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A new species of *Raphidascaris* (Nematoda: Raphidascarididae) infecting the freshwater humphead *Gymnogeophagus balzanii* (Cichlidae) from the Pantanl wetlands, Brazil and a taxonomic update of the subgenera of *Raphidascaris* based on molecular phylogeny

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Supplementary Material S1. Detailed polymerase chain reactions (PCR) and cycling conditions used in the present study, for each respective genetic region.

18S rDNA: 25µl consisted of 2.5 µl of 10X PCR buffer minus $MgCl_2$, 1.25 µl of $MgCl_2$ (50mM), 0.5 µl of dNTP's (10mM), 0.5 µl of each oligonucleotide primer (10µM), 0.2 µl of Recombinant Taq DNA polymerase (5 U/µl) (Invitrogen, Carlsbad, California), 1.25 µl of BSA (10 µg/µl), 16.3 µl of H_2O and 2.0 µl of genomic DNA (about 50ng). PCR primers were Nema18F (5'-CGC GAA TRG CTC ATT ACA ACA GC-3') and Nema18SR (5'-GGG CGG TAT CTG ATC GCC-3').
Cycling conditions: denaturation at 94°C for 5 min, followed by 36 cycles of 94°C for 30s, annealing at 52.7°C for 30s and extension at 72°C for 1min, followed by a post-amplification extension at 72°C for 5 min.

28S rDNA: 25µl consisted of 2.5 µl of 10X PCR buffer minus $MgCl_2$, 1.5 µl of $MgCl_2$ (50mM), 0.5 µl of dNTP's (10mM), 0.5 µl of each oligonucleotide primer (10µM), 0.2 µl of Recombinant Taq DNA polymerase (5 U/µl) (Invitrogen, Carlsbad, California), 1.25 µl of BSA (10 µg/µl), 16.05 µl of H_2O and 2.0 µl of genomic DNA (about 50ng). PCR primers were D2A (5'-ACA AGT ACC GTG AGG GAA AGT-3') and D3B (5'-TGC GAA GGA ACC AGC TAC TA-3').
Cycling conditions: denaturation at 94°C for 5 min, followed by 36 cycles of 94°C for 15s, annealing at 51.2°C for 30s and extension at 72°C for 30min, followed by a post-amplification extension at 72°C for 5 min.

ITS1-5.8S-ITS2: 25µl consisted of 2.5 µl of 10X PCR buffer minus $MgCl_2$, 1.0 µl of $MgCl_2$ (50mM), 0.5 µl of dNTP's (10mM), 0.5 µl of each oligonucleotide primer (10µM), 0.2 µl of Recombinant Taq DNA polymerase (5 U/µl) (Invitrogen, Carlsbad, California), 1.25 µl of BSA (10 µg/µl), 16.55 µl of H_2O and 2.0 µl of genomic DNA (about 50ng). PCR primers were A (5'-GTC GAA TTC GTA GGT GAA CCT GCG GAA GGA TCA-3') and B (5'-GCC GGA TCC GAA TCC TGG TTA GTT TCT TTT CCT-3').
Cycling conditions: denaturation at 94°C for 5 min, followed by 30 cycles of 94°C for 30s, annealing at 56.7°C for 30s and extension at 72°C for 70s, followed by a post-amplification extension at 72°C for 7 min.

***cox*1 mtDNA:** 25µl consisted of 2.5 µl of 10X PCR buffer minus $MgCl_2$, 1.25 µl of $MgCl_2$ (50mM), 0.5 µl of dNTP's (10mM), 0.5 µl of each oligonucleotide primer (10µM), 0.2 µl of Recombinant Taq DNA polymerase (5 U/µl) (Invitrogen, Carlsbad, California), 1.25 µl of BSA (10 µg/µl), 16.3 µl of H_2O and 2.0 µl of genomic DNA (about 50ng). PCR primers were CO1F (5'- TTT TTT GGT CAT CCT GAG GTT TAT-3') and CO1R (5'- ACA TAA TGA AAA TGA CTA ACA AC-3').
Cycling conditions: denaturation at 94°C for 5 min, followed by 30 cycles of 94°C for 30s, annealing at 50.9°C for 30s and extension at 72°C for 30s, followed by a post-amplification extension at 72°C for 5 min.

Table S1. Models of evolution and its fixed parameters, chosen according to the Akaike information criterion using the software jModelTest, of the phylogenetic reconstructions from the present study, associated with their respective dataset.

Dataset	Model of evolution	Gamma shape	Proportion of invariable sites	Nucleotide frequency	Substitution rates
18S + 28S rDNA	TPM2uf+I+G	0.7430	0.7060	A = 0.2062 C = 0.2208 G = 0.3070 T = 0.2661	[AC] = 1.5168 [AG] = 9.4624 [AT] = 1.5168 [CG] = 1.0000 [CT] = 9.4624 [GT] = 1.0000
ITS1-5.8S-ITS2	TIM3ef+G	0.6090	–	A = 0.2351 C = 0.2434 G = 0.2757 T = 0.2458	[AC] = 0.5165 [AG] = 2.3590 [AT] = 1.0000 [CG] = 0.5165 [CT] = 2.9112 [GT] = 1.0000
18S + ITS1-5.8S-ITS2 + 28S rDNA	TIM3+I+G	0.6150	0.4700	A = 0.2274 C = 0.2256 G = 0.2932 T = 0.2538	[AC] = 0.6939 [AG] = 3.4509 [AT] = 1.0000 [CG] = 0.6939 [CT] = 4.4533 [GT] = 1.0000
*cox*1	TPM2uf+I+G	0.1460	0.3060	A = 0.1998 C = 0.1114 G = 0.2106	[AC] = 0.0001 [AG] = 27.8037 [AT] = 0.0001

	T = 0.4782	[CG] = 1.0000 [CT] = 27.8037 [GT] = 1.0000