

Supplementary material:

Tracing elevational changes in microbial life and organic carbon sources in soils of the Atacama Desert

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Table S1: Significant differences in soil properties between Aroma transect samples in dependence on elevation or depth. Differences were assessed based on Kruskal-Wallis H (χ^2) tests.

Soil characteristic	elevation		depth	
	<i>H</i>	<i>P</i>	<i>H</i>	<i>P</i>
pH	1.33	0.857	10.61	0.005
CaCO ₃	11.42	0.022	0.22	0.897
OC	12.43	0.014	0.06	0.970
TN	5.95	0.203	5.51	0.063
NO ₃ -N	10.35	0.035	1.75	0.416
$\delta^{13}\text{C}$	10.03	0.040	0.59	0.746
$\delta^{15}\text{N}$	10.43	0.034	0.78	0.677
Olsen-P	7.43	0.115	0.02	0.990
Ex-P _{tot}	6.89	0.142	0.18	0.912
Ex-S _{tot}	10.96	0.027	1.76	0.414
Ex-Fe _{tot}	9.03	0.060	0.74	0.691
Fe _o	9.71	0.046	1.83	0.401
Fe _d	6.80	0.147	5.58	0.061
Fe _o /Fe _d	8.25	0.083	0.54	0.762
Na	6.40	0.171	1.04	0.595
Mg	6.70	0.153	1.94	0.379
Si	9.63	0.047	2.94	0.230
P	6.62	0.158	0.85	0.653
CaSO ₄	11.97	0.018	1.68	0.432
Clay	7.77	0.101	1.04	0.595
Silt	9.23	0.056	1.14	0.566

Table S2: Pearson correlation coefficients between soil properties (n = 18 samples). *P*-values were Bonferroni-Holm corrected for all 20 correlations each parameter underwent to account for multiple comparisons. Significant correlations are highlighted in yellow (*P* < 0.01) and green (*P* < 0.05).

	pH	CaCO ₃	OC	δ ¹³ C	δ ¹⁵ N	Olsen-P	Ex-P _{tot}	Ex-S _{tot}	TN	NO ₃ -N	Fe _o	Fe _d	Fe _d /Fe _d	Ex-Fe _{tot}	Na	Mg	Si	P	CaSO ₄	Clay
CaCO ₃	-0.040																			
OC	0.148	-0.329																		
δ ¹³ C	0.074	-0.519	0.838																	
δ ¹⁵ N	0.294	-0.660	0.615	0.802																
Olsen-P	-0.450	-0.137	0.188	-0.026	-0.084															
Ex-P _{tot}	0.434	-0.474	0.528	0.754	0.798	-0.208														
Ex-S _{tot}	0.119	0.704	-0.296	-0.563	-0.561	0.118	-0.526													
TN	0.379	0.259	0.597	0.371	0.115	-0.087	0.262	0.316												
NO ₃ -N	0.009	0.648	-0.050	-0.307	-0.600	-0.034	-0.393	0.538	0.533											
Fe _o	0.137	-0.327	0.423	0.535	0.749	-0.050	0.641	-0.324	0.143	-0.367										
Fe _d	-0.279	-0.615	0.170	0.378	0.552	-0.054	0.148	-0.511	-0.405	-0.567	0.479									
Fe _o /Fe _d	0.363	0.115	0.332	0.279	0.381	0.006	0.580	0.044	0.490	0.042	0.706	-0.276								
Ex-Fe _{tot}	0.207	-0.763	0.229	0.580	0.782	-0.348	0.641	-0.711	-0.276	-0.766	0.529	0.724	0.008							
Na	0.002	0.465	-0.525	-0.648	-0.468	0.191	-0.589	0.832	-0.100	0.272	-0.320	-0.188	-0.214	-0.478						
Mg	0.333	0.299	-0.121	0.048	-0.060	-0.613	0.326	-0.054	0.293	0.300	-0.052	-0.484	0.325	-0.050	-0.352					
Si	-0.053	-0.722	0.292	0.617	0.672	-0.294	0.559	-0.883	-0.261	-0.694	0.396	0.656	-0.072	0.882	-0.694	-0.023				
P	0.058	-0.606	0.529	0.801	0.818	-0.191	0.846	-0.768	0.054	-0.521	0.605	0.460	0.304	0.745	-0.758	0.198	0.823			
CaSO ₄	0.214	0.575	-0.406	-0.597	-0.467	0.150	-0.453	0.930	0.100	0.390	-0.307	-0.423	-0.020	-0.580	0.921	-0.124	-0.848	-0.754		
Clay	0.078	0.335	0.199	0.080	-0.095	-0.410	0.128	-0.018	0.430	0.531	0.249	-0.176	0.395	-0.212	-0.398	0.574	-0.120	0.138	-0.207	
Silt	0.112	-0.591	0.545	0.686	0.750	-0.198	0.701	-0.583	0.128	-0.317	0.774	0.576	0.394	0.676	-0.645	0.113	0.613	0.805	-0.560	0.405

Table S3: Number of culturable cells on NB and R2A medium, and gypsum content in soils at Yungay.

Site, elevation (m a.s.l.)	Depth	NB	R2A	CaSO ₄
	cm	CFU/g soil	CFU/g soil	%
Yungay, 1020	0-1	n.d.	31± 35	2.7
	1-5	1.7x10 ⁵ ± 0.4x10 ⁵	0.4x10 ⁵ ± 0.5x10 ⁵	7.4
	5-10	0.9x10 ⁵ ± 0.6x10 ⁵	0.6x10 ⁵ ± 0.5x10 ⁵	19.1

n.d. = not detected

CFU = colony forming units

Table S4: DNA recovery upon extraction from soil.

Site	Depth cm	DNA ng g ⁻¹ soil
Ar_2720	0-1	224.5
	1-5	702.0
	5-10	226.3
Ar_2455	0-1	167.9
	1-5	309.8
	5-10	257.4
Ar_2020	0-1	61.4
	1-5	162.4
	5-10	261.2
Ar_1680	0-1	<2.0
	1-5	40.4
	5-10	5.8
Ar_1340	0-1	33.3
	1-5	6.4
	5-10	2.8
Yu_1020	0-1	3.2
	1-5	7.1
	5-10	4.6

Table S5: Pearson correlation coefficients for microbial abundance parameters in relation to soil properties (n = 18 samples). Numbers of colony-forming units (CFU) were quantified on NB and R2A medium and log-transformed for correlation. *P*-values were Bonferroni-Holm corrected due to the multiple comparisons performed for each dependent variable. Significant results (*P* < 0.05) are highlighted in bold and are illustrated in Fig. S3.

Soil characteristic	DNA recovery		CFU on NB		CFU on R2A	
	<i>R</i>	<i>P</i>	<i>R</i>	<i>P</i>	<i>R</i>	<i>P</i>
pH	0.167	1.000	0.014	1.000	0.054	1.000
CaCO ₃	-0.581	0.182	-0.467	0.5621	-0.544	0.276
OC	0.728	0.011	0.779	0.002	0.748	0.008
TN	0.195	1.000	0.350	1.000	0.247	1.000
NO ₃ -N	-0.430	0.598	-0.155	1.000	-0.363	0.973
δ ¹³ C	0.773	0.004	0.715	0.012	0.726	0.014
δ ¹⁵ N	0.766	0.004	0.519	0.413	0.626	0.103
Olsen-P	0.002	1.000	-0.081	1.000	-0.058	1.000
Ex-P _{tot}	0.469	0.596	0.350	1.000	0.401	0.794
Ex-S _{tot}	-0.465	0.596	-0.451	0.604	-0.420	0.744
Ex-Fe _{tot}	0.591	0.167	0.478	0.536	0.596	0.142
Fe _o	0.290	1.000	0.391	0.977	0.484	0.458
Fe _d	0.491	0.503	0.505	0.456	0.602	0.139
Fe _o /Fe _d	-0.055	1.000	0.048	1.000	0.063	1.000
Na	-0.460	0.560	-0.570	0.2312	-0.445	0.644
Mg	-0.119	1.000	-0.171	1.000	-0.338	1.000
Si	0.560	0.234	0.531	0.374	0.597	0.142
P	0.608	0.133	0.487	0.525	0.513	0.382
CaSO ₄	-0.446	0.596	-0.578	0.216	-0.499	0.419
Clay	-0.166	1.000	0.223	1.000	0.021	1.000
Silt	0.518	0.386	0.639	0.082	0.620	0.110

Table S6: Fitting of environmental and microbial parameters to microbial community composition as seen in an NMDS plot. Fitting was done with the Envfit function. *P*-values were Bonferroni-Holm corrected to account for multiple comparisons. Significant results ($P < 0.05$) are highlighted in bold and are illustrated in Fig. 4.

Soil characteristic	R^2	P
pH	0.15	1.000
CaCO ₃	0.52	0.252
OC	0.76	<0.022
TN	0.19	1.000
NO ₃ -N	0.31	0.672
δ¹³C	0.70	<0.022
δ¹⁵N	0.69	0.022
Olsen-P	0.18	1.000
Ex-P _{tot}	0.31	0.749
Ex-S _{tot}	0.43	0.473
Ex-Fe_{tot}	0.72	0.022
Fe _o	0.39	0.500
Fe _d	0.38	0.504
Fe _o /Fe _d	0.05	1.000
Na	0.43	0.442
Mg	0.17	1.000
Si	0.50	0.252
P	0.49	0.252
CaSO ₄	0.45	0.442
Clay	0.01	1.000
Silt	0.50	0.24
CFU NB	0.60	0.247
CFU R2A	0.52	0.434
DNA recovery	0.57	0.255

CFU = colony forming units

Table S7: Archaeal community composition in samples along the Aroma transect. Relative abundance of the taxa is given as well as total read number for archaea and bacteria per sample.

Phylum	Class	Order	Ar_1340			Ar_1680			Ar_2020			Ar_2455			Ar_2720			Negative controls ¹		
			0-1	1-5	5-10	0-1	1-5	5-10	0-1	1-5	5-10	0-1	1-5	5-10	0-1	1-5	5-10	Extr. 1	Extr. 2	PCR
Archaea			0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
<i>Crenarchaeota</i>	MBGB		0.0	0.0	4.0	0.0	0.0	0.0	<0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
<i>Crenarchaeota</i>	MCG		0.0	0.0	0.0	0.0	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
<i>Crenarchaeota</i>	MCG	GrfC26	0.0	0.0	0.0	0.0	0.0	0.0	4.1	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	12.3	23.5	0.0
<i>Crenarchaeota</i>	<i>Thaumarchaeota</i>	<i>Nitrososphaerales</i>	100.0	100.0	50.0	99.7	100.0	99.8	64.2	100.0	99.7	100.0	100.0	99.9	99.5	99.7	99.6	0.0	16.6	100.0
<i>Crenarchaeota</i>	<i>Thermoprotei</i>		0.0	0.0	0.0	0.0	0.0	0.0	0.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
<i>Euryarchaeota</i>	DSEG	104A5	0.0	0.0	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
<i>Euryarchaeota</i>	<i>Methanobacteria</i>	<i>Methanobacteriales</i>	0.0	0.0	0.0	0.0	0.0	0.0	1.2	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	20.4	6.7	0.0
<i>Euryarchaeota</i>	<i>Methanomicrobia</i>		0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.4	0.0
<i>Euryarchaeota</i>	<i>Methanomicrobia</i>	<i>Methanosarcinales</i>	0.0	0.0	0.0	0.0	0.0	0.2	1.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	11.4	0.0
<i>Euryarchaeota</i>	<i>Thermoplasmata</i>		0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0
<i>Euryarchaeota</i>	<i>Thermoplasmata</i>	E2	0.0	0.0	46.0	0.1	0.0	0.0	27.9	0.0	0.3	0.0	0.0	0.1	0.3	0.3	0.4	67.3	39.5	0.0
Total archaeal read number			4	6	50	2,893	11,758	10,084	9,935	23,114	10,672	34,596	40,047	29,342	30,090	37,963	17,984	162	1,929	9
Total bacterial read number			178	0	0	544	1,580	943	166	2,484	3,598	567	1,559	1645	91	6,864	327	15	158	0

¹ Negative controls: Extr. 1 = extraction blank number 1; Extr. 2 = extraction blank number 2; PCR = PCR blank

Table S8: Lipid biomarker concentrations in soils from the Aroma transect.

Compound	Ar_2720	Ar_2455	Ar_2020	Ar_1680	Ar_1340
<u>saturated fatty acids (ng/g DW)</u>					
C _{12:0}	12.4	3.3	3.3	3.2	2.6
C _{14:0}	25.1	5.5	7.7	5.9	6.1
C _{15:0}	17.0	2.9	2.7	2.5	3.4
C _{16:0}	337.2	100.9	117.9	83.3	87.0
C _{17:0}	4.7	n.d.	n.d.	n.d.	n.d.
C _{18:0}	112.3	54.7	61.6	33.3	39.7
C _{20:0}	24.0	3.0	2.3	0.8	1.1
C _{21:0}	7.0	n.d.	n.d.	n.d.	n.d.
C _{22:0}	152.2	17.2	7.3	1.4	0.7
C _{23:0}	64.9	4.5	n.d.	n.d.	n.d.
C _{24:0}	305.7	16.9	7.8	n.d.	n.d.
C _{25:0}	38.2	n.d.	n.d.	n.d.	n.d.
C _{26:0}	123.7	6.3	6.9	n.d.	n.d.
C _{28:0}	40.3	n.d.	n.d.	n.d.	n.d.
<u>unsaturated fatty acids (ng/g DW)</u>					
C _{16:1}	146.5	4.5	6.6	4.1	6.3
C _{18:1}	79.4	18.6	17.7	9.3	5.0
C _{18:2}	36.0	n.d.	n.d.	n.d.	n.d.
C _{20:1}	12.7	3.4	3.7	2.3	0.7
C _{22:1}	287.4	78.2	95.0	67.9	9.0
total fatty acids (ng/g DW)	1826.7	320.0	340.4	214.0	151.6
<u>saturated <i>n</i>-alkanes (ng/g DW)</u>					
C _{21:0}	5.3	n.d.	n.d.	n.d.	n.d.
C _{22:0}	5.3	n.d.	n.d.	n.d.	n.d.
C _{23:0}	4.4	4.5	n.d.	5.8	n.d.
C _{24:0}	4.4	4.8	n.d.	5.7	6.0
C _{25:0}	10.7	9.2	6.9	6.4	5.4
C _{26:0}	3.4	n.d.	n.d.	n.d.	3.3
C _{27:0}	14.4	15.7	15.5	8.0	4.6
C _{28:0}	4.0	3.5	3.6	1.7	1.4
C _{29:0}	20.4	19.4	n.d.	n.d.	1.8
C _{30:0}	1.4	1.7	n.d.	n.d.	n.d.
C _{31:0}	13.9	6.7	n.d.	n.d.	n.d.
C _{32:0}	0.6	n.d.	n.d.	n.d.	n.d.
C _{33:0}	2.4	n.d.	n.d.	n.d.	n.d.
C _{34:0}	1.6	n.d.	n.d.	n.d.	n.d.
C _{35:0}	1.7	n.d.	n.d.	n.d.	n.d.
Total <i>n</i> -alkanes (ng/g DW)	93.9	65.4	26.0	27.7	22.6
ACL	28.4	27.9	26.4	26.1	26.4
CPI	5.4	7.5	6.2	5.1	1.8

Compound	Ar_2720	Ar_2455	Ar_2020	Ar_1680	Ar_1340
<u>isoprenoid GDGTs (pg/g DW)</u>					
GDGT-0	530.1	212.1	317.3	16.8	n.d.
GDGT-1	232.8	146.6	255.8	16.0	n.d.
GDGT-2	633.2	303.3	538.0	38.3	n.d.
GDGT-3	390.1	236.2	371.3	20.1	n.d.
GDGT-4	373.5	248.2	226.7	14.1	n.d.
crenarchaeol	2229.5	1777.7	2435.5	91.4	n.d.
crenarchaeol isomer	439.0	207.0	124.6	9.3	n.d.
<u>branched GDGTs (pg/g DW)</u>					
GDGT-IIIa	1417.6	791.8	2405.4	n.d.	n.d.
GDGT-IIIb	14.4	14.3	112.2	n.d.	n.d.
GDGT-IIIc	0.6	5.4	15.2	n.d.	n.d.
GDGT-IIa	3035.3	1434.2	2266.0	n.d.	n.d.
GDGT-IIb	122.8	84.6	792.4	n.d.	n.d.
GDGT-IIc	1.7	14.9	94.7	n.d.	n.d.
GDGT-Ia	655.5	428.0	757.5	n.d.	n.d.
GDGT-Ib	49.6	54.6	240.9	n.d.	n.d.
GDGT-Ic	5.2	6.2	35.0	n.d.	n.d.
Total GDGTs (pg/g DW)	1599.3	905.0	1691.3	30.7	n.d.

n.d.: not detected.

ACL: average chain length: $(25(C_{25:0}) + 27(C_{27:0}) + 29(C_{29:0}) + 31(C_{31:0})) / (C_{25:0} + C_{27:0} + C_{29:0} + C_{31:0})$

CPI: carbon preference index:

$0.5((C_{25:0} + C_{27:0} + C_{29:0} + C_{31:0}) / (C_{24:0} + C_{26:0} + C_{28:0} + C_{30:0})) / ((C_{25:0} + C_{27:0} + C_{29:0} + C_{31:0}) / (C_{26:0} + C_{28:0} + C_{30:0} + C_{32:0}))$

Table S9: Pearson correlation coefficients for lipid abundance in relation to selected soil properties. *P*-values were Bonferroni-Holm corrected due to the multiple comparisons performed for each dependent variable. Significant results ($P < 0.05$) are highlighted in bold ($n = 5$).

Soil characteristic	<i>n</i> -fatty acids		<i>n</i> -alkanes		PLFAs		GDGTs	
	<i>R</i>	<i>P</i>	<i>R</i>	<i>P</i>	<i>R</i>	<i>P</i>	<i>R</i>	<i>P</i>
OC	0.960	0.019	0.904	0.105	0.616	0.806	0.767	0.311
δ ¹³ C	0.936	0.019	0.837	0.107	0.520	0.806	0.800	0.311
δ ¹⁵ N	0.976	0.013	0.873	0.107	0.512	0.806	0.617	0.311



Figure S1: Photographs taken at the different study sites. Sparse vegetation is seen at Ar_2720. Stones covering the surface were removed before sampling, as seen in the photograph of Ar_2455.

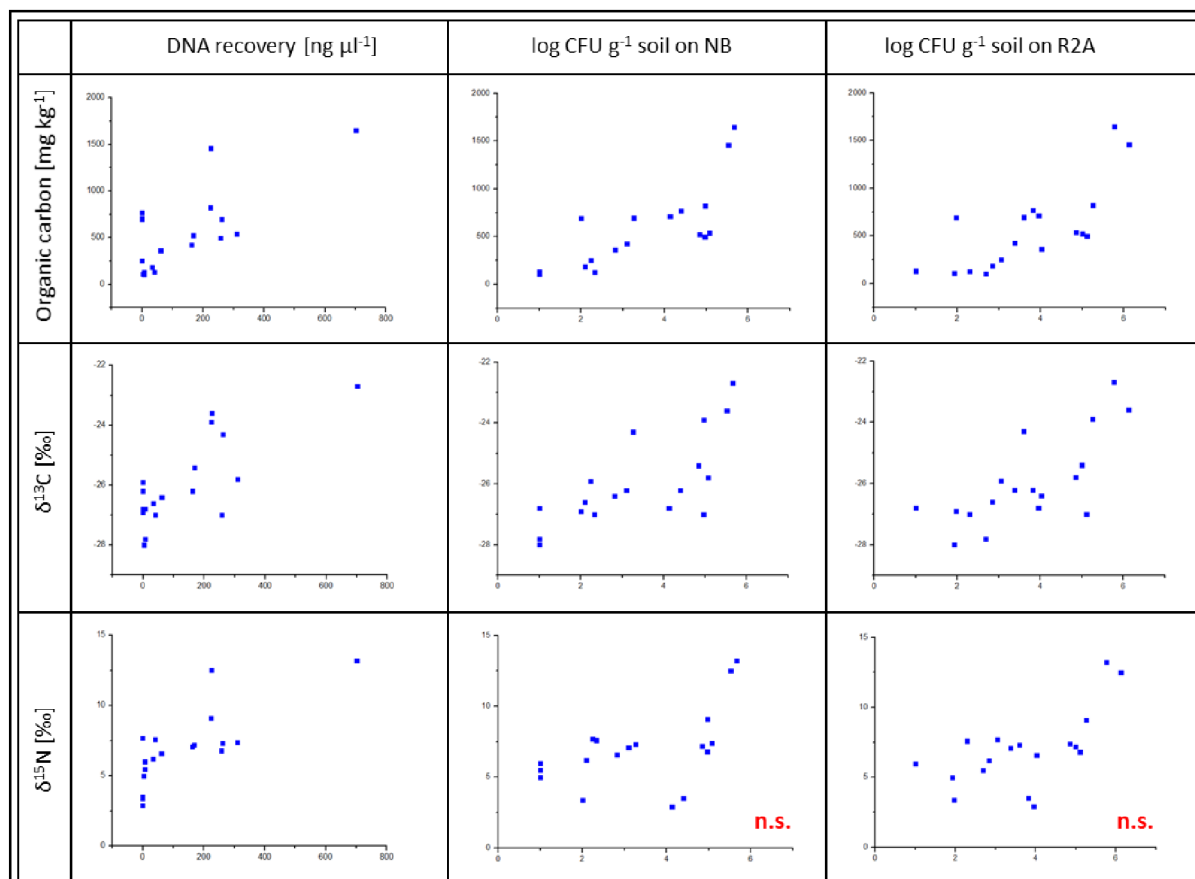


Figure S2: Plots displaying significant correlations between microbial abundance parameters and soil properties. Correlations were analyzed according to Table S3.

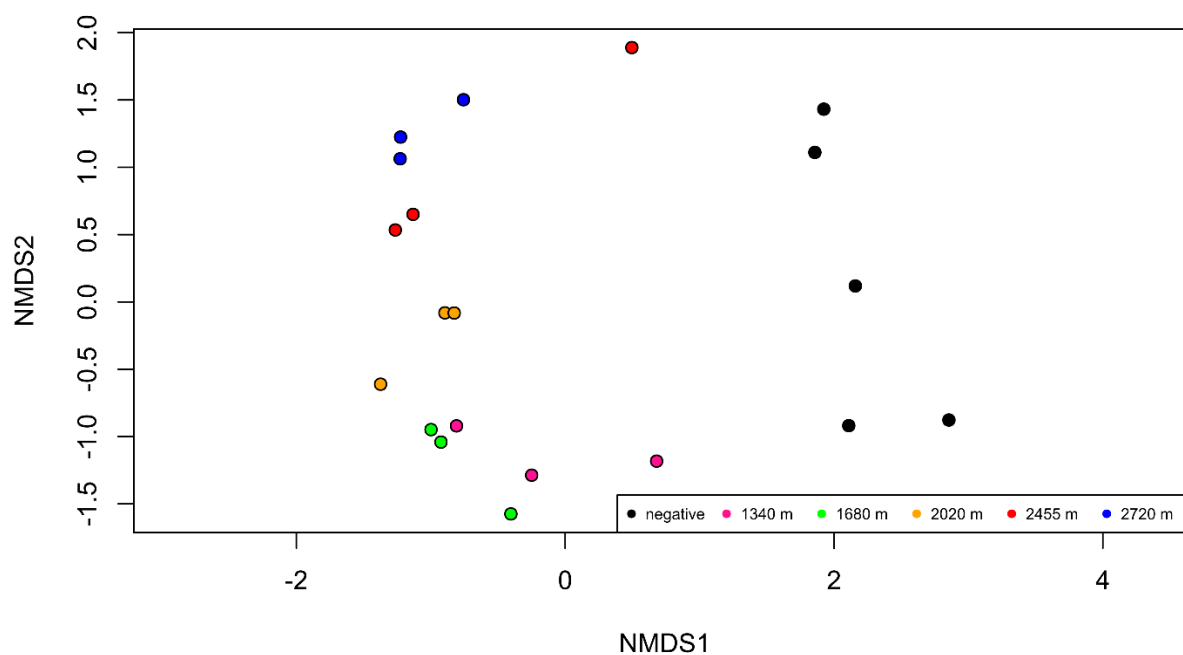


Figure S3: NMDS plot showing the dissimilarities in bacterial community composition between samples taken along the Aroma transect and negative controls included during DNA extraction and PCR. A clear separation of data from samples and controls is evident and was supported by ANOSIM ($R = 0.83$, $P = 0.001$). Based on these results, negative controls were excluded in all further analyses.

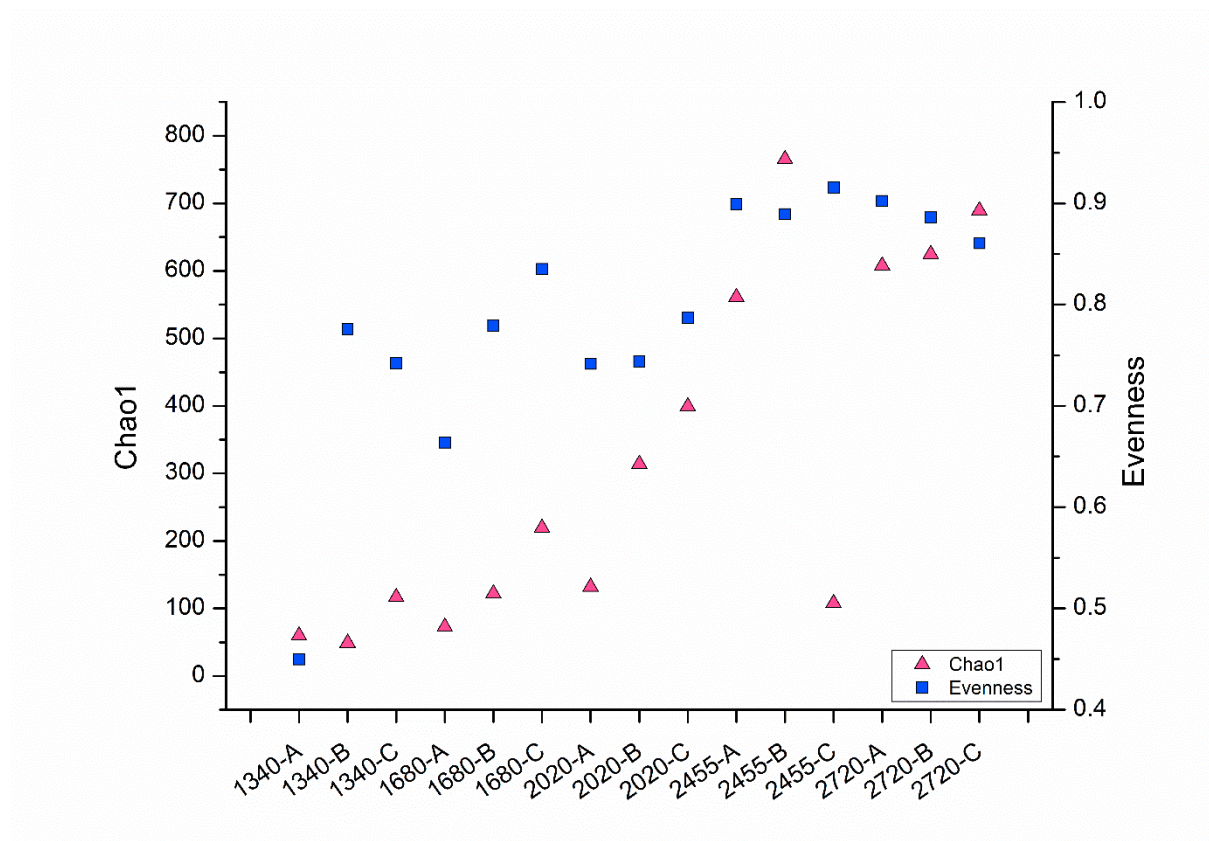


Figure S4: Bacterial diversity in the soil samples along the Aroma elevational transect. Estimated diversity based on the Chao1 index as well as Pielou's evenness index are presented. The labeling of soil samples from different depths is as follows: A = 0 – 1 cm, B = 1 – 5 cm, C = 5 – 10 cm. The indices were calculated based on a rarefied dataset of 2177 amplicon sequencing variants (ASVs). This low number allowed to include even the samples from the driest site (1340 m a.s.l.), where only weak PCR products were obtained. Rarefaction curves were made and confirmed that this number was still sufficient to estimate diversity also in samples from all other sites.

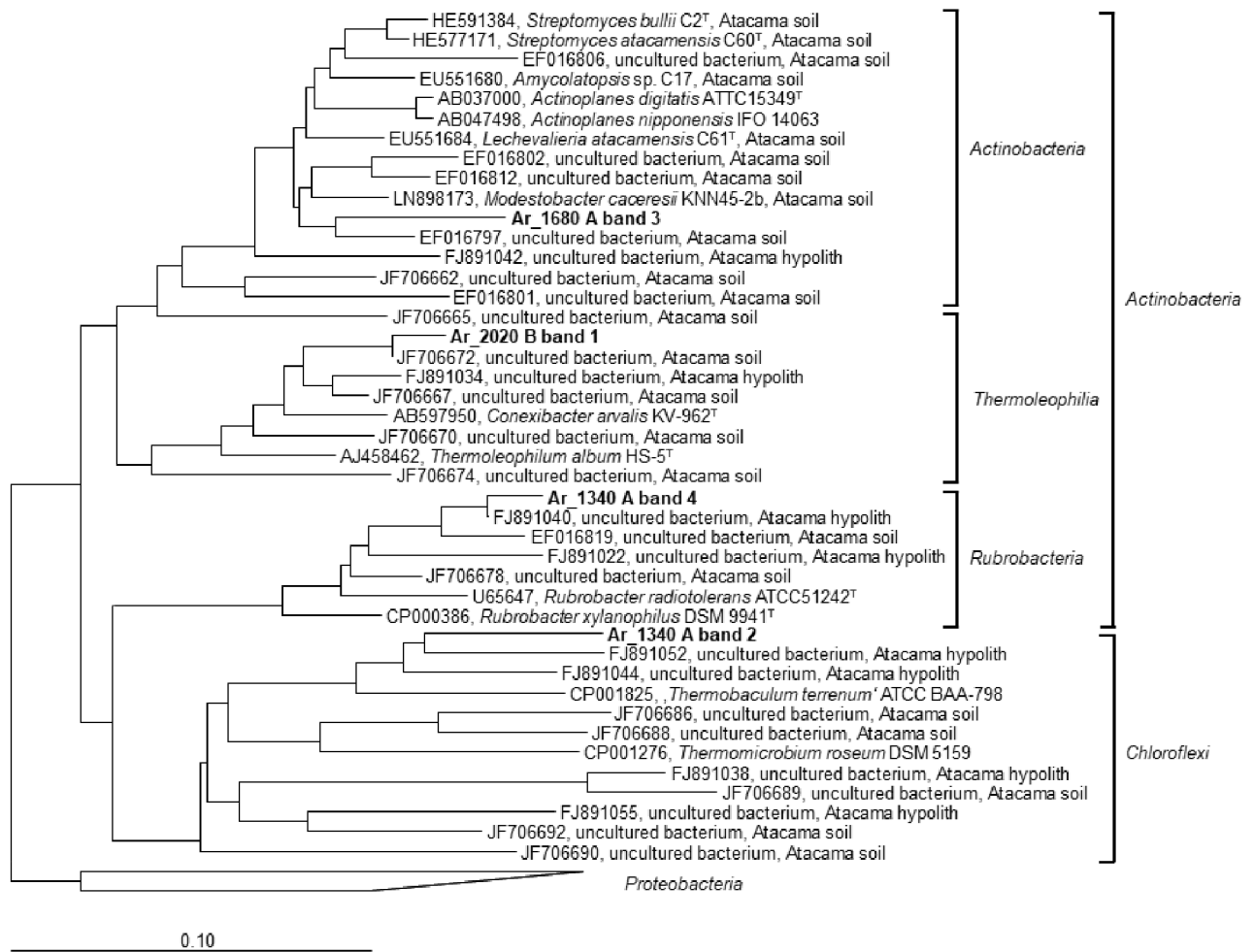


Figure S5: Identity of bacterial taxa that were derived from Sanger sequencing of dominant of a detaturing gradient gel electrophoresis gel from a pre-experiment. The phylogenetic tree was calculated based on 592 nucleotide positions using the neighbor joining algorithm with Jukes-Cantor correction. The bar denotes 10 % sequence divergence. Sequences obtained in this study are highlighted in bold. They are shown in relation to the most closely related sequences from cultivated strains and to related sequences that were derived from other Atacama soil studies. The tree was rooted using ten sequences of *Proteobacteria*. Phyla and classes are indicated by brackets. The sequences have been deposited in the DDBJ/EMBL/Genbase databases under the accession numbers MK968112 – MK968115.

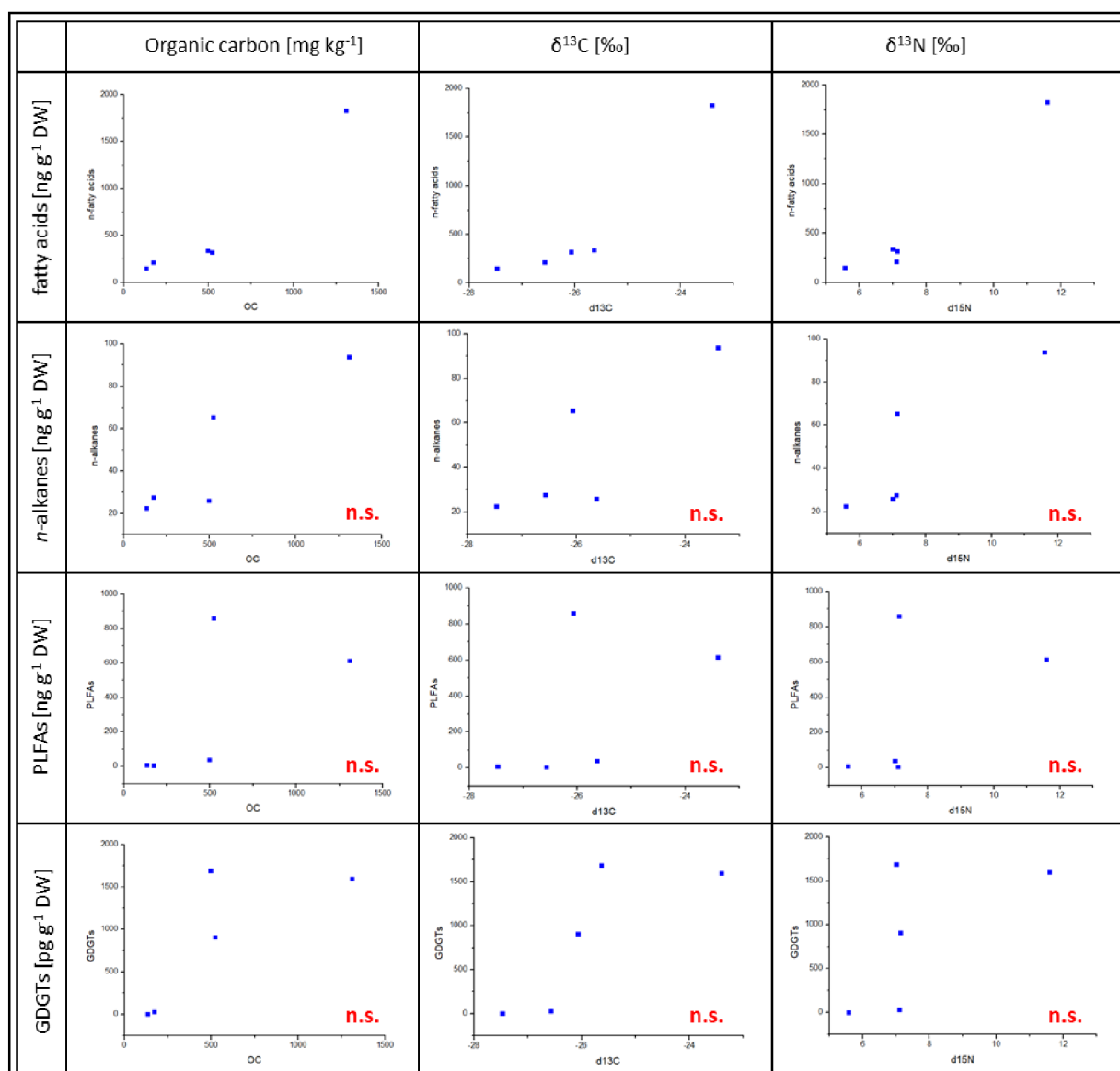


Figure S6: Plots displaying correlations between soil parameters and lipid concentrations. Correlations were analyzed according to Table S9 (n.s. = no significant correlation).