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Correction to: A comprehensive fungispecific 18S rRNA gene sequence primer toolkit suited for diverse research issues and sequencing platforms



Stefanos Banos¹, Guillaume Lentendu^{2,3}, Anna Kopf⁴, Tesfaye Wubet^{2,5,7}, Frank Oliver Glöckner^{4,6} and Marlis Reich^{1*}

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Following publication of the original article [1], we have been notified that three of the primer names identified as most promising candidates for fungal community surveys were incorrectly renamed following the primer nomenclature system proposed by Gargas & DePriest [2]. Their positioning on the reference sequence had to be shifted 1bp towards the 3'-end (see Table 1 for the correct naming). The same error occurred in some primer names listed in the additional files (see attached Supplementary information).

As consequence, the number of identical nucleotides shared by the most promising primers and the newly designed blocking oligo sequences changed (see Table 2).

In this correction, the revised supplementary materials are included.

Supplementary information

Supplementary information accompanies this paper at https://doi.org/10. 1186/s12866-019-1628-y.

Additional file 1. List of the 164 fungi-specific primers detected by a literature research. For each primer, performance, characteristics and source literature are provided.

Additional file 2. List of the 436 fungi-specific primer pairs tested for their performance by in silico PCR. Primer pairs were grouped according to the expected amplicon size into three groups: S for small (\leq 600 bp), M for medium (600–1000 bp), and L for large size (> 1000 bp).

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¹Molecular Ecology, Institute of Ecology, FB02, University of Bremen, Leobener Str. 2, 28359 Bremen, Germany

Full list of author information is available at the end of the article

Additional file 3. List of the seven most promising primer pairs for biodiversity assessments identified by in silico PCR. Primer pairs are suitable for different sequencing methods dependent on the expected amplicon size. Sequence coverage rate of diverse fungal and non-fungal eukaryotic groups as revealed by in silico PCR.

Additional file 4. Annealing temperatures empirically evaluated for the most promising primer pairs. Two fungal strains, one of the Basidiomycota and one of the Ascomycota, served as template DNA. Intensity of the color indicates the strength of the amplification product detected by ethidium bromide staining. Red, template DNA from *Taphrina deformans*; Green, template DNA from *Agaricus bisporus*; *, optimal annealing temperature.

Additional file 5. Performance of the most promising primer pairs empirically tested on 12 fungal strains.

Additional file 7. Primer pairs suitable for the amplification of specific fungal phyla/subphyla. Characteristics of the primer pair and sequence coverage rate of the target group is indicated.

Additional file 8. List of the designed annealing blocking oligonucleotides for the eukaryotic groups Stramenopiles, Alveolata, Rhizaria and *Telonema*. Characteristics and sequence coverage rates of fungal and non-fungal eukaryotic groups are given.

Additional file 11. Taxonomic composition of three environmental samples. Barchart indicates relative sequence abundance of the different fungal classes/subgroups amplified by the primer pair nu-SSU-1334-5'/nu-SSU-1648-3' (FF390/FR-1). Others: Blastocladiomyetes, Glomeromycetes, Monoblepharidomycetes, Pucciniomycotina_Incertae sedis.

Author details

¹Molecular Ecology, Institute of Ecology, FB02, University of Bremen, Leobener Str. 2, 28359 Bremen, Germany. ²Department of Soil Ecology, Helmholtz Centre for Environmental Research GmbH – UFZ, Halle-Saale, Germany. ³Department of Ecology, University of Kaiserslautern, Kaiserslautern, Germany. ⁴Microbial Genomics and Bioinformatics Research Group, Max Planck Institute for Marine Microbiology, Bremen, Germany. ⁵Present Address: Department of Community Ecology, Helmholtz Centre for Environmental Research GmbH – UFZ, Halle-Saale, Germany. ⁶Department of Life Sciences and Chemistry, Jacobs University Bremen gGmbH, Bremen, Germany. ⁷German Centre for Integrative Biodiversity Research (iDiv) Halle-Jena-Leipzig, Leipzig, Germany.



^{*} Correspondence: reich@uni-bremen.de

Banos et al. BMC Microbiology (2019) 19:249 Page 2 of 2

Table 1 Characteristics and *in silico* performance of the best primer pairs. Primer pairs were grouped according to the expected amplicon size into three groups: S for small (≤600 bp), M for medium (600 – 1,000 bp), and L for large size (>1,000 bp). Fungal and non-fungal eukaryotic sequence coverage rates tested by *in silico* PCR. Individual primer sequence and characteristics are listed in the Additional file 1. For primer pairs see Additional file 2

Primer pair	Old name	name Amplicon Var. regions covered (nt)		Fungi (%)	Co-Amplif. (%)	
				(0M/1M)	(0M/1M)	
Group S						
nu-SSU-1334-5'/nu-SSU-1648-3'	FF390/FR-1	348	V7, V8	80.4/92.7	0.2/5.0	
nu-SSU-1430-5'/nu-SSU-1648-3'	SR14R/FR-1	235	V8	76.8/86.0	0.8/2.5	
nu-SSU-0062-5'/nu-SSU-0531-3'	TW9/GEO2	503	V1, V2, V3	73.7/89.1	1.5/8.0	
Group M						
nu-SSU-0817-5'-24/nu-SSU-1648-3'	nu-SSU-0817-5 ' /FR-1	870	part of V4, V5, V6, V7, V8	75.8/86.2	0.5/4.5	
nu-SSU-0777-5'/nu-SSU-1648-3'	Basid 3/FR-1	904	part of V4, V5, V6, V7, V8	68.3/80.8	2.9/14.7	
Group L						
nu-SSU-0068-5'-20/nu-SSU-1648-3'	Fun18S1/FR-1	1615	all except V9	82.3/90.3	2.3/6.8	
nu-SSU-0550-5'/nu-SSU-1648-3'	GEO3/FR-1	1133	V4, V5, V7, V8	73.1/88.4	0.9/2.0	

Amplicon (nt), length of generated amplicon

Fungi, coverage rate of fungal sequences with zero (0M) and one (1M) mismatch

Co-Amplif., non-fungal eukaryotic co-amplification

Table 2 Characteristics of the best blocking oligonucleotides complementing the primer pair nu-SSU-1334-5'/nu-SSU-1648-3' (FF390/FR-1). Fungal and non-fungal eukaryotic sequence coverage rate tested by *in silico* analysis

Target	Sequence	ComPrim	#nt	T _m (°C)	Fungi (%)	Alv. (%)	Rhiz. (%)	Stram. (%)	Tel. (%)
Alveolata	gtcgctcctaccgattga	nu-SSU-1648-3'	12	50.3	0.08	52.6	6.3	0.9	3.3
Rhizaria	ttaacgaacgagacctcga	nu-SSU-1334-5′	16	48.9	0	0	24.3	0.3	0
Stramenopiles	tcgcacctaccgattgaa	nu-SSU-1648-3′	13	48.3	0	0.5	0.3	77.1	1.7
Telonema	gaccttaacctactaaatagtta	nu-SSU-1334-5′	5	48.1	0	0.3	0	0	39.2

ComPrim, sequence complement to the indicated primer.

#nt, number of identical nt's shared by primer and blocking oligo sequence

T_m, annealing temperature

%Fungi, coverage rate for fungal sequences

%Alv., coverage rate for Alveolata sequences

%Rhiz., coverage rate for Rhizaria sequences

%Stram., coverage rate for Stramenopiles sequences

%Tel., coverage rate for Telonema sequences



References

- Banos, et al. A comprehensive fungi-specific 18S rRNA gene sequence primer toolkit suited for diverse research issues and sequencing platforms. BMC Microbiol. 2018;18:190. https://doi.org/10.1186/s12866-018-1331-4.
- Gargas A, DePriest PT. A nomenclature for fungal PCR primers with examples from intron-containing SSU rDNA. Mycologia. 1996;88(5):745–8. https://doi.org/10.2307/3760969.