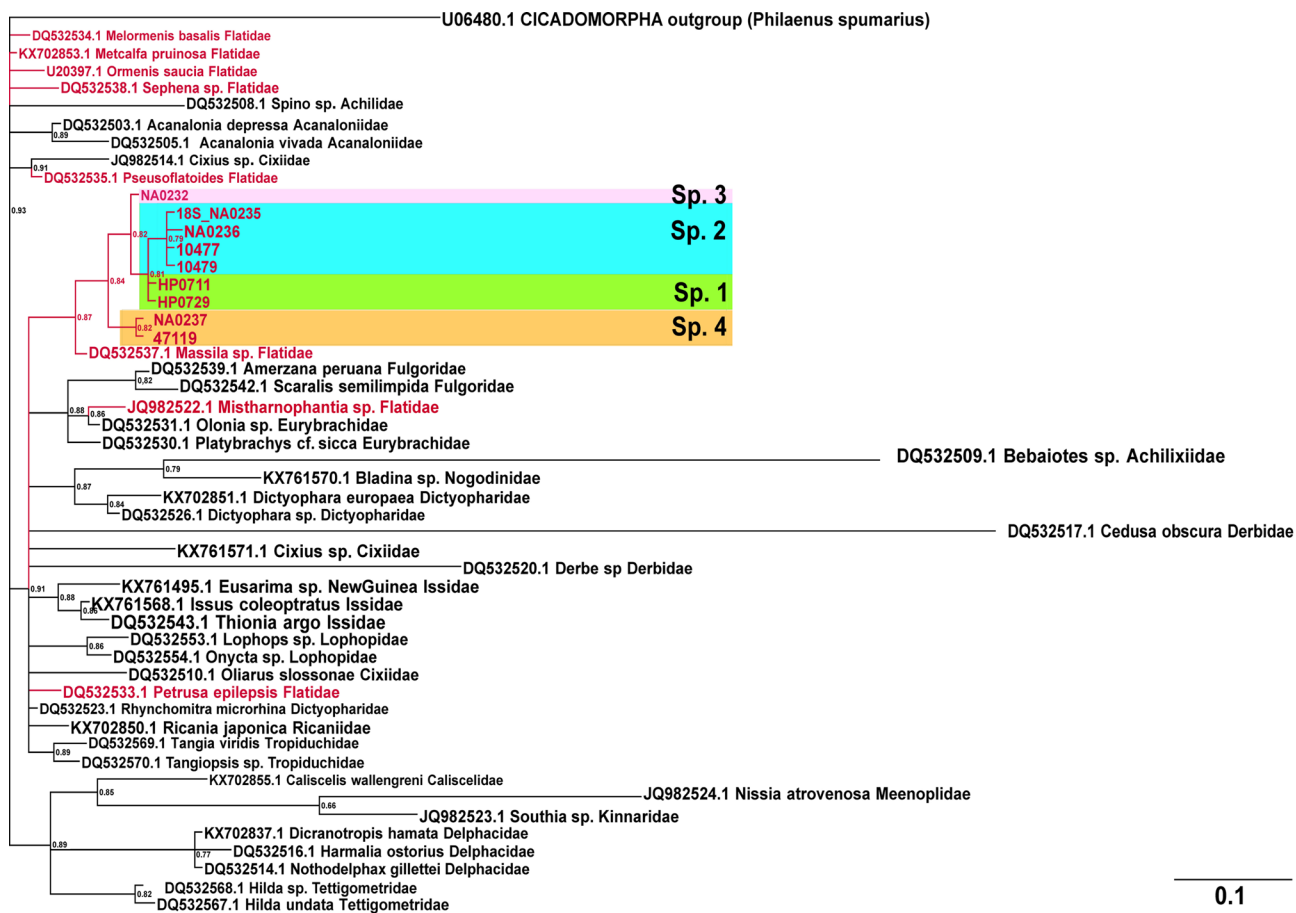


Fig. S1: The 50% majority-rule consensus tree of Fulgoromorpha estimated from the Bayesian analysis based on 18S gene (530 bp fragment). Posterior probabilities of the branches are noted at each resolved nodus ( $> 0.5$ ). Each branch represents a species (with its GenBank / sample code, species and family name). Representatives of 19 families of Fulgoroidea are included, with multiple species per family where possible. The new samples of Fulgoroidea symbionts of *Pseudolasius* are highlighted by the respective morphospecies; family Flatidae marked in red. See methods section for details.

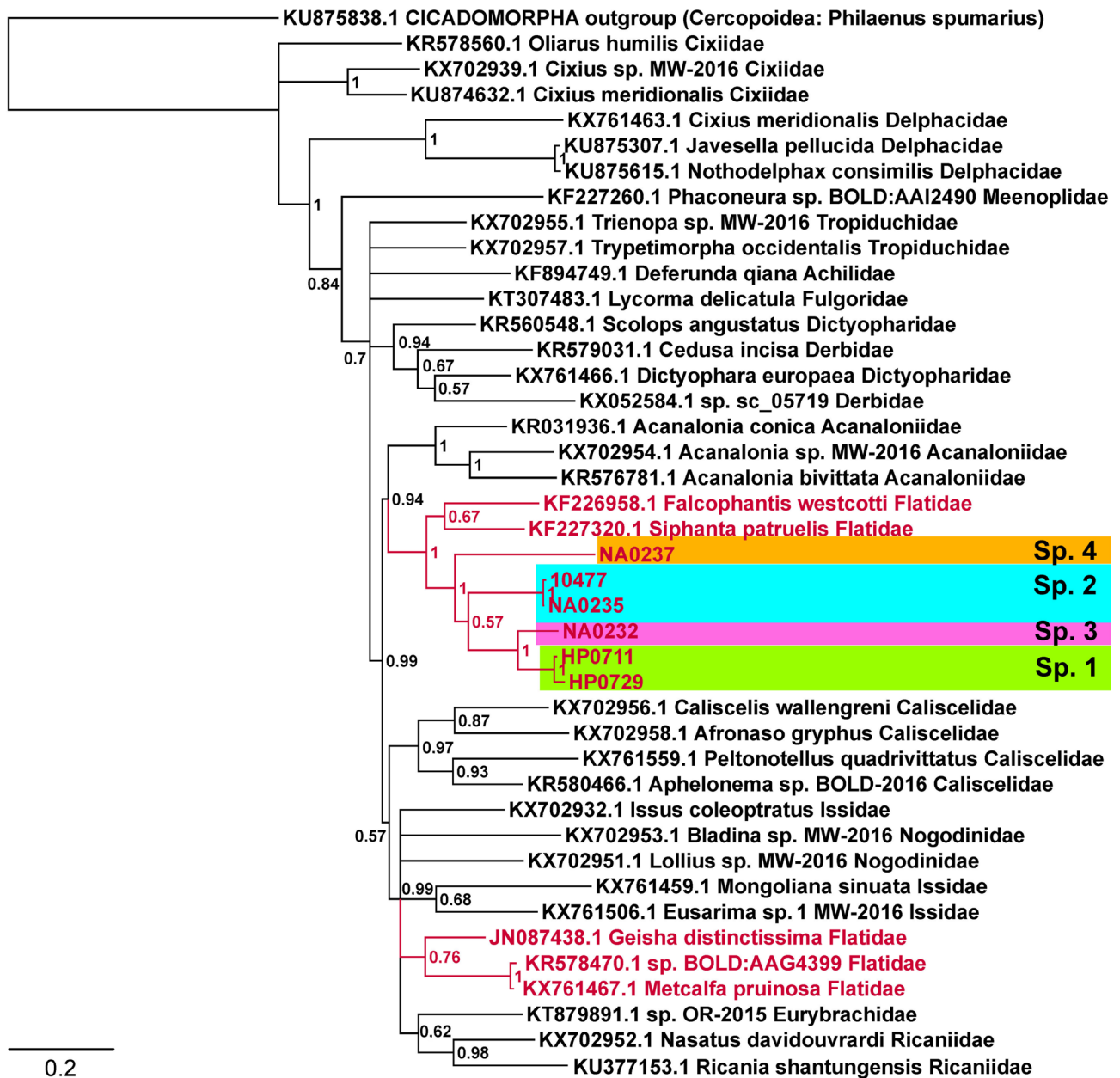


Fig. S2: The 50% majority-rule consensus tree of Fulgoromorpha estimated from the Bayesian analysis based on COI gene (615 bp fragment). Posterior probabilities of the branches are noted at each resolved nodus ( $> 0.5$ ). Each branch represents a species (with its GenBank / sample code, species and family name). Representatives of 15 families of Fulgoroidea are included, with multiple species per family where possible. The new samples of Fulgoroidea symbionts of *Pseudolasius* are highlighted by the respective morphospecies; family Flatidae marked in red. See methods section for details.

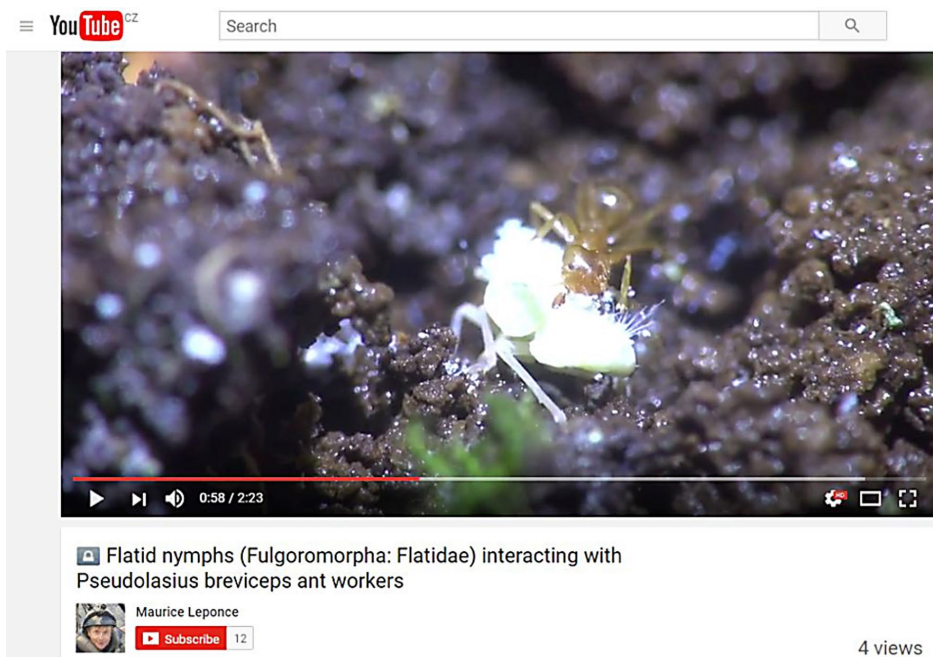


Fig. S3: Video records of interaction of flatid nymph (Sp. 4: sample ML47119) with ant worker (*Pseudolasius breviceps*) after the soil cover of the nest has been broken on tree trunk. Openly accessible video-record at: <https://www.youtube.com/watch?v=c74H-AFbG0I&feature=youtu.be>

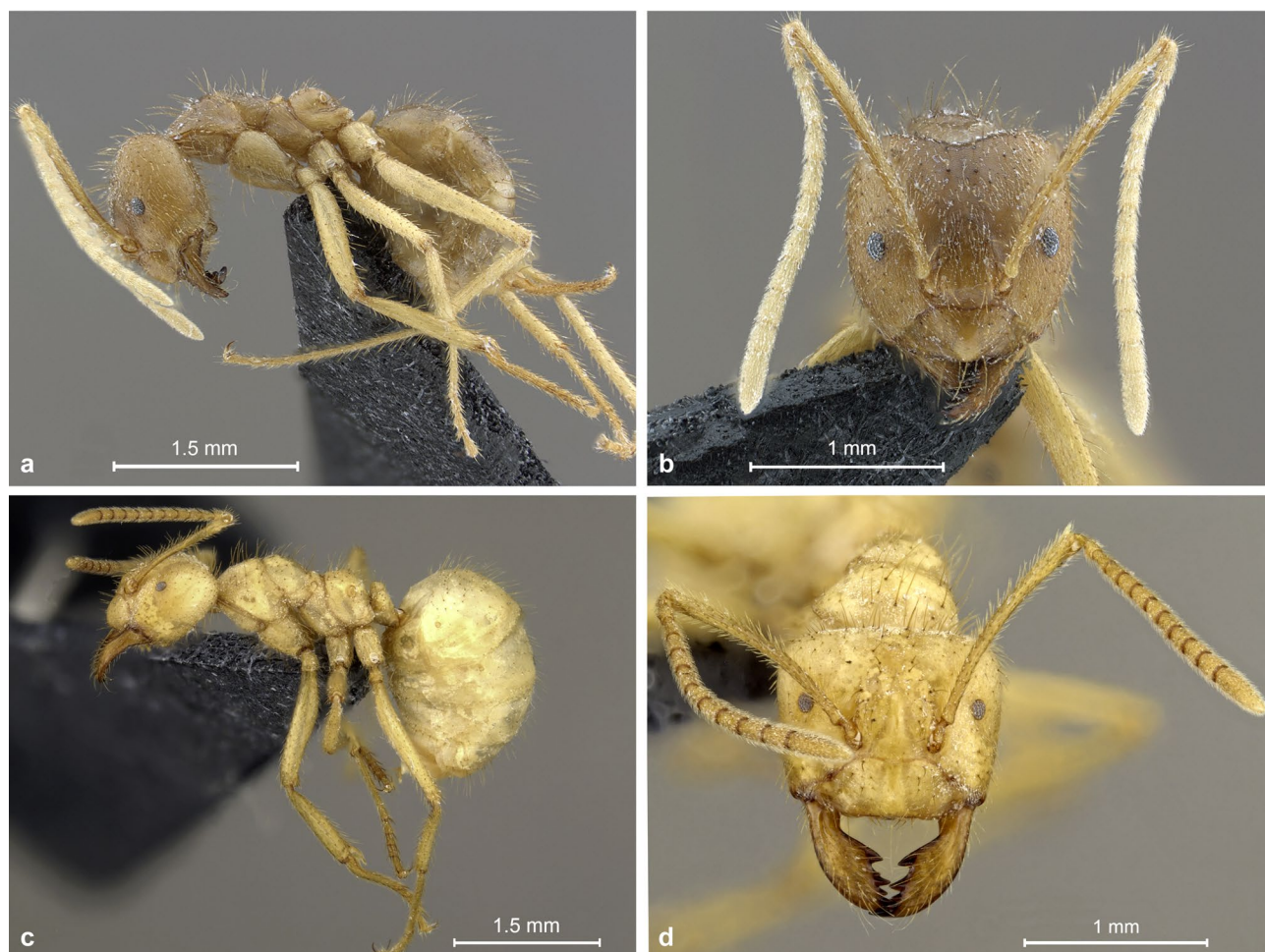


Fig. S4: *Pseudolasius* ant species that interact in their nests with planthopper nymphs (Flatidae). *Pseudolasius breviceps* sample 47119: (a) lateral, (b) frontal; *P. karawajewi* sample HP0711: (c) lateral, (d) frontal (see Tab. 1 for more information on the samples).



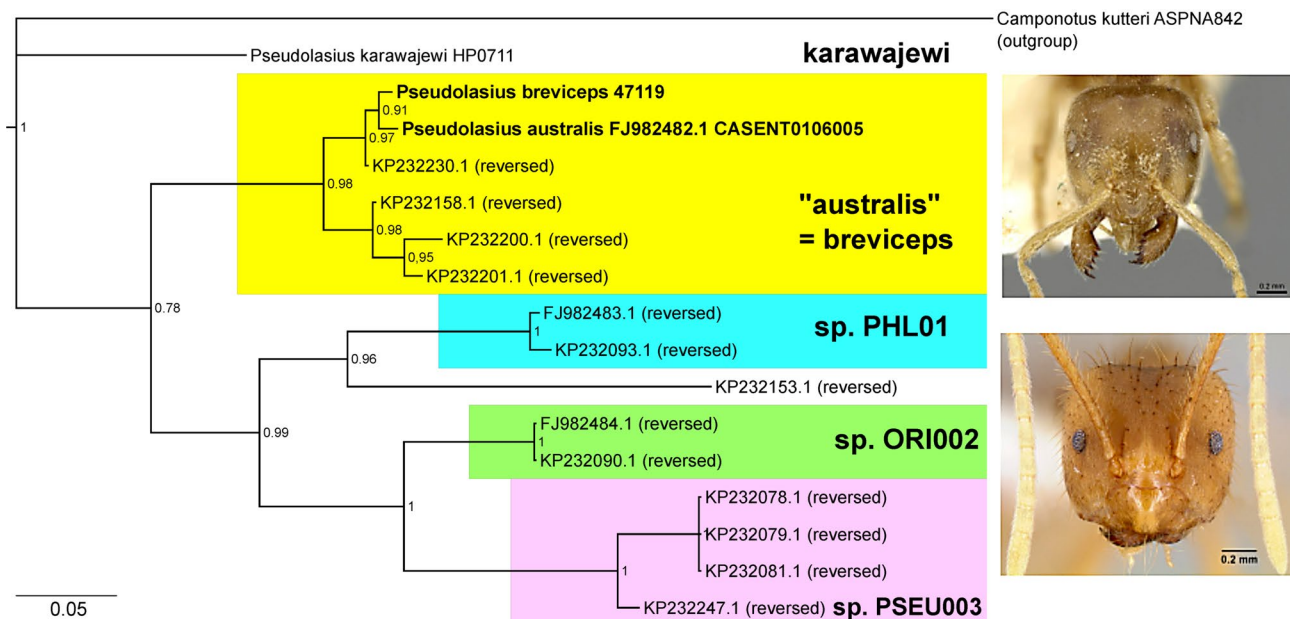


Fig. S5: The 50% majority-rule consensus tree of Australasian *Pseudolasius* estimated from the Bayesian analysis based on COI gene (658 bp fragment). The sequences were loaded from GenBank (BENSON & al. 2013) and compared with COI barcode obtained in this study (samples ML47119 and HP0711: Fig. S3). On the right are frontal head images from Antweb (ANTWEB 2017) for *P. australis* type (top) and CASENT0106005 specimen (bottom), with the latter matching morphologically *P. breviceps* from our study. The "australis" clade in GenBank database thus refers to *P. breviceps*, not *P. australis*. The latter species has not been thus likely sequenced previously, including this study. The clades are highlighted and named by species or morphospecies from GenBank.

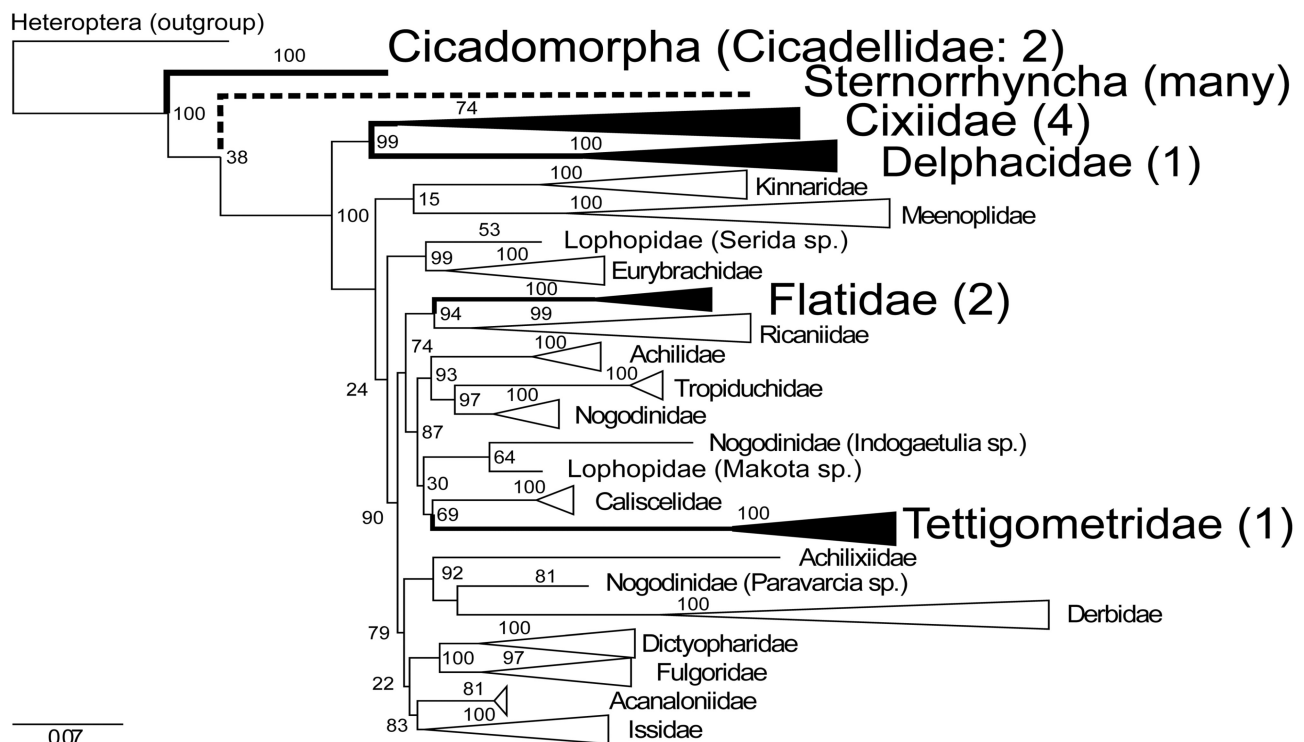


Fig. S6: ML tree of the Hemiptera main groups and 19 Fulgoromorpha families (Cixiidae to Issidae) based on several mitochondrial and nuclear genes adapted from SONG & LIANG (2013). The branches (families) based on more than one species are collated to triangles; digits present their bootstrap support. The groups with known trophobiosis with ants in their nests are marked by enlarged letters in bold with the number of the studies on obligate symbiosis in brackets (see Tab.2 for more). Sternorrhyncha are marked in dashed line due its uncertain phylogenetic position depending on the molecular data used (see Fig. 5 for alternative phylogeny based on nuclear genes only but a wider sampling across whole Auchenorrhyncha).

## References

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- BENSON, D.A., CAVANAUGH, M., CLARK, K., KARSCH-MIZRACHI, I., LIPMAN, D.J., OSTELL, J. & SAYERS, E.W. 2013: GenBank. – *Nucleic Acids Research* 41: D36-42.
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