

# pr2-primers: an 18S rRNA primer database for protists

Daniel Vaultot <sup>1,2,\*</sup>, Stefan Geisen <sup>3,4,5</sup>, Frédéric Mahé <sup>6,7</sup>, David Bass <sup>8,9</sup>

<sup>1</sup> UMR 7144, ECOMAP, CNRS, Sorbonne Université, CNRS, Station Biologique de Roscoff, 29680 Roscoff, France

<sup>2</sup> Asian School of the Environment, Nanyang Technological University, 50 Nanyang Avenue, Singapore 639798

<sup>3</sup> Department of Terrestrial Ecology, Netherlands Institute of Ecology (NIOO-KNAW), PO Box 50, 6700 AB Wageningen, The Netherlands

<sup>4</sup> Laboratory of Nematology, Wageningen University, Wageningen, The Netherlands

<sup>5</sup> Nanjing Agricultural University, Nanjing, 210095, P.R. China

<sup>6</sup> CIRAD, UMR PHIM, F-34398 Montpellier, France

<sup>7</sup> PHIM, Univ Montpellier, CIRAD, INRAE, Institut Agro, Montpellier, France

<sup>8</sup> Centre for Environment, Fisheries and Aquaculture Science, Barrack Road, The Nothe, Weymouth, Dorset DT4 8UB, UK

<sup>9</sup> Department of Life Sciences, The Natural History Museum, Cromwell Road, London SW7 5BD, UK

## ORCID

- Daniel Vaultot: 0000-0002-0717-5685
- Stefan Geisen: 0000-0003-0734-727X
- Frédéric Mahé: 0000-0002-2808-0984
- David Bass: 0000-0002-9883-7823

\* Corresponding author: [vaultot@gmail.com](mailto:vaultot@gmail.com)

## Supplementary Material

**Table S1:** List of 18S rRNA primers in the pr2-primers database ordered by start position relative to the sequence of the yeast *Saccharomyces cerevisiae* (FU970071). Some specific primers do not match the yeast sequence and are found at the bottom of the table. DOI for reference can be found in the on-line web application.

id	Name	Sequence	Direction	Start	Specificity	Reference
71	PF1	TGCGTACCTGGTTGATCCTGCC	fwd	-5		Keeling, (2002)
78	EukA	AACCTGGTTGATCCTGCCAGT	fwd	0		Medlin et al. (1988)
81	Euk328F	ACCTGGTTGATCCTGCCAG	fwd	1		Moon et al. (2001)
138	18SV1V2F	ACCTGGTTGATCCTGCCA	fwd	1	non-Metazoa	Clerissi et al. (2018)
331	Heterokonta For	ACCTGGTTGATCCTGCCAGTAGTCATAC	fwd	1	Heterokonta	Scheckenbach et al. (2010)
220	NSF4/18	CTGGTTGATYCTGCCAGT	fwd	3		Hendriks et al. (1989)
333	18SForBodo	CTGGTTGATTCTGCCAGTAGT	fwd	3	Kinetoplastea	Scheckenbach et al. (2010)
168	Pbr1	GGTTGATCCTGCCAGTAGTC	fwd	5	Plasmodiophora	Niwa et al. (2011)
169	Pbr1r	GACTACTGGCAGGATCAACC	rev	5	Plasmodiophora	Niwa et al. (2011)
109	SF2Dark	GTTGATCCTGCCAGTAGTGT	fwd	6	Myxomycetes	Fiore-Donno (2016)
334	kineto14F	CTGCCAGTAGTCATATGCTTGTTC AAGGA	fwd	13	Kinetoplastea	von der Heyden and Cavalier-Smith (2005)
155	NS1	GTAGTCATATGCTTGTCTC	fwd	19		White et al. (1990)
142	25F	CATATGCTTGTCTCAAAGATTAAGCCA	fwd	24		Cavalier-Smith et al. (2009)
346	Thx25F	CATATGCTTGTCTCAAAGATTAAGCCA	fwd	24		Cavalier-Smith and von der Heyden (2007)
79	63f	ACGCTTGTCTCAAAGATTA	fwd	27		Lepere et al. (2011)
136	F04	GCTTGTCTCAAAGATTAAGCC	fwd	29		Blaxter et al. (1998)
255	HapGenFor33	TTGYCTYAAAGATTAAGCCATGCA	fwd	31	Haplosporidia	Ward et al. (2019)
234	18S-42F	CTCAARGAYTAAGCCATGCA	fwd	35		López-García et al. (2003)
294	73F	GCCATGCATGTCTAAGTATAAACWTC	fwd	47	Glissomonads	Howe et al. (2009)
329	Tel103-126F	TACACGGTGAGACTGCGAAT	fwd	74	Telonemia	Bréte et al. (2010)
246	Plas1f	TCAGTGAATCTGCGGATGGC	fwd	77	Plasmodiophorids	Neuhauser et al. (2014)
107	SFAca22	CGGYGAGACTGCGGATGG	fwd	78	Acanthamoeba	Fiore-Donno (2016)
256	HapGenFor84	CTGTGAAACTGCAKATGGCTC	fwd	78	Haplosporidia	Ward et al. (2019)
300	sA4-gra	CNGTGAACWGCAGATGG	fwd	78	Granofilosea incl. Reticulamoeba spp.	Bass et al. (2012)
231	18S-82F	GAAACTGCGAATGGCTC	fwd	82		López-García et al. (2003)
313	NSF83	GAAACTGCGAATGGCTCATT	fwd	82		Hendriks et al. (1989)
366	18S V2i F	CTGTGAATGGCTCCTTACATCAG	fwd	86	Euglenids	Guminska et al. (2021)
104	Kineto 80	CATCAGACGYAATCTGCCGC	fwd	103	Kinetoplastea	Lentendu, G. et al. (2014)
82	152+	TTACATGGATAACCGTGGTAATTC	fwd	135	oligotrich, choreotrich	Tamura et al. (2011)
280	LGD-135	TAAGACGACGATTGCTGATTT	rev	135	Cyclotrichiid Ciliates	Bass et al. (2009)
301	V2f-d5	GGATAGCCGTAATAATTGTGG	fwd	142	Reticulamoeba spp.	Bass et al. (2012)
368	150+	AHTTACATGGATAACCGTGG	fwd	150	ciliates	Doherty et al. (2007)
98	Chryso 240	GGAAACCAATGCGGGGCAAC	fwd	216	Chrysophyceae	Lentendu, G. et al. (2014)
289	243F	CCAATGCACCCTCTGGGTGGTT	fwd	243	Cercomonas-clade A	Karpov et al. (2006)
310	Diphy257F	AAGWGGARTCATAATAACTTTTGCG	fwd	257	Diphyllatea	Orr et al. (2018)
88	Cer2F	ATTTCTGCCCTATCAGCT	fwd	300	Cercozoa	Lentendu, G. et al. (2014)
315	Oom278F	CTATCAGCTTTGGATGGTAGGA	fwd	309	Oomycetes	Holt et al. (2018)
252	Hap-E312f	CATAGCAGATGGAAGTTTGAGG	fwd	312	Haplosporidium sp.	Ward et al. (2019)
308	AU2	TTTCGATGGTAGGATAGDGG	fwd	317	Fungi	Vandenkoornhuyse et al. (2002)
139	18SV1V2R	GTARKCCWMTAYMYTACC	rev	324	non-Metazoa	Clerissi et al. (2018)
302	C3f-d5	GACATCTGAGGTGATAACGAA	fwd	344	Reticulamoeba spp.	Bass et al. (2012)
292	369R	TCGCATTACGTATCGCATTTCGCTG	rev	369		Karpov et al. (2006)
367	18S V2i R	GCTSCCTCTCCGGAATCRAACC	rev	371	Euglenids	Guminska et al. (2021)
137	R22	GCCTGCTGCCCTTCTTGA	rev	412		Blaxter et al. (1998)
251	Hap-M412r	CGAGGTTGCCAAGTCTTTTCG	rev	412	Minchinia mytili	Ward et al. (2019)
195	Par 18S-F	GCAGCAGCGGYGAAAC	fwd	422	Parabasalids	Michaud et al. (2020)
227	545F	AGGCGCGTAAATTACCCAATC	fwd	427		Kawachi et al. (2016)
311	Diphy453F	CGCAAATTACCCAATCCTG	fwd	432	Diphyllatea	Orr et al. (2018)
254	Hap-E449r	TTGGATGCACTTTCAAGATTACC	rev	449	Haplosporidium sp.	Ward et al. (2019)
151	SAR V3 F	AYTCAGGGAGGTAGTGACAAG	fwd	451	SAR	Sisson et al. (2018)
242	mik451F	GCCGAGAYGGTTAAWAGGCCTCCT	fwd	451	Mikrocytid	Hartikainen et al. (2014)
83	528-	CCCGGCCGTTATTTCTTGT	rev	467	oligotrich, choreotrich	Tamura et al. (2011)
344	MARa 502F	CAGAGATTTCAATGGGGATATTTAAYG	fwd	502	Neobodo designis marine clade	von der Heyden and Cavalier-Smith (2005)
250	Hap-M258f	AACTTTTAGCGTCCAGCCCA	fwd	528	Minchinia mytili	Ward et al. (2019)
232	Euk-516r	ACCAGACTGGCCCTCC	rev	547		Amann et al. (1990)
65	Uni18SF	AGGGCAAKYCTGGTGCCAGC	fwd	549		Zhan et al. (2013)
9	3NDf	GGCAAGTCTGGTGCCAG	fwd	551		Cavalier-Smith et al. (2009)
156	NS2	GGCTGCTGGCACCAGACTTGC	rev	552		White et al. (1990)
157	NS3	GCAAGTCTGGTGCCAGCAGCC	fwd	552		White et al. (1990)

id	Name	Sequence	Direction	Start	Specificity	Reference
13	515F	GTGCCAGCMGCCGCGGTAA	fwd	561		Parfrey et al. (2014)
19	515FY	GTGYCAGCMGCCGCGGTAA	fwd	561		Parada et al. (2015)
25	EUK581-F	GTGCCAGCAGCCGCG	rev	561	non-Metazoan	Carnegie et al. (2003)
31	515F Univ	GTGYCAGCMGCCGCGGTAA	fwd	561		Needham and Fuhrman (2016)
320	530R	CCGCGGCKGCTGGCAC	rev	561	Microsporidia	Williams et al. (2018)
4	563f	GCCAGCAVCYGGGTAAY	fwd	563		Hugerth et al. (2014)
7	V4 1f	CCAGCASCYGGGTAATWCC	fwd	564		Bass et al. (2016)
8	TAREuk454FWD1	CCAGCASCYGGGTAATWCC	fwd	564		Stoeck et al. (2010)
67	Claudia Vannini (F)	CCAGCASCYGGGTAATWCC	fwd	564	ciliates	Boscaro et al. (2017)
187	EuF-V4	CCAGCASCYGGGTAATWCC	fwd	564		Belevich et al. (2017)
218	TAREuk454FWD1 Choi	CCAGCAGCCGCGGTAATWCC	fwd	564		Choi and Park (2020)
1	F-566	CAGCAGCCGCGGTAATWCC	fwd	565		Hadziavdic et al. (2014)
369	568	GGTSTAAATTCRKYTCATTKC	rev	568	ciliates	Doherty et al. (2007)
10	EUKAF	GCCGCGGTAATCCAGCTC	fwd	570		Moreno et al. (2018)
69	ParaV45F	GCYCGGTAATWCCAGCTCT	fwd	570	Parabasalids	Jasso-Selles et al. (2017 )
12	E572F	CYCGGTAATCCAGCTC	fwd	571		Comeau et al. (2011)
14	528F	CCGCGGTAATCCAGCTC	fwd	571		Zhu et al. (2005)
17	NSF563	CGCGGTAATCCAGCTCCA	fwd	572		Mangot et al. (2013)
75	SSU566F	CGCGGTAATCCAGCTYC	fwd	572	dinoflagellates	Smith et al (2017)
2	A-528F	GCGGTAATCCAGCTCAA	fwd	573		Cheung et al. (2010)
73	DIV4for	GCGGTAATCCAGCTCCAATAG	fwd	573	diatoms	Visco et al. (2015)
3	574*f	CGGTAAYTCCAGCTCYV	fwd	574		Hugerth et al. (2014)
15	590F	CGGTAATCCAGCTCCAATAGC	fwd	574		Venter et al. (2017)
32	Euk528F	CGGTAATCCAGCTCC	fwd	574		Edgcomb et al. (2011)
132	574f	CGGTAAYTCCAGCTCYAV	fwd	574		Hugerth et al. (2014)
20	D512for	ATCCAGCTCCAATAGCG	fwd	579	diatoms	Zimmermann et al. (2011)
199	FF1100	CCAGCTCCAATAGCGTATATTA	fwd	582	Fungi	Vainio and Hantula (2000)
207	S32 J	CCAGCTCCAATAGCGTATAC	fwd	582	Radiolaria	Decelle et al. (2012)
208	S32 TASN	CCAGCTCCAATAGCGTATRC	fwd	582	Radiolaria	Ishitani et al. (2012)
21	Cerc479F	TGTTGCAGTAAAAAGCTCGT	fwd	608		Harder et al. (2016)
16	EK-565F-NGS	GCAGTAAAAAGCTCGTAGT	fwd	612		Simon et al. (2015)
152	SAR V3 R	RACTACGAGCTTTTAACTGC	rev	612	SAR	Sisson et al. (2018)
5	616f	TTAAAAGVYTCGTAAGTYG	fwd	616		Hugerth et al. (2014)
6	616*f	TTAAARVGYTCGTAAGTYG	fwd	616		Hugerth et al. (2014)
90	S616F Cerco	TTAAAAGCTCGTAGTTG	fwd	616	Cercozoa	Fiore-Donno et al. (2018)
91	S616F Eocer	TTAAAAGCGCGTAGTTG	fwd	616	Cercozoa	Fiore-Donno et al. (2018)
253	Hap-E620r	GGAGCCAAATCCGAGGACTT	rev	620	Haplosporidium sp.	Ward et al. (2019)
376	AMV4.5NF	AAGCTCGTAGTTGAATTTCC	fwd	621	mycorrhizal fungi	Sato et al. (2005)
342	FWb 681F	GGAGTCGGTTACGTCCCRCTCCGRRYCG	fwd	681	Neobodo designis freshwater clade	von der Heyden and Cavalier-Smith (2005)
279	LGD-698	GCTTAGTCTTCTCGTCTTAGGA	fwd	698	Cyclotrichiid Ciliates	Bass et al. (2009)
99	Chryso 651	CTATTTTGCTCAGATAAATGACGAG	rev	735	Chrysophyceae	Lentendu, G. et al. (2014)
303	V4r-d5b	GGATGACAATGTTTCCGGTGA	rev	740	Reticulamoeba gemmipara	Bass et al. (2012)
105	Kineto 651	TTGGTCGCRCTTYTTAGTCACAG	rev	746	Kinetoplastea	Lentendu, G. et al. (2014)
274	817F	TTAGCATGGAATAATRRATAGGA	fwd	793		Yang et al. (2020)
23	ChloroF	TGGCCTATCTTGTGGTCTGT	fwd	822	Chlorophyceae	Valiente Moro et al. (2009)
190	HaptoR1	CGAAACCAACAAAATAGCAC	rev	823	Prymnesiophyceae	Egge et al. (2013)
296	Gv847F	ATCATTYAGCATGGAATAAACAYAAC	fwd	847	Sainouroids	Bass et al. (2016)
304	V4r-d5a	CTCGGATTCCTGAAACCAATG	rev	850	Reticulamoeba spp.	Bass et al. (2012)
206	S879	CCAACGTCCCTATCAATCAT	rev	855	Radiolaria	Decelle et al. (2012)
377	AMDGR	CCCAACTATCCCTATTAATCAT	rev	855	mycorrhizal fungi	Sato et al. (2005)
244	mik868F	GGACTACCAGWGGCGAAAGCGCCT	fwd	868	Mikrocytid	Hartikainen et al. (2014)
89	Cer1R	ATACTAGCACCCCAACT	rev	870	Cercozoa	Lentendu, G. et al. (2014)
52	Cerc750R	TGAATACTAGCACCCCAAC	rev	871	Cercozoa	Harder et al. (2016)
74	DIV4rev3	CTCTGACAATGGAATACGAATA	rev	879	diatoms	Visco et al. (2015)
22	DimA	RGGACRGGTGAATAGGATG	fwd	893	diplonemids	Cannon et al. (2018)
76	SSU911R	ATYCAAGAATTTACCTCTGAC	rev	894	dinoflagellates	Smith et al. (2017)
146	690R	ATCCAAGAATTTACCTCTGAC	rev	894		Alves-de-Souza et al (2011)
77	B-706R	AATCCRAGAATTTACCTCT	rev	897		Cheung et al. (2010)
127	897f	AGAGGTGRAATTCTHRGA	fwd	897		Hugerth et al. (2014)
128	897r	TCYDAGAATTCACCTCT	rev	897		Hugerth et al. (2014)
337	Kin1240rev	GCCTTCGCTGTAGTTTCGTC	rev	924	Kinetoplastea	Scheckenbach et al. (2010)
35	R-952	TTGGCAAATGCTTTCGC	rev	935		Hadziavdic et al. (2014)
49	NSR951	TTGGYRAATGCTTTCGC	rev	935		Mangot et al. (2013)

id	Name	Sequence	Direction	Start	Specificity	Reference
93	S947R Cerco	AAGAAGACATCCTTGGTG	rev	946	Cercozoa	Fiore-Donno et al. (2018)
55	PRYM01+7	GATCAGTGAAAACATCCCTGG	rev	948	Haptophyta	Egge et al. (2013)
43	V4 18S Next.Rev	ACTTTCGTTCTTGATYRATGA	rev	960		Piredda et al. (2017)
92	S963R Cerco	CAACTTTCGTTCTTGATTA	rev	962	Cercozoa	Fiore-Donno et al. (2018)
37	TAREukREV3	ACTTTCGTTCTTGATYRA	rev	963		Stoeck et al (2010)
38	V4RB	ACTTTCGTTCTTGATYRR	rev	963		Balzano and Leterme (2015)
42	EUKAR	CYTTCCGYCTTGATTRA	rev	963		Moreno et al. (2018)
219	TAREukREV3 Choi	ACTTTCGTTCTTGATTA	rev	963		Choi and Park (2020)
196	Par 18S-R	CCTACTCTCGCYCTTGATCG	rev	964	Parabasalids	Michaud et al. (2020)
188	picoR2	AKCCCCYAACTTTCGTTCTTGAT	rev	966		Belevich et al. (2017)
202	V4r	ACTTTCGTTCTTGAT	rev	966		Bradley et al. (2016)
46	Nex 18S 0964 R	GATCCCYAACTTTCGTTCTTGA	rev	967		Kim et al. (2016)
228	1119R	TCCCCTAACTTTCGTTCTTG	rev	968		Kawachi et al. (2016)
68	Claudia Vannini (R)	TCTGRTYGTCTTGGATCCCYTA	rev	981	ciliates	Boscaro et al. (2017)
216	LABY-A	GGGATCGAAGATGATTAG	fwd	984	Labyrinthulomycetes	Stokes et al. (2002)
40	V4 euk R1	GACTACGACGGTATCTRATCRTCTTCG	rev	989		Bráte et al. (2010)
41	V4 euk R2	ACGGTATCTRATCRTCTTCG	rev	989		Bráte et al. (2010)
44	E1009R	AYGGTATCTRATCRTCTTYG	rev	989		Comeau et al. (2011)
66	Uni18SR	GRCGGTATCTRATCGYCTT	rev	991		Zhan et al. (2013)
51	D978rev	GACTACGATGGTATCTAATC	rev	996	diatoms	Zimmermann et al. (2011)
321	CM-V5F	GATTAGANACNNNGTAGTTC	fwd	996	Microsporidia	Trzebný et al. (2020)
324	18S Naslb R	GAGACTACGACGGTATCTGATC	rev	996	Nassellaria	Sandin et al. (2019)
193	Oxy 18S-F	ATCAGAWACCGYCGTAGTC	fwd	997	Oxymonads	Michaud et al. (2020)
198	FF700	GATACCGTNGTAGTCT	fwd	1001	Fungi	Vainio and Hantula (2000)
257	C5f-Hapl	GTAGTCCARCAYATAAACBATGTC	fwd	1010	Haplosporidia	Hartikainen et al. (2014b)
316	Oom1024R	CTCATAACGGTCTGACAAGG	rev	1024	Oomycetes	Holt et al. (2018)
370	1199+	GCCGACTCGGGATCGGGGGC	fwd	1031	ciliates	Doherty et al. (2007)
380	1199+	GCCGACTCGGGATCGGGGGC	fwd	1031	oligotrich, choreotrich	Tamura et al. (2011)
305	V5r-d5b	GTCAACGCTCGTGATCCCTG	rev	1055	Reticulamoeba spp.	Bass et al. (2012)
298	Gv1063F	AGCRAAAGCATTCATCAAT	fwd	1063	Sainouroids	Bass et al. (2016)
306	V5r-d5a	GGTGCCAACGAGGTCGTTTCA	rev	1075	Reticulamoeba spp.	Bass et al. (2012)
230	s14F3	ACGCAMGTGTGAAACTTG	fwd	1119	Foraminifera	Holzmann et al. (2003)
48	EUK1134-R	TTTAAGTTTCAGCCTTGCG	rev	1120		Carnegie et al. (2003)
265	UNonMet DB	CTTTAARTTTCASYCTTGCG	rev	1120	non-Metazoan	Bass and del Campo (2020)
30	926wF	AAACTYAAAKGAATTGRCGG	fwd	1130		Wilkins et al. (2013)
60	926R	CCGYCAATYMTTTRAGTTT	rev	1130		Needham and Fuhrman (2016)
159	NS5	AACTTAAAGGAATTGACGGAAG	fwd	1131		White et al. (1990)
36	1132r	CCGTCAATTHCTTYAART	rev	1132		Hugerth et al. (2014)
211	1132rmod	TCCGTCAATTYCTTTAAGT	rev	1132		Geisen et al. (2018)
158	NS4	CTTCGTCAATTCCTTTAAG	rev	1133		White et al. (1990)
224	1132R modified	CCGTCAATTHCTTYAAR	rev	1133		Hu et al. (2016)
70	ParaV45R	AAGRAATTGACGGAAGNGCA	rev	1137	Parabasalids	Jasso-Selles et al. (2018)
45	1119r	GGTGCCCTCCGTCA	rev	1144		Parfrey et al. (2014)
96	s14f1	AAGGGCACACAAGAACGC	fwd	1150	Foraminifera	de Vargas et al. (1997)
322	CM-V5R	TAANCAGCACAMTCCACTC	rev	1164	Microsporidia	Trzebný et al. (2020)
53	DimB	CAAATTGAGCCGAGACTCC	rev	1168		Cannon et al. (2018)
225	960F	GGCTTAATTTGACTCAACRCG	fwd	1177		Gast et al. (2004)
34	R-1200	CCCGTGTGAGTCAAATTAAGC	rev	1178		Hadziavdic et al. (2014)
129	F-1183	AATTGACTCAACACGGG	fwd	1182		Hadziavdic et al. (2014)
275	1196R	TCTGGACCTGGTGAGTTTCC	rev	1199		Yang et al. (2020)
291	1259F	GGTCRRGACAYAGTRAGGATTGACAGATTGAAG	fwd	1211	Cercozoa	Karpov et al. (2006)
94	1301f	GATTGAAGCTCTTTCTTGATCACTTC	fwd	1236	Plasmodiophorida	Bass et al. (2018)
288	1256R	GCACCACCACCAYAGAATCAAGAAAGAWCTTC	rev	1242	Cercozoa	Bass and Cavalier-Smith (2004)
343	FWb 1244R	TATTCTCTTTGGCGGGMTCAAGCAAGCGAG	rev	1244	Neobodo designis freshwater clade	von der Heyden and Cavalier-Smith (2005)
97	s15.3	CCTATCACATAATCATGAAAAG	rev	1247	Foraminifera	Pawlowski, J., et al., (2014)
148	1055R	ACGGCCATGCACCACCACCCAT	rev	1260		Alves-de-Souza et al (2011)
147	1055F	GGTGGTGCATGGCCCTTCTT	fwd	1266		Alves-de-Souza et al (2011)
47	1300R	CACCAACTAAGAACGGCCATGC	rev	1272		Venter et al (2017)
266	SSR-F1 289	TGGAGYGATTTGTCTGGTTDATTCCG	fwd	1292		Nagai et al. (2016)
54	ChloroR	GAATCAACCTGACAAGGCAAC	rev	1295	Chlorophyceae	Valiente Moro et al. (2009)
299	hxx1295R	TCAATCCACTCACTCCCAAAGGC	rev	1295	Sainouroids	Bass et al. (2016)
197	FF390	CGATAACGAACGAGACCT	fwd	1316	Fungi	Vainio and Hantula (2000)
345	MARa 1321R	GGACGTGCTGAGGATATCCCGWTA	rev	1321	Neobodo designis marine clade	von der Heyden and Cavalier-Smith (2005)

id	Name	Sequence	Direction	Start	Specificity	Reference
245	mik1340	TGCATCACGGACCTACCTTWGACC	rev	1340	Mikrocytid	Hartikainen et al. (2014)
217	LABY-Y	CWCRAAATTCCTCCGGT	rev	1400	Labyrinthulomycetes	Stokes et al. (2002)
160	NS6	GCATCACAGACCTGTATTGCCTC	rev	1415		White et al. (1990)
161	NS7	GAGGCAATAACAGGTCTGTGATGC	fwd	1415		White et al. (1990)
221	Nex 18S 1434 F	GAGGCAATAACAGGTCTGTGATG	fwd	1415		Kim et al. (2016)
201	V8f	ATAACAGGTCTGTGATGCCCT	fwd	1421		Bradley et al. (2016)
203	1422f	ATAACAGGTCTGTGATGC	fwd	1421		Hadziavdic et al. (2016)
226	NSR1438	GGGCATCACAGACCTGTAT	rev	1421		Van De Peer et al. (2000)
194	Oxy 18S-R	GGGCATMACRGACCTGTTA	rev	1422	Oxymonads	Michaud et al. (2020)
204	1424f	AACAGGTCHGWRATGCC	fwd	1423		Hugerth et al. (2014)
131	R-1443	AAGGCATCACAGACCTG	rev	1425		Hadziavdic et al. (2014)
297	hxx1442R	ATCTAAGAGCATCACGGACCTTTTATC	rev	1442	Sainouroids	Bass et al. (2016)
243	mik1511	CCTATTACGCGCTCTGTTGAGA	rev	1511	Mikrocytid	Hartikainen et al. (2014)
238	SL175pr5F	ACGAGGAATGCCTAGTAAGCGCAA	fwd	1569	Mantoniella antarctica	Gast et al. (2014)
330	Tel3250-3230R	GACGTAATCAGGGCGTCT	rev	1590	Telonemia	Brâte et al. (2010)
277	ParaGENrGW	GTGTACAAGGRCAGGGACT	rev	1614	Paramyxids	Ward et al. (2016)
27	1380F	CCCTGCCHTTTGTACACAC	fwd	1617		Amaral Zettler et al (2009)
318	Microsp1342r	ACGGGCGGTGTACAAAAGAACAG	rev	1619	Microsporidia	Stentiford et al. (2017)
62	U1391R	GGGCGGTGTGTACAARGR	rev	1623		Edgcomb et al. (2011)
28	1389F	TTGTACACACCGCCC	fwd	1626		Amaral Zettler et al (2009)
29	1388F	TTGTACACACCGCCGTCGC	fwd	1626		Piredda et al. (2017)
59	1392-R	ACGGGCGGTGTGTRC	rev	1628		Wilkins et al. (2013)
145	18r71	GCGACGGGCGGTGTATC	rev	1628		Alves-de-Souza et al (2011)
166	ITS9MUNgs	TACACACGCCCGTCG	fwd	1629		Tedersoo and Lindahl (2016)
200	FR1	ANCCATTCAATCGGTANT	rev	1647	Fungi	Vainio and Hantula (2000)
309	ALU4	RTCTCACTAAGCCATTC	rev	1657	Fungi	Vandenkoornhuyse et al. (2002)
295	1682R	ATCCGTGAAGCTCACTAATC	rev	1682	Glissomonads	Howe et al. (2009)
290	1733R	TGATCAAGTTTGATTAGTTCTCGGAT	rev	1733	Cercomonas-clade A	Karpov et al. (2006)
260	sB2hap	CCTTGTTACGACTTBTYCTTCCTC	rev	1744	Haplosporidia	Hartikainen et al. (2014b)
307	sB2-d5	CCTTGTTACGACTTTTGC	rev	1750	Reticulamoeba spp. + some eukaryotes	Bass et al. (2012)
332	Heterokonta Rev	GGTTCACCTACGAAACCTTGTACGACTTCA	rev	1752	Heterokonta	Scheckenbach et al. (2010)
95	1801r	ACGGAAACCTTGTACGACTTC	rev	1753	Plasmodiophorida	Bass et al. (2018)
61	U1492R	GGTTACCTTGTACGACTT	rev	1754		Edgcomb et al. (2011)
63	U1517R	ACGGCTACCTTGTACGACTT	rev	1754		Edgcomb et al. (2011)
80	1818r	ACGGAAACCTTGTACGA	rev	1757		Lepere et al. (2011)
267	SSR-R1 772	TCACCTACGAAACCTTGTACG	rev	1758		Nagai et al. (2016)
235	18S-1498R	CACCTACGAAACCTTGTTA	rev	1760		López-García et al. (2003)
222	Nex 18S 1757 R	CAGGTTCACTACGAAACCT	rev	1765		Kim et al. (2016)
371	1765-	CCCCAKCACGACDCMTATTGCTG	rev	1765	ciliates	Doherty et al. (2007)
237	RS11pr4R	CTGCAGGTTACCTACGAAACC	rev	1766	Pyramimonas cf. tychotreta	Gast et al. (2014)
162	NS8	TCCGCAGTTTACCTACGGA	rev	1770		White et al. (1990)
259	sB1N	GATCCHTCYGCAGTTACCTACG	rev	1772		Hartikainen et al. (2014b)
262	Sb1n	GATCCHTCYGCAGTTACCTACG	rev	1772	Paradinids	Ward et al. (2018)
57	EukB	TGATCCTTCTGCAGTTACCTAC	rev	1773		Medlin et al. (1988)
58	1510R	CCTTCYGCAGTTACCTAC	rev	1773		López-García et al. (2003)
143	1801R	TGATCCTTCTGCAGTTACCT	rev	1775		Cavalier-Smith et al. (2009)
336	18SRevBodo	TGATCCAGCTGCAGTTACCC	rev	1776	Kinetoplastea	Scheckenbach et al. (2010)
312	Diphy1881R	CGACAAAACCTCAAAGATTC	rev	1860	Diphyllatea	Orr et al. (2018)
347	Helio1979R	CACACTTACWAGGAYTTCCTCGTTSAGACG	rev	1979	Centrohelid heliozoa	Cavalier-Smith and von der Heyden (2007)
335	kineto2026R	GATCCTTCTGCAGTTACCTACGCT	rev	2026	Kinetoplastea	von der Heyden and Cavalier-Smith (2005)
106	SRAca28	CCAATTACAAGACTCTTRTCGAG	fwd		Acanthamoeba	Fiore-Donno (2016)
108	SR19Dark	GTCCTTAATTGTTACTCGAD	fwd		Myxomycetes	Fiore-Donno (2016)
112	Pdir1	GATTCGGGGCGGGTTCCA	fwd		Pedinophyceae	Milyutina et al. (2019)
113	Pdir2	GATCGGGCTTCGGTTCCGAG	fwd		Pedinophyceae	Milyutina et al. (2019)
114	Pprev2	CTCGCGAACTCGAACCGAAG	rev		Pedinophyceae	Milyutina et al. (2019)
115	Pdir3	CCTCAGCCTGCTAAATAGCTAC	fwd		Pedinophyceae	Milyutina et al. (2019)
116	Pdir4	GACTTTCGGGGTTTTACCCGGA	fwd		Pedinophyceae	Milyutina et al. (2019)
134	S19F	GTGCATGGCCGTTCTTAGTTC	rev		Foraminifera	Morard et al. (2011)
135	S15rF	CCCGTACRAGGCATTCTAG	fwd		Foraminifera	Morard et al. (2011)
144	329R	GTGAACCTGCRGAAGGATCA	rev			Alves-de-Souza et al (2011)
170	Pb121	GGATACAAAACCAACCTGGC	fwd		Plasmodiophora	Niwa et al. (2011)
171	Pb121r	GCCAGGTTGGTTTTGTATCC	rev		Plasmodiophora	Niwa et al. (2011)
186	SB	GTAGGTGAACCTGCAGAAGGATCA	rev			Sogin (1990)

id	Name	Sequence	Direction	Start Specificity	Reference
192	PRYM03+3	GTA AATTGCCCGAATCCTG	fwd	Prymnesiophyceae	Egge et al. (2013)
205	17	CGGTCACGTTTCGTTGC	rev	Foraminifera	Cordier et al. (2019)
210	S51 TAS	YAAGAATTTACCTCTCGCTT	rev	Radiolaria	Ishitani et al. (2012)
214	QPX-F	ATCCTCGGCCTGCTTTTAGTAG	fwd	Quahog parasite	Stokes et al. (2002)
215	QPX-R2	GAAGTCTCTACCTTTCTTGCGA	rev	Quahog parasite	Stokes et al. (2002)
236	RS11pr4F	ATGTTCCGATCGCGGCGAGAC	fwd	Pyramimonas cf. tychotreta	Gast et al. (2014)
239	SL175pr5R	TAGAAAGCCACGGTCCGAACGC	rev	Mantoniella antarctica	Gast et al. (2014)
240	Gempr2F	TGGGATTGCTGGGTAGAACTTCGT	fwd	Geminigera cryophila	Gast et al. (2014)
241	Gempr2R	CACCTACGGGAAACCTTGTTACGAC	rev	Geminigera cryophila	Gast et al. (2014)
247	Plas1r	GGTGCSKCKAGRTVCAAGAGGC	rev	Plasmodiophorids	Neuhauser et al. (2014)
248	Plas2f	TGGATGTACGAGAGTACTACATGG	fwd	Plasmodiophorids	Neuhauser et al. (2014)
249	Plas2r	CGTTGAACCTAGCATTGTAGCG	rev	Plasmodiophorids	Neuhauser et al. (2014)
258	V5f-Hapl	GGACTCRGGGGGAAGTATGCT	fwd	Haplosporidia	Hartikainen et al. (2014b)
261	V4fAsce	GGAATAATAWAGATAGGACTTCRGCA	fwd	Paradinids	Ward et al. (2018)
263	V5fAsce	GYTCRGACCKTATTYGAGAAATCA	fwd	Paradinids	Ward et al. (2018)
264	EndoR1	CGACTTCTCCTTCCTAARYRDTAWG	rev	Paradinids	Ward et al. (2018)
276	Para1fGW	GGGCGAGGGGTAAATCT	fwd	Paramyxids	Ward et al. (2016)
278	Para3fGW	GGCTTYTGGGAGAKTACGG	fwd	Paramyxids	Ward et al. (2016)
281	myxo 617F all	CGCGCAAATACCAMTCCA	fwd	Myxozoa	Hartikainen et al. (2016)
282	myxo 764F all	CCGCGTAATCCAGCTCCAG	fwd	Myxozoa	Hartikainen et al. (2016)
283	myxo 2313R all	CGTTACCGGAATRRCTGACAG	rev	Myxozoa	Hartikainen et al. (2016)
284	myxo 1817 v1	ATTTACCTCTCGCCATCGA	rev	Myxozoa	Hartikainen et al. (2016)
285	myxo 1817 v2	ATTTACCTCTCGCGCMAA	rev	Myxozoa	Hartikainen et al. (2016)
286	myxo 1817 v3	ATTTACCTCTCGCTGCCAA	rev	Myxozoa	Hartikainen et al. (2016)
317	CTMicrosp	CACCAGGTTGATTCTGCCTGACG	fwd	Microsporidia	Stentiford et al. (2017)
319	V1F	CACCAGGTTGATTCTGCCTGAC	fwd	Microsporidia	Williams et al. (2018)
323	APU-1R	CTTCTTTGGTTAAACAC	rev	Apusomonads	Torruella et al. (2017)
325	18S NassII F	AGCATGGAATAATAACTGATGA	fwd	Nassellaria	Sandin et al. (2019)
326	18S NassII R	CACCARTTCATCCAATCGGTAG	rev	Nassellaria	Sandin et al. (2019)
340	DiploF	GATATCTAAACCTGTC	fwd	diplonemids	Lara et al. (2009)
341	DiploR	GCATTCTCATTCAAGGA	rev	diplonemids	Lara et al. (2009)
378	BaciF	AGATTGCCAGGCCTCTCG	fwd	Bacillariophyceae	Valiente Moro et al. (2009)
379	BaciR	CCATCGTAGTCTTAACCATAAAC	rev	Bacillariophyceae	Valiente Moro et al. (2009)
381	1765-	CCCCACACGACDCMTATTGCTG	rev	oligotrich, choreotrich	Tamura et al. (2011)
172	Pbr2r	CTCTATGCCGAATCGCTTC	rev	Plasmodiophora	Niwa et al. (2011)
173	Pbr4	GTGTCGCTTAAGATATAGTC	fwd	Plasmodiophora	Niwa et al. (2011)
174	Pbr4r	GACTATATCTTAAGCGACAC	rev	Plasmodiophora	Niwa et al. (2011)

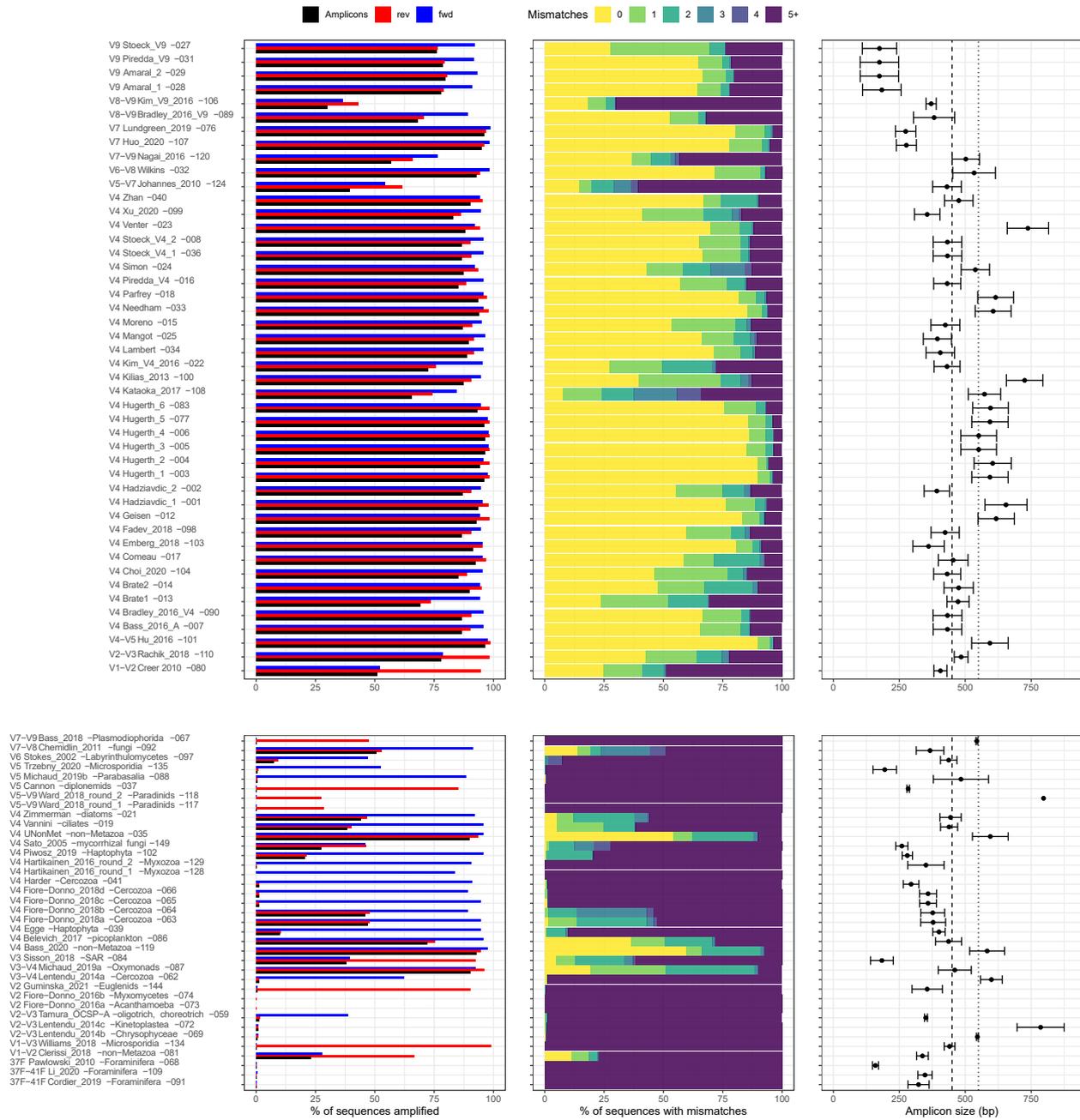
**Table S2:** List of 18S rRNA primer sets used for metabarcoding in the pr2-primers database. Size corresponds to the average amplicon size (bp) for sequences from the PR2 database. DOI for reference can be found in the on-line web application.

id	Name	Primer fwd	Primer rev	Region	Size	Specificity	Reference
1	Hadziavdic 1	F-566	R-1200	V4	654		Hadziavdic et al. (2014)
2	Hadziavdic 2	A-528F	R-952	V4	392		Hadziavdic et al. (2014)
3	Hugerth 1	574*f	1132r	V4	594		Hugerth et al. (2014)
4	Hugerth 2	563f	1132r	V4	604		Hugerth et al. (2014)
5	Hugerth 3	616f	1132r	V4	551		Hugerth et al. (2014)
6	Hugerth 4	616*f	1132r	V4	551		Hugerth et al. (2014)
7	Bass 2016 A	V4 1f	TAReukREV3	V4	433		Bass et al. (2016)
8	Stoeck V4 2	TAReuk454FWD1	TAReukREV3	V4	433		Stoeck et al (2010)
12	Geisen	3NDf	1132rmod	V4	617		Geisen et al. (2018)
13	Brate1	3NDf	V4 euk R1	V4	473		Br�ate et al. (2010)
14	Brate2	3NDf	V4 euk R2	V4	475		Br�ate et al. (2010)
15	Moreno	EUKAF	EUKAR	V4	425		Moreno et al. (2018)
16	Piredda V4	TAReuk454FWD1	V4 18S Next.Rev	V4	432		Piredda et al. (2017)
17	Comeau	E572F	E1009R	V4	454		Comeau et al. (2011)
18	Parfrey	515F	1119r	V4	615		Parfrey et al. (2014)
19	Vannini	Claudia Vannini (F)	Claudia Vannini (R)	V4	439	ciliates	Boscaro et al. (2017)
21	Zimmerman	D512for	D978rev	V4	444	diatoms	Zimmermann et al. (2011)
22	Kim V4 2016	528F	Nex 18S 0964 R	V4	431		Kim et al. (2016)
23	Venter	590F	1300R	V4	738		Venter et al (2017)
24	Simon	EK-565F-NGS	EUK1134-R	V4	538		Simon et al. (2015)
25	Mangot	NSF563	NSR951	V4	394		Mangot et al. (2013)
27	Stoeck V9	1391F	EukB	V9	175		Stoeck et al (2010)
28	Amaral 1	1380F	1510R	V9	184		Amaral Zettler et al (2009)
29	Amaral 2	1389F	1510R	V9	175		Amaral Zettler et al (2009)
31	Piredda V9	1388F	1510R	V9	175		Piredda et al. (2017)
32	Wilkins	926wF	1392-R	V6-V8	534		Wilkins et al. (2013)
33	Needham	515F Univ	926R	V4	606		Needham and Fuhrman (2016)
34	Lambert	515FY	NSR951	V4	405		Lambert et al. (2019)
35	UNonMet	EUK581-F	EUK1134-R	V4	595	non-Metazoa	Carnegie et al. (2003)
36	Stoeck V4 1	TAReuk454FWD1	V4RB	V4	433		Balzano and Leterme (2015)
37	Cannon	DimA	DimB	V5	284	diplonemids	Cannon et al. (2018)
39	Egge	A-528F	PRYM01+7	V4	400	Haptophyta	Egge et al. (2013)
40	Zhan	Uni18SF	Uni18SR	V4	476		Zhan et al. (2013)
41	Harder	Cerc479F	Cerc750R	V4	295	Cercozoa	Harder et al. (2016)
59	Tamura OCSP-A	152+	528-	V2-V3	351	oligotrich, choreotrich	Tamura et al. (2011)
62	Lentendu 2014a	Cer2F	Cer1R	V3-V4	599	Cercozoa	Lentendu et al. (2014)
63	Fiore-Donno 2018a	S616F Cerco	S963R Cerco	V4	378	Cercozoa	Fiore-Donno et al. (2018)
64	Fiore-Donno 2018b	S616F Eocer	S963R Cerco	V4	377	Cercozoa	Fiore-Donno et al. (2018)
65	Fiore-Donno 2018c	S616F Cerco	S947R Cerco	V4	359	Cercozoa	Fiore-Donno et al. (2018)
66	Fiore-Donno 2018d	S616F Eocer	S947R Cerco	V4	359	Cercozoa	Fiore-Donno et al. (2018)
67	Bass 2018	1301f	1801r	V7-V9	544	Plasmodiophorida	Bass et al. (2018)
68	Pawlowski 2010	s14f1	s15.3	37F	160	Foraminifera	Pawlowski and Lecroq (2010)
69	Lentendu 2014b	Chryso 240	Chryso 651	V2-V3	546	Chrysophyceae	Lentendu et al. (2014)
72	Lentendu 2014c	Kineto 80	Kineto 651	V2-V3	786	Kinetoplastea	Lentendu et al. (2014)
73	Fiore-Donno 2016a	SRAca28	SFAca22	V2		Acanthamoeba	Fiore-Donno et al. (2016)
74	Fiore-Donno 2016b	SR19Dark	SF2Dark	V2		Myxomycetes	Fiore-Donno et al. (2016)
76	Lundgreen 2019	F-1183	R-1443	V7	274		Lundgreen et al. (2019)
77	Hugerth 5	574f	1132r	V4	594		Hugerth et al. (2014)
80	Creer 2010	F04	R22	V1-V2	406		Creer et al. (2010)
81	Clerissi 2018	18SV1V2F	18SV1V2R	V1-V2	338	non-Metazoa	Clerissi et al. (2018)
83	Hugerth 6	A-528F	1132r	V4	596		Hugerth et al. (2014)
84	Sisson 2018	SAR V3 F	SAR V3 R	V3	184	SAR	Sisson et al. (2018)
86	Belevich 2017	EuF-V4	picoR2	V4	437	picoplankton	Belevich et al. (2017)
87	Michaud 2019a	Oxy 18S-F	Oxy 18S-R	V3-V4	460	Oxymonads	Michaud et al. (2020)
88	Michaud 2019b	Par 18S-F	Par 18S-R	V5	484	Parabasalia	Michaud et al. (2020)
89	Bradley 2016 V9	V8f	1510R	V8-V9	382		Bradley et al. (2016)
90	Bradley 2016 V4	TAReuk454FWD1	V4r	V4	433		Bradley et al. (2016)
91	Cordier 2019	s14f1	17	37F-41F	323	Foraminifera	Cordier et al. (2019)
92	Chemidlin 2011	FF390	FR1	V7-V8	367	fungi	Chemidlin Prevost-Boure et al. (2011)

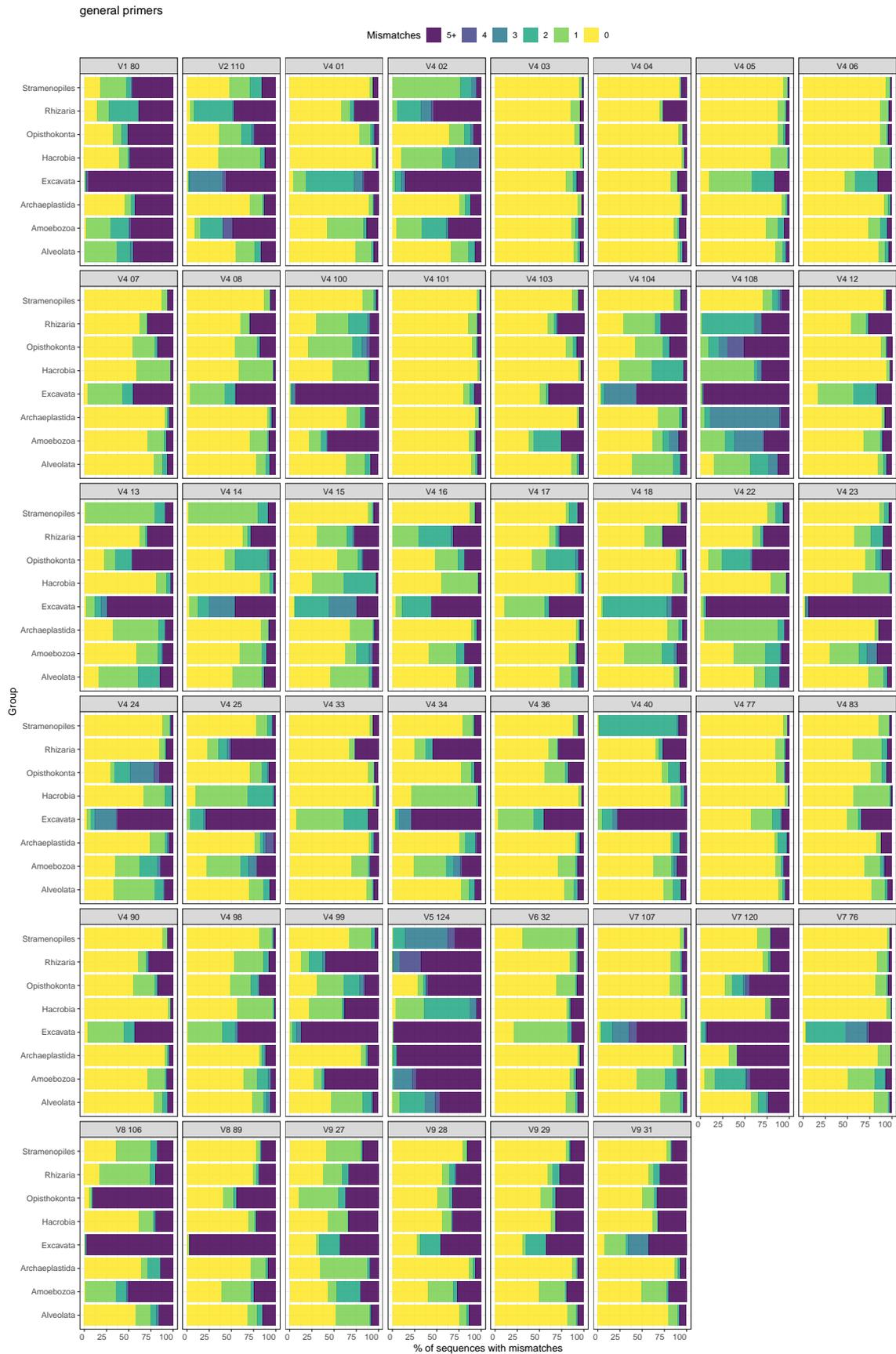
id	Name	Primer fwd	Primer rev	Region	Size	Specificity	Reference
97	Stokes 2002	LABY-A	LABY-Y	V6	437	Labyrinthulomycetes	Stokes et al. 2002
98	Fadeev 2018	A-528F	V4RB	V4	424		Fadeev et al. (2018)
99	Xu 2020	A-528F	B-706R	V4	356		Xu et al. (2020)
100	Kiliias 2013	A-528F	1055R	V4	725		Kiliias et al. (2013).
101	Hu 2016	574*f	1132R modified	V4-V5	594		Hu et al. (2016)
102	Piwosz 2019	TAReuk454FWD1	HaptoR1	V4	280	Haptophyta	Piwosz (2019)
103	Emberg 2018	E572F	897r	V4	361		Emberg et al. (2018)
104	Choi 2020	TAReuk454FWD1 Choi	TAReukREV3 Choi	V4	432		Choi and Park (2020)
106	Kim V9 2016	Nex 18S 1434 F	Nex 18S 1757 R	V8-V9	371		Kim et al. (2016)
107	Huo 2020	960F	NSR1438	V7	277		Huo et al. (2020)
108	Kataoka 2017	545F	1119R	V4	573		Kataoka et al. (2017)
109	Li 2020	s14F3	17	37F-41F	347	Foraminifera	Li et al. (2020)
110	Rachik 2018	18S-82F	Euk-516r	V2-V3	484		Rachik et al. (2018)
117	Ward 2018 round 1	V4fAsce	Sb1n	V5-V9	995	Paradinids	Ward et al. (2018)
118	Ward 2018 round 2	V5fAsce	EndoR1	V5-V9	797	Paradinids	Ward et al. (2018)
119	Bass 2020	574*f	UNonMet DB	V4	583	non-Metazoa	Bass and del Campo (2020)
120	Nagai 2016	SSR-F1 289	SSR-R1 772	V7-V9	502		Nagai et al. (2016)
124	Johannes 2010	817F	1196R	V5-V7	431		Yang et al. (2020)
128	Hartikainen 2016 round 1	myxo 617F all	myxo 2313R all	V4	996	Myxozoa	Hartikainen et al. (2016)
129	Hartikainen 2016 round 2	myxo 764F all	myxo 1817 v1	V4	351	Myxozoa	Hartikainen et al. (2016)
134	Williams 2018	V1F	530R	V1-V3	441	Microsporidia	Williams et al. (2018)
135	Trzebny 2020	CM-V5F	CM-V5R	V5	195	Microsporidia	Trzebny et al. (2020)
144	Guminska 2021	18S V2i F	18S V2i R	V2	356	Euglenids	Guminska et al. (2021)
149	Sato 2005	AMV4.5NF	AMDGR	V4	260	mycorrhizal fungi	Sato et al. (2005)

**Table S3:** Overall statistics for *in silico* % amplification of PR<sup>2</sup> sequences for primer sets listed in the pr2-primers database.

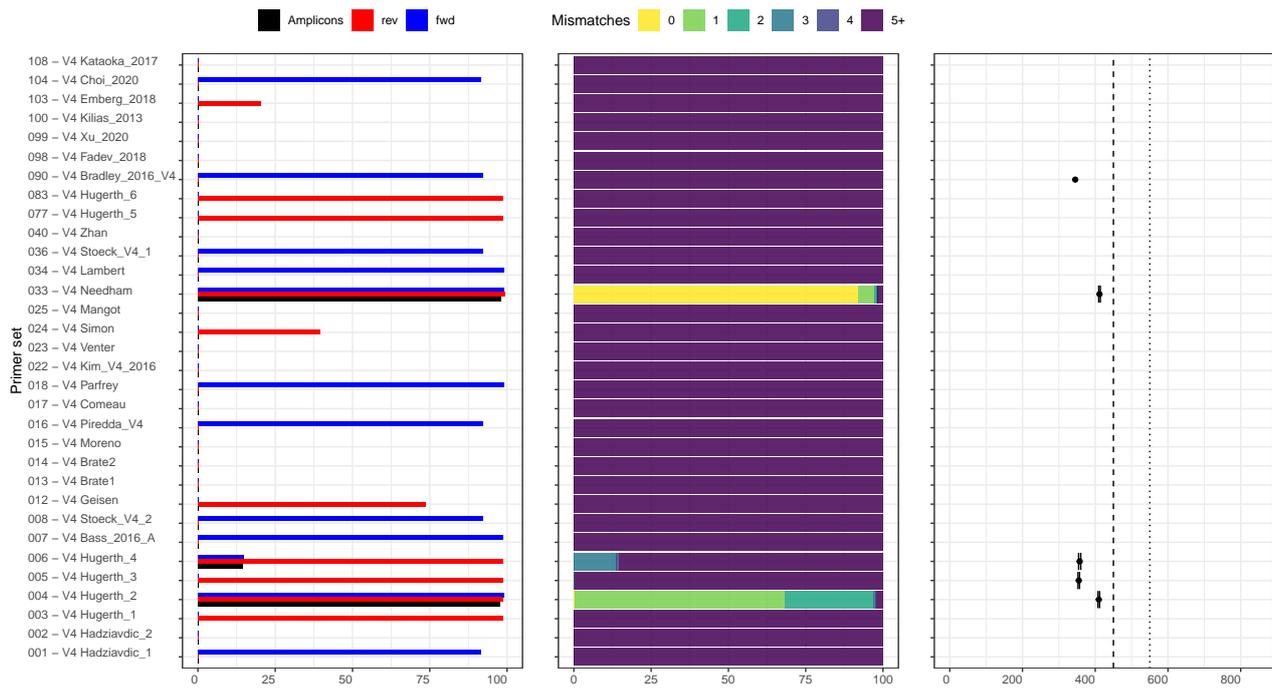
	general	specific
<b>forward primers</b>		
min	36.4	0.0
mean	91.0	49.7
max	98.7	97.6
<b>reverse primers</b>		
min	43.2	0.0
mean	88.7	32.4
max	98.6	98.9
<b>primer sets</b>		
min	30.0	0.0
mean	83.4	18.6
max	96.5	92.7



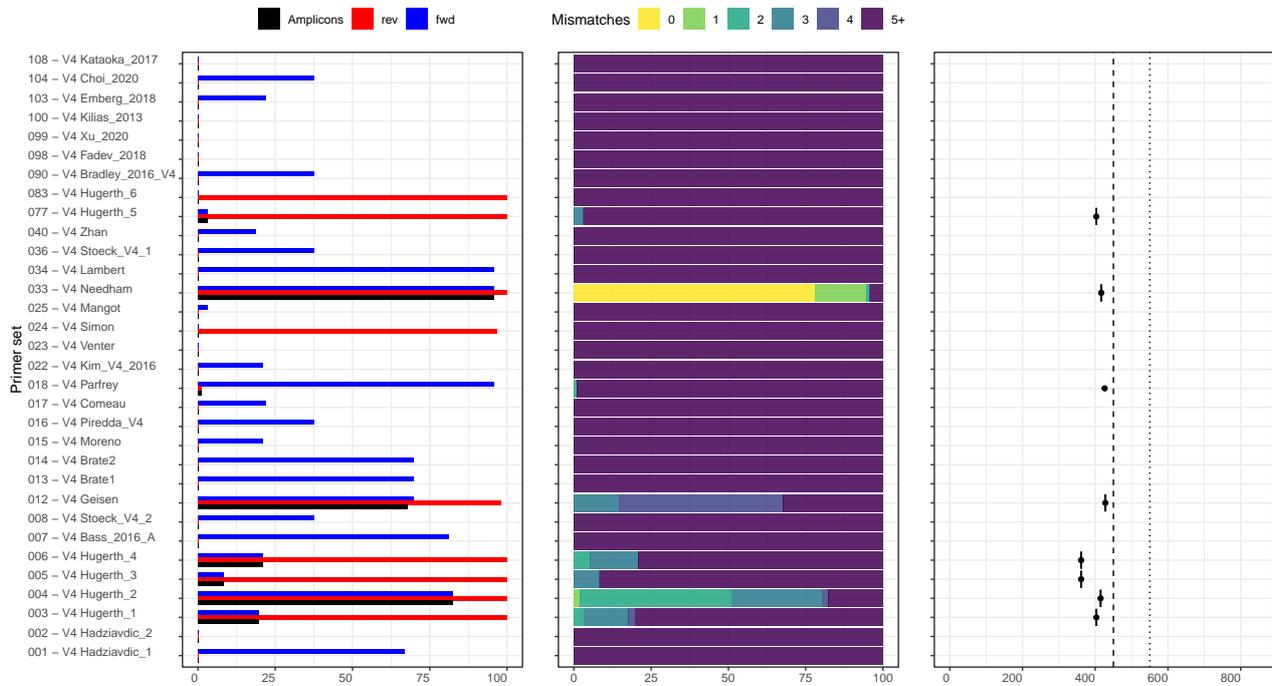
**Figure S1:** Evaluation of all general (top) or specific (bottom) primer sets (Table S2) for the 18S rRNA gene against the PR<sup>2</sup> reference database (version 4.12.0). See Fig. 2 for legend.



**Figure S2:** Number of mismatches for general primer sets as a function of the supergroup.



**Figure S3:** Evaluation of general primer sets (Table S2) targeting the V4 region of the 18S rRNA gene against bacterial 16S rRNA sequences from the Silva seed reference database (version 132). Legend as in Figure 2.



**Figure S4:** Evaluation of general primer sets (Table S2) targeting the V4 region of the 18S rRNA gene against archaeal 16S rRNA sequences from the Silva seed reference database (version 132). Legend as in Figure 2.



**Figure S5:** Number of mismatches for specific primer sets as a function of the supergroup. Target group is indicated inside the corresponding supergroup bar (e.g., Foraminifera are inside Cercozoa).

