

Supplementary material

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The use of extracellular DNA as a proxy for specific microbial activity

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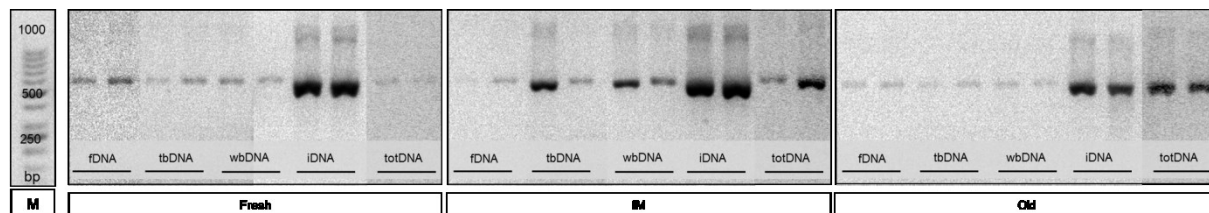
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Fig. S1 Gel electrophoreses images of the purified DNA for two replicates of the various DNA fractions for the fresh, intermediate and old fungal cultures. M = marker, IM = intermediate, ladder = ThermoFisher 50bp GeneRuler. fDNA= free external DNA, wbDNA= weakly bound external DNA, tbDNA= tightly bound external DNA, iDNA=internal DNA, totDNA= total DNA obtained by classical extraction approach, without previous elimination of exDNA.



Tab. S1 qPCR conditions and primers used in this study. FW = fresh weight

	Total Bacteria	Total Methanogens	Anaerobic fungi (<i>Neocallimastigomycota</i>)
Target	Bacterial 16S rRNA gene	Universal <i>mcrA</i> gene	28S rRNA gene
Forward primer	1055f	mlas-mod-F	GGNL1F
Reverse primer	1392r	<i>mcrA</i> -rev	GGNL4R
Forward primer sequence (5'-3')	ATGGCTGTCGTCAGCT	GGYGGTGTMGDDTTCA CMCARTA	CATAGAGGGTGAGAA TCCCGTA
Reverse primer sequence (3'-5')	ACGGGCGGTGTGTAC	CGTTCATBGCCTAGTT VGGRTAGT	TCAACATCCTAAGCG TAGGTA
Amplicon size (bp)	352	469	570
Pure culture for standards	<i>Nitrosomonas europaea</i> (DSMZ 21879)	<i>Methanosarcina barkeri</i> (DSMZ 800)	<i>Caecomycetes communis</i> (SF011204) *
Range of standards	10 ⁷ -10 ²	10 ⁵ -10 ¹	10 ⁵ -10 ¹
Detection limit (gene copies g⁻¹ FW^a)	1.6 x10 ⁵	1.6 x10 ³	1.6 x10 ³
Primer conc. (μM)	0.8	1	0.6
R² of standard curve	≥0,999	≥0,998	≥0,999
Reference	Ferris et al. 1996	Angel et al. 2012	This study
Initial denaturation	95 °C, 10 min	95 °C, 10 min	95 °C, 10 min
Nr. of cycles	40	45	45
Denaturation	95°C, 20 s	95°C, 30 s	95°C, 25 s
Annealing	58°C, 15 s	66°C, 30 s	58°C, 20 s
Elongation	72°C, 30 s	72°C, 30 s	72°C, 30 s

* WDCM collection number 919

^aFresh weight