

Improved 18S rDNA amplification protocol for assessing protist diversity in oxygen-deficient marine systems

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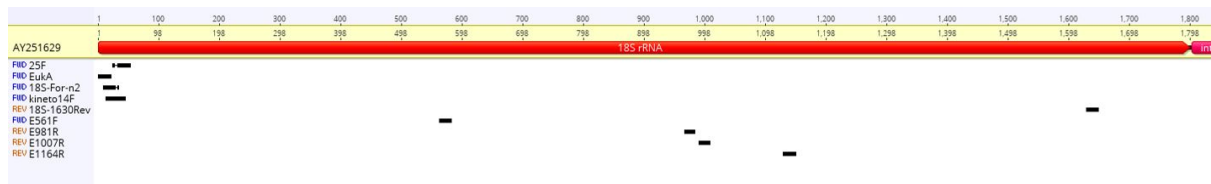


Fig. S1. Primer binding sites of all used primers for eukaryotic pre-amplification and V4 or V4/V5 amplification shown with respect to *Saccharomyces cerevisiae* accession number AY251629. Sequences of these primers are shown in Table 1.

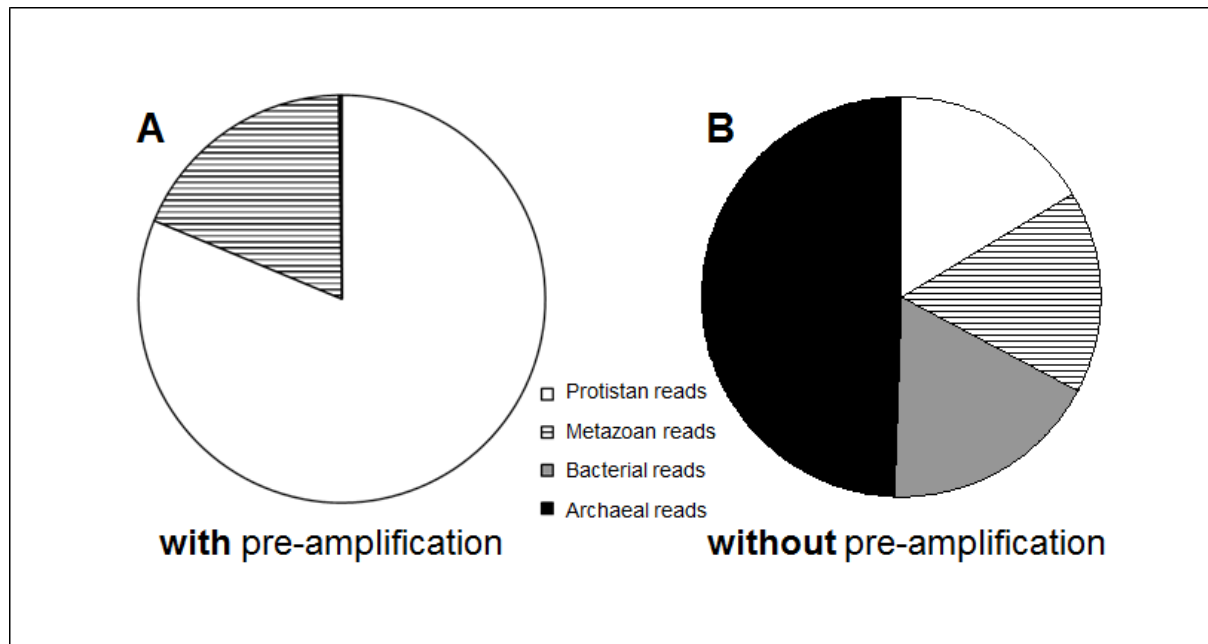


Fig. S2. Distribution of reads among the different prokaryotic and eukaryotic groups as detected in preamplified (A) and not pre-amplified (B) amplicons.

Table S1. Relative proportion of protists on the sample filter (cellular level) and in cDNA and DNA libraries (amplicon level) after a denoising procedure (Acacia).

Taxon	Cells on the filter (%)	cDNA reads (%)	DNA reads (%)
<i>Heterocapsa</i>	2.0	0.06	10.39
<i>Spumella</i>	19.6	-	10.17
<i>Cafeteria</i>	19.6	0.11	0.54
<i>Codosiga</i>	19.6	0.22	0.20
<i>Cyclidium</i>	19.6	99.61	78.69
<i>Bodo</i>	19.6	-	0.01

Table S2. Read abundances of the two Black Sea samples taken from oxygen-deficient waters (suboxic, 130 m; sulfidic, 155 m), resulting in 106 OTUs. Clone library sequences (57 OTUs; cut-off 98%; Wylezich and Jürgens, 2011) and pyrosequencing reads (92 OTUs; cut-off 95%; this study) are compared.

	130_clone_lib_cDNA	130_454-cDNA	130_454-DNA	155_clone_lib_cDNA	155_454-cDNA	155_454-DNA
1				15	174	5
2				16		1
3				1		
4				4	446	21
67					57	
5	1	1	1			
6				15	112	10
7	25	21				
8	515	61	30	64		
69						19
9	2					
10	8	1				
11	1	5				
12	1			1		
13	19	148	5	1		
14				8	29	1
15				7	155	19
16				2	203	
68					27	
17	3					
18	52	100		67		
19	1					
20	3	1040	9			
70			2			
22	1	1				

	130_clone_lib_cDNA	130_454-cDNA	130_454-DNA	155_clone_lib_cDNA	155_454-cDNA	155_454-DNA
25	2					
94		1	18			
92		40	38			
21	9	524	610			7
93		1	27			
23	21	6826	11250			7
24	1	650	1093			
95		41			3	
26		32		1	5	
27	5	1057		1		
96					4	
97						4
98						6
99			2			93
100			2			
101			5			
102			2			
103			24			
104			2			
105		30				
106		12				
83			22			37
28	4	63				
29	23	87	22	5	2	
30	1				13	5
31	2					
32				1	278	
33				5	486	
37				1		
75					4	
76					4	
77					15	
78		5				
71					8	9
34	1					
47	1				4	10
81					2	
82		30				
35				8	4	
36				1		
38				2	5	

	130_clone_lib_cDNA	130_454-cDNA	130_454-DNA	155_clone_lib_cDNA	155_454-cDNA	155_454-DNA
39				2		
40	2	70		13	48	119
41	4	9		4		
42				1		
43	13	4		31		
44	4	4	6	3	169	143
45	5		1	4	6	
46	2	22	111		13	
73		34	2			
79		1				
72		90	1			
74					735	
80						1
48	1					
49				1		
91		3				
50			36	1	210	856
51				1	40	
52				3	28	16
58					25	116
59						1
60						1
61		17	45			7
62					1	
63					221	30
64					5	
65		1	4			1
55				1		
54				2	1	
66					393	360
53	1			80	2686	1
84						8
85					9	45
86					1	
87		41				
88		42				
89		8				
90		19				
56				6	7	
57				1	6	2