

Table S1. Strains used for mock community, concentration of isolated DNA, and percentage contribution of used DNA.

No	Taxa	Strain information	Strain ID	DNA concentration, ng/μl	% of total DNA in MIX
1	<i>Phormidium autumnale</i> (<i>Microcoleus autumnalis</i>)	benthic, isolated from microbial mat (Eastern Pamir)	L01	142.5	23.9
2	<i>Phormidium autumnale</i> (<i>Microcoleus autumnalis</i>)	benthic, isolated from microbial mat (Eastern Pamir)	L02	21.2	3.6
3	<i>Phormidium</i> sp.	benthic, isolated from microbial mat (Eastern Pamir)	L03	53.1	8.9
4	<i>Geitlerinema ionicum</i>	benthic, isolated from microbial mat (Eastern Pamir)	L04	16.5	2.8
5	<i>Geitlerinema ionicum</i>	benthic, isolated from microbial mat (Eastern Pamir)	L05	50.5	8.5
6	<i>Oscillatoria</i> sp.	benthic, isolated from microbial mat (Eastern Pamir)	L06	52.6	8.8
7	<i>Nostoc</i> sp.	benthic, isolated from microbial mat (Eastern Pamir)	L07	13.4	2.3
8	<i>Hillbrichtia pamiria</i> genus <i>novum species novum</i>	benthic, isolated from microbial mat from the hot spring (Eastern Pamir)	L08	56.5	9.5
9	<i>Nostoc paludosum</i>	benthic, isolated from microbial mat (Eastern Pamir)	L09	18.8	3.2
10	<i>Anabaena</i> sp.	benthic, isolated from microbial mat (Eastern Pamir, Tajikistan)	L10	17.3	2.9
11	<i>Nostoc edaphicum</i>	benthic, isolated from microbial mat (Eastern Pamir, Tajikistan)	L11	12.1	2.0
12	<i>Calothrix</i> sp.	lithobiontic, isolated from calcite (Eastern Pamir, Tajikistan)	L12	79.7	13.4
13	<i>Synechocystis</i> sp.	planktonic (Baltic Sea)	L13	15.6	2.6
14	<i>Microcystis aeruginosa</i>	SAG 14.85	L14	10.25	1.7
15	<i>Synechococcus</i> sp.	planktonic (lake, Poland)	L15	35.4	6.0

Table S2. The reads obtained for studied samples before and after filtering.

Sample-id	Input reads	Filtered reads	Percentage of input passed filter
Cyx15a_V3-V4	74752	63107	84.4
<i>Cyx15a_V3-V4</i>	116810	102931	88.2
Cyx15a_V6	116713	105513	90.4
Cyx8_V3-V4	71955	64286	89.3
<i>Cyx8_V4-V6</i>	97293	84287	86.6
Cyx8_V6	91616	83944	91.6
Cyx9_V3-V4	97344	85560	87.9
<i>Cyx9_V4-V6</i>	99722	87687	87.9
Cyx9_V6	93486	84433	90.3
Mix_V3-V4	74943	64641	86.3
<i>Mix_V4-V6</i>	117286	103375	88.1
Mix_V6	104716	96093	91.8

Table S3. The number of bacterial and cyanobacterial ASVs with the percentage contribution of cyanobacterial ASVs using studied primer pairs.

	ASV bacteria	ASV cyanobacteria	Cyanobacteria % of total ASV
Cyx15a V3-V4	8919	667	7%
<i>Cyx15a V4-V6</i>	68024	66917	98%
Cyx15a V6	75844	73340	97%
Cyx8 V3-V4	10631	6306	59%
<i>Cyx8 V4-V6</i>	83276	81677	98%
Cyx8 V6	84149	83128	99%
Cyx9 V3-V4	20572	855	4%
<i>Cyx9 V4-V6</i>	62777	60711	97%
Cyx9 V6	70235	67951	97%
Mix V3-V4	9391	5996	64%
<i>Mix V4-V6</i>	47514	46473	98%
Mix V6	72762	71460	98%

Table S4. The results of the amplicon sequencing of the mock community (MIX). The taxa names of the ASVs were verified based on the Cydrasill package with our matched 16S sequences from the mock community. The interactive tree is available under the link iTOL, https://itol.embl.de/personal_page.cgi login: CyanoMIX, password: CyanoMIX2021

	Strain ID	V3-V4-MIX	V4-V6-MIX	V6-MIX
Calothrix_PCC6303 L12 Calothrix sp. (Calothrix klad 0.999999)	L12	338	8122	0
Cyanobacteriaceae L08	L08	8	0	0
Cyanobacteriales L12 Calothrix sp. (Calothrix klad 0.999911)	L12	0	0	15014
Cyanobacteriales		16	35	3448
Cyanobacteriales L08	L08	457	0	0
Cyanobacteriia		964	0	2205
Cyanobacteriia MG652616 <i>Oculatella kazantipica</i> KZ-19-s-2		0	0	261
Cyanobacteriia L08 0.999869	L08	59	41	53
Cyanobacteriia_Chloroplast		66	19	0
Cyanobium_PCC6307	L15	70	760	0
Geitlerinema_L0D9 Geitlerinema clad (Geitlerinema 0.9592812992)	L04	0	652	0
<i>Geitlerinema_L0D9_uncultured_Antarctic</i> Y423710 <i>Geitlerinema carotinosum</i> AICB 37	L05	218	4344	0
<i>Geitlerinema_uncultured_Antarctic</i>	L04, L05	2	0	0
Leptolyngbya_VRUC_135		0	209	0
<i>Leptolyngbya_VRUC_135_Oculatella mojavensis</i> MG652616 <i>Oculatella kazantipica</i> KZ-19-s-2		0	204	0
Nodosilinea_PCC7104		0	85	77
<i>Nodosilinea_PCC7104 Leptolyngbya</i> sp KIOST-1 0.987916		0	0	62
Nodosilinea_PCC7104 <i>Nodosilinea nodulosa</i> PCC 7104 <i>Leptolyngbya</i> sp RG <i>Leptolyngbya</i> PCC7104 (0.995741)		24	0	0
<i>Nodularia_PCC9350</i>	L10	46	0	0
<i>Nodularia_PCC9350</i> L10 <i>Nodularia</i> 1 (<i>Nodularia spumigena</i> CCY9414)	L10	56	0	0
<i>Nostoc_PCC73102</i>	L7	95	730	36472

Nostocaceae L10	L10	0	9162	3508
Oxyphotobacteria_Incertae_Sedis_Unknown_Famil		52	0	9
Phormidiaceae	L1, L2	0	13139	0
Sedis_Unknown_Family_uncultured_uncultured_organism		39	0	0
<i>Synechococcus_nidulans</i>	L15	14	0	0
<i>Synechocystis_PCC6803_uncultured_bacterium</i> L13 (<i>Aphanocapsa cf rivularis</i> UAM 390 JQ070058 0.3343881702)	L13	0	391	1139
<i>Trichocoleus_sociatus</i> Geitlerinema sp. PCC 7105/ <i>Planktothrix agardhii</i> NIVA-CYA 98 G <i>Planktothrix rubescens</i> NIVA-CYA 98/ <i>Planktothrix agardhii</i> NIVA-CYA 34 G <i>Planktothrix agardhii</i> NIVA-CYA 34/ <i>Planktothrix agardhii</i> NIVA-CYA 15 G <i>Planktothrix agardhii</i> NIVA-CYA 15 0.986649	L06	79	0	0
<i>Tychonema_CCAP_145911B</i>	L03	7704	21596	0
<i>Tychonema_CCAP_145911B</i> L03 <i>Oscillatoria princeps</i> PCC 10802/Uncultured cyanobacterium	L03	0	0	9719
other bacteria		3559	0	653
Sum		13866	59489	72620

Nostocaceae	0	0	0	0	0	0	0	489	579
Calothrix_PCC-6303	0	0	0	0	0	0	0	0	0
Calothrix_PCC-6303__Calothrix_sp,	0	0	0	0	0	0	0	0	0
Calothrix_PCC-6303__uncultured_bacterium	0	0	0	0	0	0	0	0	0
Nodularia_PCC-9350	0	0	0	0	0	0	0	0	0
Nostoc_PCC-73102	0	0	0	0	0	0	94	563	506
Rivularia_PCC-7116;s__uncultured_cyanobacterium	0	0	0	0	0	0	34	338	0
Chlorogloea_microcystoides	0	0	0	0	0	0	33	0	0
Oscillatoria_PCC-10802;s__uncultured_bacterium	0	18	0	0	0	0	0	0	0
Phormidiaceae	0	0	0	0	0	0	0	121	30
Lyngbya_PCC-7419	0	0	0	0	0	0	0	22	0
Trichocoleus_sociatus	0	0	0	0	0	0	0	0	0
Tychonema_CCAP_1459-11B	0	0	0	0	0	0	0	612	0
Pleurocapsa_PCC-7319	0	0	0	0	0	0	0	78	0
Leptolyngbyaceae	0	0	0	3178	81542	0	0	50	0
Leptolyngbyaceae;g_JSC-12;s__uncultured_cyanobacterium	282	24095	0	0	0	0	0	0	0
Leptolyngbyaceae;g_LB3-76	0	0	0	2380	0	0	0	0	0

Leptolyngbyaceae;g_LB3-76	0	0	0	639	0	0	0	0	0
Leptolyngbya_sp	269	42088	51123	0	0	0	0	0	0
Oxyphotobacteria_Incertae_Sedis_Unknown_Family	0	0	0	0	0	0	12	16	86
Geitlerinema_PCC-8501	0	0	0	0	0	0	0	11	0
Leptolyngbya_ANT,L52,2	0	0	0	0	18	0	0	0	0
Pegethrix_sp	0	0	0	8	0	0	0	0	0
Leptolyngbya_ANT,L67,1	0	0	0	0	0	0	0	10	19
Leptolyngbya_RV74_uncultured_cyanobacterium	0	35	0	0	0	0	0	0	0
Leptolyngbya_VRUC_135	0	0	0	0	0	0	0	0	0
Oculatella_mojaviensis	0	0	0	0	0	0	0	0	0
Phormidium_CYN64	0	0	0	2	40	33	0	0	0
Oxyphotobacteria_Incertae_Sedis	0	0	0	0	0	0	31	0	0
Oxyphotobacteria_Incertae_Sedis	0	0	0	0	0	0	0	0	0
Nodosilinea_PCC-7104	0	0	0	0	0	0	524	57899	38957
Pseudanabaenaceae	54	0	0	0	0	0	0	0	0
uncultured_Synechococcus	0	326	0	0	0	0	0	0	0
Cyanobiaceae	0	0	0	0	10	4	0	0	0

Cyanobium_PCC-6307	0	0	0	0	0	0	0	0	0
Synechococcus_nidulans	0	0	0	0	0	0	0	0	0
Thermosynechococcus_elongatus	49	309	0	0	0	0	0	0	0
Candidatus_Melainabacteria	0	0	0	0	6	0	8	0	0
uncultured_Merismopedia	0	0	0	0	0	0	6	0	0
Gastranaerophilales;s__uncultured_bacterium	0	0	0	0	0	0	0	25	0
Vampirovibrionaceae	0	0	0	0	0	0	62	0	0
Vampirovibrionales;s__uncultured_bacterium	0	0	0	0	0	0	0	87	0
Sum	667	66917	73340	6306	81677	83128	855	60711	67951