

**Supplementary material**

**Phylogeny, systematics and rarity assessment of New Zealand endemic *Saphydrus* beetles and related enigmatic larvae (Coleoptera : Hydrophilidae : Cylominae)**

*Matthias Seidel*<sup>A,B</sup>, *Yûsuke N. Minoshima*<sup>C</sup>, *Richard A. B. Leschen*<sup>D</sup> and *Martin Fikáček*<sup>A,B,E</sup>

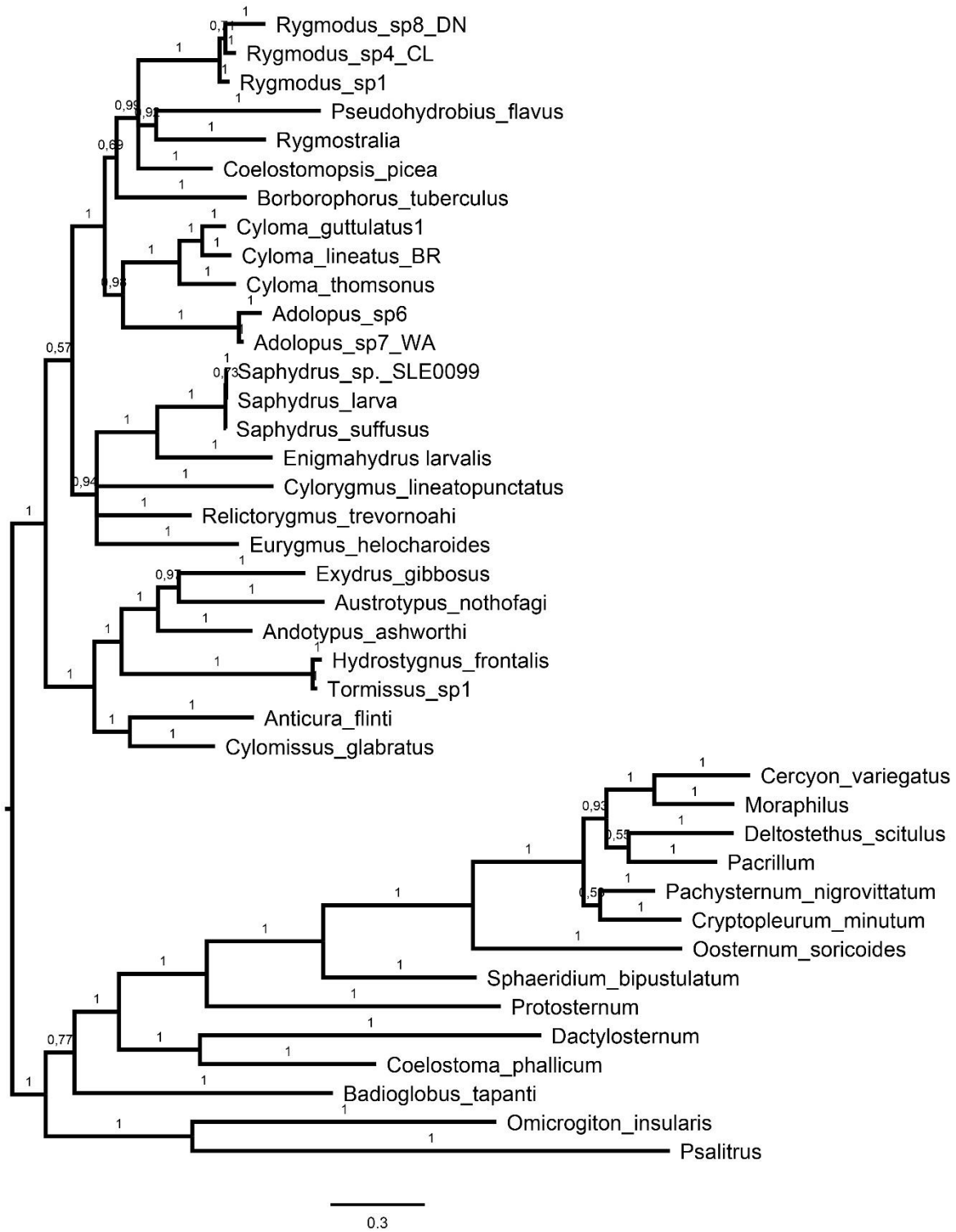
<sup>A</sup>Department of Zoology, Faculty of Science, Charles University, Viničná 7,  
CZ-12843 Praha 2, Czech Republic.

<sup>B</sup>Department of Entomology, National Museum in Prague, Cirkusová 1740,  
CZ-19300 Praha 9 – Horní Počernice, Czech Republic.

<sup>C</sup>Natural History Division, Kitakyushu Museum of Natural History and Human History,  
2-4-1 Higashida, Yahatahigashi-ku, Kitakyushu-shi, Fukuoka, 805-0071 Japan.

<sup>D</sup>Manaaki Whenua – Landcare Research, New Zealand Arthropod Collection,  
Auckland, New Zealand.

<sup>E</sup>Corresponding author. Email: mfikacek@gmail.com



**Figure S1.** The 50% majority rule tree from the Bayesian analysis.

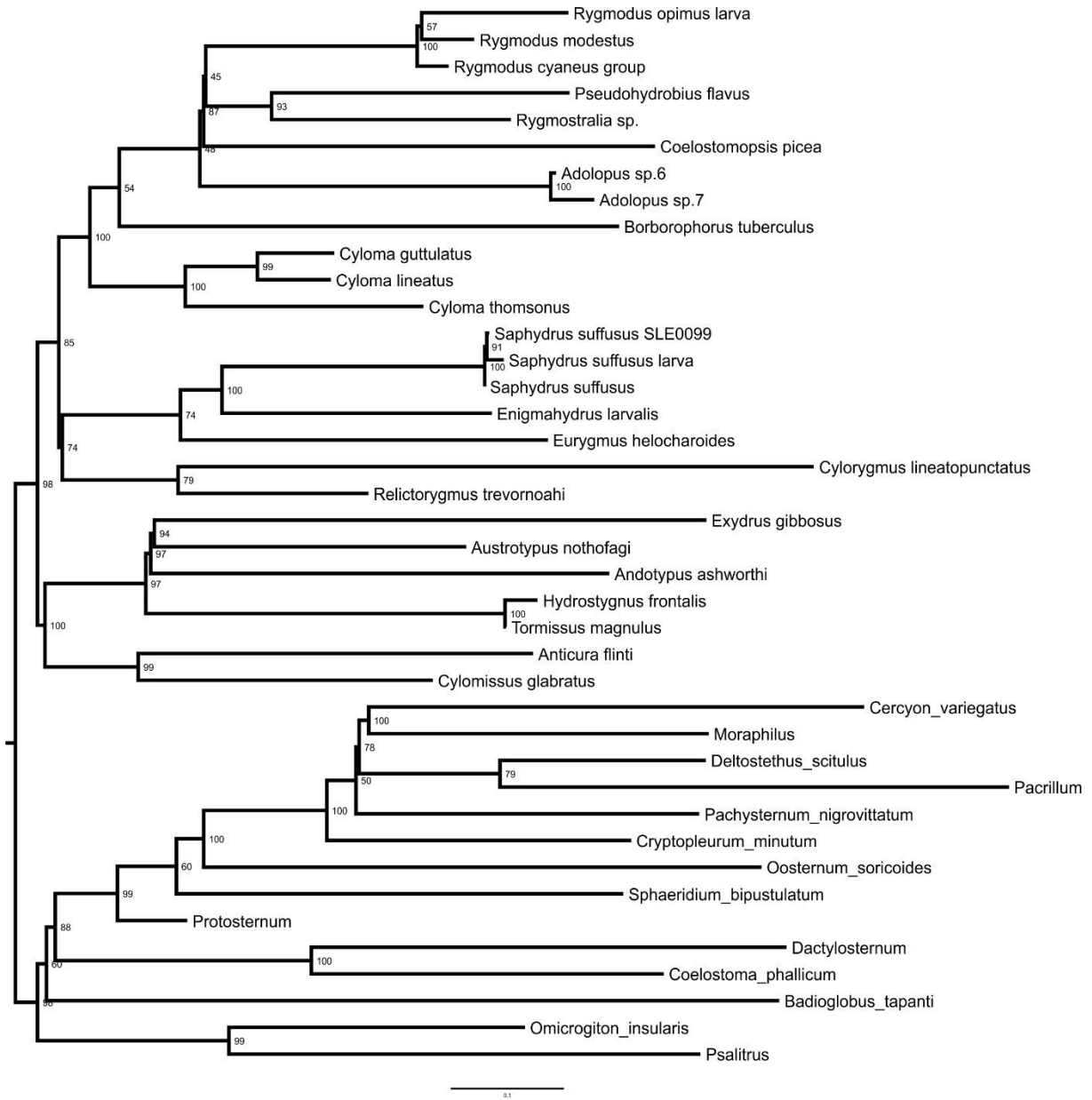


Figure S2. Maximum likelihood tree (1000 bootstrap replicates).

**Table S1. Primer sequences and programs used for PCRs for gene fragments used in our study**

cox1-3'							
SJerryF	CAACATYATATTYTGATTYTTTGG (Timmermans <i>et al.</i> 2010)						
SPatR	GCACTAWTCTGCCATATTAGA (Timmermans <i>et al.</i> 2010)						
PROGRAM							
Step	1	2	3	4	5	6	7
Temp.	95°C	95°C	50°C	72°C	repeat 2-4	72°C	12°C
Time	5:00	0:30	0:40	2:00	40×	8:00	forever
16S							
16S-1472-JJ	GGTCCTTTCGTAATAA (Astrin and Stüben 2008)						
16S-ar-JJ	CRCCTGTTTATTAATAACAT (Astrin and Stüben 2008)						
PROGRAM							
Step	1	2	3	4	5	6	7
Temp.	94°C	94°C	50°C	72°C	repeat 2-4	72°C	12°C
Time	3:00	0:30	0:45	1:00	34×	8:00	forever
18S5							
18S5'	GACAACCTGGTTGATCCTGCCAGT (Shull <i>et al.</i> 2001)						
18Sb5.0	TAACCGCAACAACCTTAAT (Shull <i>et al.</i> 2001)						
PROGRAM							
Step	1	2	3	4	5	6	7
Temp.	95°C	95°C	50°C	72°C	repeat 2-4	72°C	4°C
Time	5:00	0:30	0:40	2:00	40×	8:00	forever
18Sce							
18Sai	CTTGAGAAACGGCTACCACATC (Whiting <i>et al.</i> 1997)						
18Sb0.5	GTTTCAGCTTTGCAACCAT (Whiting <i>et al.</i> 1997)						
PROGRAM							
Step	1	2	3	4	5	6	7
Temp.	95°C	95°C	50°C	72°C	repeat 2-4	72°C	4°C
Time	5:00	0:30	0:40	2:00	40×	8:00	forever
18S3							
18Sa1.0	GGTGAAATTCTTGGACCGTC (Shull <i>et al.</i> 2001)						
18S3'I	CACCTACGGAAACCTTGTTACGAC (Shull <i>et al.</i> 2001)						
PROGRAM							
Step	1	2	3	4	5	6	7
Temp.	95°C	95°C	50°C	72°C	repeat 2-4	72°C	4°C
Time	5:00	0:30	0:40	2:00	40×	8:00	forever
28S							
28S NLF	ACCCGCTGAAYTTAAGCATAT (Van der Auwera <i>et al.</i> 1994)						
184-21							
28S LS1041R	TACGGACRTCCATCAGGGTTTCCCCTGACTTC (Maddison 2008)						
PROGRAM							
Step	1	2	3	4	5	6	7
Temp.	94°C	94°C	52°C	72°C	repeat 2-4	72°C	12°C
Time	3:00	0:30	0:45	1:00	34×	10:00	forever

**Table S2. Taxon names, codes and GenBank accession numbers of sequences [when available]**

Taxon name	Voucher ID	Cox1-3'	28S	18S	16S	classification
<i>Adolopus</i> sp6	NZ168.1	-	MT073251	-	-	Cylominae
<i>Adolopus</i> sp6	NZ565.1	MT072685		MT073224	MT073242	Cylominae
<i>Adolopus</i> sp7	NZ672	MT072682	MT073252	MT073225	MT073241	Cylominae
<i>Adolopus</i> sp7	NZ649.1	-	MT073252	-	-	
<i>Andotypus ashworthi</i>	MF792	MT072675	KP419352.1	KP419000.1	-	Cylominae
<i>Anticura flinti</i>	MF793	KM262054.1	MT073260	KT447582.1	MT073231	Cylominae
<i>Austrotypus nothofagi</i>	MF540	MT072674	MT073258	MT073219	MT073230	Cylominae
<i>Borborophorus tuberculus</i>	COL666	KC935232.1	KJ845053.1	KC935009.1	-	Cylominae
<i>Coelostomopsis picea</i>	COL119	KC935245.1	KC992551.1	KC935022.1	-	Cylominae
<i>Cyloma guttulatus</i>	NZ111.1	-	-	MT073223	-	Cylominae
<i>Cyloma guttulatus</i>	NZ243.1	MT072676	MT073249	-	MT073238	Cylominae
<i>Cyloma lineatus</i>	NZ83	MT072677	MT073250	MT073221	MT073240	Cylominae
<i>Cyloma thomsonus</i>	NZ158	MT072669	-	MT073222	MT073239	Cylominae
<i>Cyloma thomsonus</i>	NZ248.1	-	MT073248	-	-	Cylominae
<i>Cylomissus glabratus</i>	SLE0098	KC935251.1	KC992556.1	KC935028.1	KC992663.1	Cylominae
<i>Cylorygmus lineatopunctatus</i>	MF790	MT072669	KJ845012.1	MT073220	-	Cylominae
<i>Enigmahydrus larvalis</i>	MF665	MT072678	MT073262	MT073217	MT073232	Cylominae
<i>Eurygmus helocharoides</i>	COL2028	MT072679	MT073253	MT073213	MT073233	Cylominae
<i>Exydrus gibbosus</i>	NZ114.1	-	-	-	MT073228	Cylominae
<i>Exydrus gibbosus</i>	COL1791	MT072673	MT073259	MT073218	-	Cylominae
<i>Hydrostygnum frontalis</i>	SLE0097	KC935285.1	KC992591.1	KC935066.1	KC992687.1	Cylominae
<i>Pseudohydrobius flavus</i>	COL803	KF801992.1	KF802155.1	MT073215	KF801827.1	Cylominae
<i>Relictorygmus trevornoahi</i>	MF1727.2	MT072671	MT073254	MT073211	MT073235	Cylominae
<i>Rygmodes modestus</i>	COL1841	MT072681	MT073256	MT073226	MT073237	Cylominae
<i>Rygmodes opimus</i>	NZ162	MT072680	MT073255	MT073227	MT073236	Cylominae
<i>Rygmodes</i> sp1	SLE0129	KC935318.1	KC992627.1	KC935094.1	KC992705.1	Cylominae
<i>Rygmodes</i> sp1	COL800	KF801989.1	KF802152.1	MT073216	KF801824.1	Cylominae
<i>Saphydrus suffusus</i>	NZ462	MT072683	-	-	MT073234	Cylominae
<i>Saphydrus suffusus</i>	COL1786	-	MT073261	MT073214	-	
<i>Saphydrus suffusus</i>	SLE0099	KC935319.1	KC992628.1	KC935095.1	KC992706.1	Cylominae
<i>Saphydrus suffusus larva</i>	MF529	MT072684	-	-	-	Cylominae
<i>Tormissus magnulus</i>	NZ461.1	-	-	-	MT073229	Cylominae
<i>Tormissus magnulus</i>	COL1806	MT072672	MT073257	MT073212	-	Cylominae
<i>Sphaeridium bipustulatum</i>		KC935323.1	-	KC935099.1	KC992710.1	Sphaeridinae
<i>Psalitrus</i> sp.	MF249	KC935316.1	KC992625.1	KC935092.1	-	Sphaeridinae
<i>Protosternum</i> sp.	SLE0297	-	KC992624.1	KC935091.1	KC992703.1	Sphaeridinae
<i>Pacrillum</i> sp.	MF205	KC935309.1	KC992615.1	KC935082.1	-	Sphaeridinae
<i>Pachysternum nigrovittatum</i>	SLE0066	KC935308.1	KC992614.1	KC935081.1	-	Sphaeridinae
<i>Oosternum soricoides</i>	MF224	KC935306.1	KC992612.1	KC935079.1	-	Sphaeridinae
<i>Omicrogiton insularis</i>	MF284	KC935295.1	KC992602.1	KC935075.1	-	Sphaeridinae
<i>Moraphilus</i> sp.	SLE0128	KC935292.1	KC992597.1	KC935072.1	-	Sphaeridinae
<i>Deltostethus scitulus</i>	SLE0126	KC935255.1	KC992560.1	KC935032.1	KC992665.1	Sphaeridinae
<i>Dactylosternum</i> sp.	SLE0072	KC935254.1	KC992559.1	KC935031.1	KC992664.1	Sphaeridinae
<i>Cryptopleurum minutum</i>	SLE0058	KC935248.1	KC992553.1	KC935024.1	-	Sphaeridinae
<i>Coelostoma phallicum</i>	MF326	KC935244.1	KC992550.1	KC935021.1	-	Sphaeridinae
<i>Cercyon variegatus</i>	SLE0020	KC935234.1	KC992539.1	KC935011.1	KC992654.1	Sphaeridinae
<i>Badioglobus tapanti</i>	SLE0053	KC935229.1	KC992534.1	KC935005.1	-	Sphaeridinae

**Table S3. Collection data of newly sequenced Cylominae specimens included in the phylogenetic study**

Species	Code	Collecting events	Collecting information
<i>Cyloma guttulatus</i> 1	NZ111.1	New Zealand, SC, Peel Forest Reserve, Emily Falls track, 43°53.9'S 171°13.8'E, 400 m, 9-Feb-2016, J. Hájek & P. Hlaváč lgt.	
<i>Cyloma guttulatus</i> 1	NZ243.1	New Zealand, FD, Borland Walk at Borland Lodge, 45°46.35–48'S 167°32.18–25'E, 180 m, 23–28-Jan-2016, Seidel & Fikáček lgt., 2016-NZ020	baited pitfall traps (rotten squid) in mossy <i>Nothofagus</i> forest, in places with numerous ferns in understory
<i>Cyloma lineatus</i>	NZ83	New Zealand, BR, Klondyke Track, Victoria Ra, 12-Jan-2011, T.R. Buckley & R. Leschen lgt., TB452, 42°18.842'S 172°07.065'E, 699 m	sifting wet dead wood
<i>Cyloma thomsonus</i>	NZ158	New Zealand, SL, Kaka Point Scenic Reserve, 46°23.04'S 169°46.45'E, 20 m, 2–6-Feb-2016, Seidel, Sýkora & Fikáček lgt., 2016-NZ050	baited pitfall traps (rotten squid) in native coastal forest with podocarps and ferns
<i>Cyloma thomsonus</i>	NZ248.1	New Zealand, SL, Catlins, McLean Falls Track at Tautuku River, 46°34.26'S 169°20.98'E, 50 m, 3–10-Feb-2016, Seidel, Sýkora & Fikáček lgt., 2016-NZ051	baited pitfall traps (rotten squid) in wet to moderately wet area in native coastal forest with numerous tree ferns and falled/rotten trunks
<i>Enigmahydrus larvalis</i>	MF665	New Zealand, Taranaki, unnamed stream 0.2 km S of Pukeiti Garden, 9 km E of Okato, 370 m, 30-Nov-2012, 39°12.1'S 173°58.9'E, Becker, Fikáček & Hájek lgt.	in mosses on stones in/along the stream in lowland <i>Nothofagus</i> forest
<i>Saphydrus suffusus</i>	NZ462	New Zealand, NN, Nelson Lakes NP, N end of Lake Rotoroa at Braeburn Track 41.7987°S 172.58421°E, 522 m, 5–9-Dec-2016, Fikáček & Seidel lgt., MM57	lowland wet <i>Nothofagus</i> forest with huge amount of sooty moulds and with continuous layer of <i>Blechnum</i> fern understory, baited pitfall traps (rotten squid)
<i>Saphydrus suffusus</i>	COL1786	New Zealand, BR, Rough Creek, Maruia Springs, 12-Jan-2011, R. Leschen & T.R. Buckley lgt., TB455, 42°22.825'S 172°16.798'E, 550 m	
<i>Saphydrus suffusus</i> larva	MF529	New Zealand: Taranaki, Mount Egmont NP, Potaema Walk, 6.8 km W of Pembroke, 650 m, 28-Nov-2012, 39°18.9'S 174°9.1'E, Becker, Fikáček & Hájek lgt.	
<i>Cylorygmus lineatopunctatus</i>	MF790	Chile, Valparaiso, PN La Campana, Sector Ocoa, 4.75 km SE of park entrance, 'La Cascada', 870 m, 20-Nov-2013, 32°57.7'S 71°3.2'W, Fikáček, Kment & Vondráček lgt., CH03	
<i>Relictorygmus trevernoahi</i>	MF1727.2	South Africa, Western Cape, 13.3 km SEE Stanford (wetland), 100 m, 4–5-Dec-2015, 34°27.84S 19°35.75E, Arriaga, Fikáček, Seidel & Vondráček, RSA50	
<i>Rygmostralia</i> sp1	COL800	Australia, QLD, Mount Bartle Frere, NW peak, 1426 m, 19-Nov-2009, 17.385°S 145.802°E, Monteith & Turco lgt.	sieved litter
<i>Exydrus gibbosus</i>	NZ114.1	New Zealand, Auckland, Auckland - Lynfield, 31-Jan-2016, Wattle Bay reserve, 36°56.1'S 174°43.6'E, 2–30 m, J.Hájek & P. Hlaváč lgt.	sifting in secondary coastal forest

Species	Code	Collecting events	Collecting information
<i>Exydrus gibbosus</i>	COL1791	New Zealand, BP, Ohope, 8-Sep-2009, R. Leschen & E. Hilario lgt., RL1493, 37.97338°S 177.07198°E	
<i>Eurygmus helocharoides</i>	COL2028	Australia, QLD, Polly Creek, Garradunga, malaise 6, 58 m, 13-Jan-14-Feb-2010, 17.458°S 146.021°E, J. Hasenpusch lgt.	
<i>Andotypus ashworthi</i>	MF792	Chile, Los Lagos, PN Puyehue, Aguas Calientes, lower part of Sendero Pionero, 520 m, 5-9-Dec-2013, 40°44.3'S 72°18.6'W, Fikáček, Kment & Vondráček lgt., CH27	baited pitfall traps (rotten fish) in Valvidian (evergreen laurel-leaf) rainforest
<i>Anticura flinti</i>	MF793	Chile, Los Lagos, PN Puyehue, Anticura, Río Anticura between Salto Pudú and confluence with Río Golgol, 350-460 m, 6-9-Dec-2013, 40°40.0-40.5'S 72°9.9-10.6'W, Fikáček, Kment & Vondráček lgt., CH29	floating, mosses from the stone in the river (submerged + just above water) and the flood debris accumulated in logjam
<i>Austrotypus nothofagi</i>	MF540	Australia, Queensland, Lamington NP, 800 m, 13-15-Mar-2013, 28°11.9'S, 153°11.2'E, G. Monteith lgt.	
<i>Adolopus</i> sp6	NZ168.1	New Zealand, FD, Borland Walk at Borland Lodge, 45°46.35-48'S 167°32.18-25'E, 180 m, 23-28-Jan-2016, Seidel & Fikáček lgt., 2016-NZ020	on/under moist rotten logs and polypore fungi
<i>Adolopus</i> sp6	NZ565.1	New Zealand, MK, Temple Valley, 10-Jan-2006, R. Leschen, T. Buckley & R. Hoare lgt., RL1043, 44.10676°N 169.81697°E	leaf litter
<i>Adolopus</i> sp7	NZ672	New Zealand, WA Waikuku Lodge, Aorangi Range, 24-Jan-2008. K. Marske, R. Leschen & T. Buckley lgt., 41°24.616'S E175°21.880'E, 458 m	ex leaf litter/rotten wood
<i>Adolopus</i> sp7	NZ649.1	New Zealand, WA, Sutherland Vehicle Track, Aorangi Range, 24-Jan-2008, K. Marske, R. Leschen & T. Buckley lgt., 41°25.239'S E175°21.551'E, 398 m	sifted wood and leaf litter in secondary forest on former grazing land
<i>Rygmodes opimus</i>	NZ162	New Zealand, DN, Taieri Mouth, Picknick Gully, 46°3.25'S 170°11.27'E, 25 m, 12-Feb-2016, Seidel, Sýkora & Fikáček lgt., 2016-NZ073	sifting of small accumulations of leaf litter + hand collecting in small remnant of native coastal forest (very dry)
<i>Tormissus magnulus</i>	NZ461.1	New Zealand, NN, Nelson Lakes NP, N end of Lake Rotoroa at Braeburn Track 41.7987°S 172.58421°E, 522 m, 5-9-Dec-2016, Fikáček & Seidel lgt., MM57	lowland wet <i>Nothofagus</i> forest with huge amount of sooty moulds and with continuous layer of <i>Blechnum</i> fern understory, baited pitfall traps (rotten squid)
<i>Tormissus magnulus</i>	COL1806	New Zealand, WN, Kapiti Island, Wilkinson Track, 21-Jan-2008, K. Marske, R. Leschen & T. Buckley lgt., 40°51.145'S 174°55.722'E, 260 m, KM227	under/in dead wood
<i>Rygmodes modestus</i>	COL1841	New Zealand, Coromandel, Tapu, Coroglen Track, 16-Nov-2009, 36°59'S 175°35'E, D. Seldon lgt., RL1580	ex <i>Cordyline australis</i> flowers
<i>Pseudohydrobius flavus</i>	COL803	Australia, QLD, Mount Lewis Road, hut 12 km NW Julatten, 1187 m, 21-Nov-2009, 16.511°S 145.269°E, Monteith & Turco lgt.	

**Table S4. PartitionFinder2 output of MrBayes block for partition definitions (Subset 1–3 = Cox1–3', Subset 4 = 16S, Subset 5 = 18S, Subset 6 = 28S)**

---

```
begin mrbayes;

  charset Subset1 = 1-786\3;
  charset Subset2 = 2-786\3;
  charset Subset3 = 3-786\3;
  charset Subset4 = 787-1359;
  charset Subset5 = 1360-3184;
  charset Subset6 = 3185-4226;

  partition PartitionFinder = 6:Subset1, Subset2, Subset3, Subset4, Subset5, Subset6;
  set partition=PartitionFinder;

  lset applyto=(1) nst=6 rates=invgamma;
  lset applyto=(2) nst=6 rates=invgamma;
  lset applyto=(3) nst=6 rates=invgamma;
  lset applyto=(4) nst=6 rates=invgamma;
  lset applyto=(5) nst=6 rates=invgamma;
  lset applyto=(6) nst=6 rates=invgamma;

  prset applyto=(all) ratepr=variable;
  unlink statefreq=(all) revmat=(all) shape=(all) pinvar=(all) tratio=(all);
```

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**Table S5. PartitionFinder2 output of IQ-Tree block for partition definitions (Subset 1–3 = Cox1–3', Subset 4 = 16S, Subset 5 = 18S, Subset 6 = 28S)**

---

```
#nexus
begin sets;
  charset Subset1 = 1-786\3;
  charset Subset2 = 2-786\3;
  charset Subset3 = 3-786\3;
  charset Subset4 = 787-1359;
  charset Subset5 = 1360-3184;
  charset Subset6 = 3185-4226;
  charpartition PartitionFinder = GTR+I+G:Subset1, GTR+I+G:Subset2,
  GTR+I+G:Subset3, GTR+I+G:Subset4, GTR+I+G:Subset5, GTR+I+G:Subset6;
end;
```

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## References

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