

Supplementary Information

Towards a global biogeography of the glacier-fed stream benthic microbiome

Leïla Ezzat^{1,2*}, Hannes Peter¹, Massimo Bourquin¹, Grégoire Michoud¹, Stilianos Fodelianakis¹, Tyler J. Kohler^{1,3}, Thomas Lamy², Susheel Bhanu Busi⁴, Daniele Daffonchio⁵, Nicola Deluigi¹, Vincent De Staercke¹, Ramona Marasco⁵, Paraskevi Pramateftaki¹, Martina Schön¹, Michail Styllas¹, Matteo Tolosano¹, and Tom J. Battin^{1*}

(1) River Ecosystems Laboratory, Alpine and Polar Environmental Research Center, Ecole Polytechnique Fédérale de Lausanne (EPFL), Sion, Switzerland

(2) MARBEC, Université de Montpellier, CNRS, Ifremer, IRD, Montpellier, France

(3) Department of Ecology, Faculty of Science, Charles University, Prague, Czechia

(4) Systems Ecology Group, Luxembourg Centre for Systems Biomedicine, University of Luxembourg, Esch-sur-Alzette, Luxembourg

(5) Marine Science Program, Biological and Environmental Science and Engineering Division, King Abdullah University of Science and Technology (KAUST), Thuwal 23955-6900, Kingdom of Saudi Arabia

(6) Red Sea Research Center, King Abdullah University of Science and Technology (KAUST), Thuwal 23955-6900, Kingdom of Saudi Arabia

Supplementary Table 1. Relative abundance and number of shared ASVs between pairs of mountain ranges.

	relative abundance_A	relative abundance_B	Number of ASVs shared
Himalayas-Scandinavian Mountains	0.419015	0.5071668	2157
Himalayas-European Alps	0.782806	0.7481819	5523
Himalayas-Caucasus Mountains	0.7829652	0.7430087	5164
Himalayas-Ecuadorian Andes	0.3199009	0.3027907	1473
Himalayas-Southwest Greenland	0.3311317	0.4864572	1301
Himalayas-Southern Alps	0.4728874	0.3906133	2579
Himalayas-Pamir & Tien Shan	0.6982064	0.7229774	4012
Himalayas-Chilean Andes	0.5717394	0.4467462	2509
Himalayas-Alaska Range	0.6443309	0.7448201	3040
Scandinavian Mountains-European Alps	0.8411826	0.630231	3676
Scandinavian Mountains-Caucasus Mountains	0.7885709	0.6201421	3370
Scandinavian Mountains-Ecuadorian Andes	0.4577847	0.4519469	1886
Scandinavian Mountains-Southwest Greenland	0.7808465	0.9018629	2511
Scandinavian Mountains-Southern Alps	0.4376911	0.305656	1803
Scandinavian Mountains-Pamir & Tien Shan	0.3357528	0.3451913	1290

Scandinavian Mountains-Chilean Andes	0.405467	0.374804	1370
Scandinavian Mountains-Alaska Range	0.5742785	0.5623666	1707
European Alps-Caucasus Mountains	0.8870318	0.8793335	7074
European Alps-Ecuadorian Andes	0.4028483	0.4862575	2169
European Alps-Southwest Greenland	0.5417904	0.8765586	2489
European Alps-Southern Alps	0.4734824	0.4019203	2870
European Alps-Pamir & Tien Shan	0.6530452	0.6912551	3932
European Alps-Chilean Andes	0.5429602	0.5074065	2611
European Alps-Alaska Range	0.7485469	0.8325259	3968
Caucasus Mountains-Ecuadorian Andes	0.414518	0.4321697	2015
Caucasus Mountains-Southwest Greenland	0.5544361	0.7960096	2232
Caucasus Mountains-Southern Alps	0.4411217	0.4055561	2754
Caucasus Mountains-Pamir & Tien Shan	0.6426268	0.6909848	3635
Caucasus Mountains-Chilean Andes	0.502204	0.4827798	2418
Caucasus Mountains-Alaska Range	0.688269	0.7949225	3350
Ecuadorian Andes-Southwest Greenland	0.3793637	0.5375735	1331
Ecuadorian Andes-Southern Alps	0.4701593	0.4118664	2200

Ecuadorian Andes-Pamir & Tien Shan	0.2170282	0.3185161	1001
Ecuadorian Andes-Chilean Andes	0.4608922	0.4305349	1733
Ecuadorian Andes-Alaska Range	0.310049	0.3010038	984
Southwest Greenland-Southern Alps	0.4803476	0.236483	1074
Southwest Greenland-Pamir & Tien Shan	0.363174	0.3104636	914
Southwest Greenland-Chilean Andes	0.4584478	0.2846576	854
Southwest Greenland-Alaska Range	0.6523664	0.4655946	1214
Southern Alps-Pamir & Tien Shan	0.2820456	0.3572127	1614
Southern Alps-Chilean Andes	0.5498252	0.5957948	2822
Southern Alps-Alaska Range	0.3242696	0.419421	1680
Pamir & Tien Shan-Chilean Andes	0.4521242	0.3678969	1857
Pamir & Tien Shan-Alaska Range	0.5058447	0.6412359	2235
Chilean Andes-Alaska Range	0.4091095	0.5133663	1634

Supplementary Table 2. Results from INEXT analyses providing estimates of the regional (gamma) diversity as ASV richness for a given mountain range based on either abundance or frequency data. s.e. stands for standard error, LCL and UCL for lower and upper confidence limits respectively while “O” and “E” stand for observed and estimated respectively.

Site	Observed	Estimator (Abundance)	s.e.	LCL	UCL	O/E	E/O
European Alps	14131.00	16856.33	125.17	16621.83	17112.90	0.84	1.19
Southern Alps	10803.00	12366.38	89.59	12200.42	12552.05	0.87	1.14
Caucasus Mountains	12070.00	14768.49	128.46	14528.22	15032.23	0.82	1.22
Pamir & Tien Shan	12333.00	14088.72	92.43	13916.71	14279.41	0.88	1.14
Southwest Greenland	4034.00	4607.47	52.817	4512.937	4720.66	0.88	1.14
Scandinavian Mountains	7765.00	9471.20	100.249	9285.755	9679.25	0.82	1.22
Ecuadorian Andes	7122.00	8185.54	72.615	8052.469	8337.63	0.87	1.15
Himalayas	8516.00	9697.49	76.193	9557.341	9856.50	0.88	1.14
Alaska Range	7584.00	8851.12	80.573	8702.784	9019.12	0.86	1.17
Chilean Andes	7584.00	8851.12	80.573	8702.784	9019.12	0.86	1.17

Site	Observed	Estimator (Frequency)	s.e.	LCL	UCL	O/E	E/O
European Alps	14130.00	22289.34	272.77	21772.00	22841.71	0.63	1.58
Southern Alps	10802.00	15755.29	191.22	15394.46	16144.48	0.69	1.46
Caucasus Mountains	12069.00	19926.15	277.32	19401.14	20488.75	0.61	1.65

Pamir & Tien Shan	12332.00	20760.37	286.73	20216.87	21341.34	0.59	1.68
Southwest Greenland	4033.00	7871.90	217.448	7468.817	8322.26	0.51	1.95
Scandinavian Mountains	7764.00	15547.69	324.57	14937.11	16210.25	0.50	2.00
Ecuadorian Andes	7121.00	13113.42	256.91	12630.67	13638.47	0.54	1.84
Himalayas	8515.00	14356.44	244.21	13897.08	14855.01	0.59	1.69
Alaska Range	7072.00	14988.88	342.96	14344.73	15690.09	0.47	2.12
Chilean Andes	7583.00	16084.59	341.27	15441.59	16780.19	0.47	2.12

Supplementary Table 3. Occurrence of endemic and unique ASVs across mountain ranges

		endemic ASVs							
	n _{samples}	ASVs	unique ASVs (n / %)	endemic ASVs (n / %)	mean occurrence (n)	maximum occurrence (n)	total relative abundance (%)	average relative abundance (%)	maximum relative abundance(%)
			1546	285					
Alaska	15	7072	21.9%	4.0%	2.73	7	3.3	0.06	0.99
			2261	914					
European Alps	26	14130	16.0%	6.5%	2.74	15	2.7	0.03	0.80
			1650	563					
Caucasus Mountains	19	12069	13.7%	4.7%	2.84	15	4.4	0.05	8.47
			2055	492					
Chilean Andes	10	7583	27.1%	6.5%	2.59	7	15.3	0.09	2.42
			2012	830					
Ecuadorian Andes	9	7121	28.3%	11.7%	2.88	9	19.1	0.05	2.10
			452	65					
Southwest Greenland	6	4033	11.2%	1.6%	2.22	5	0.7	0.03	0.21
			1546	916					
Pamir & Tien Shan	17	8515	18.2%	10.8%	3.84	13	12.4	0.04	1.75
			2617	1040					
Himalayas	16	12332	21.2%	8.4%	2.64	11	6.6	0.03	1.35
			2555	2456					
Southern Alps	20	10802	23.7%	22.7%	3.68	17	27.3	0.04	2.32

		1798							
Scandinavian Mountains	9	7764	23.2%	379	2.36	6	2.8	0.03	0.29

Supplementary Note 1. Detailed results from variation partitioning

Partitioning the variance for the entire microbiome, we found that the variation in GFS microbiome was structured over broad spatial scales, as the spatial trend and the among mountain range components of variation explained 12.5% ($F=7.43$, $p=0.001$) and 16.7% ($F=4.5$, $p=0.001$) of the total variation in GFS microbiome. That said, 14.5% ($F=1.54$, $p=0.001$) of the total variation in GFS microbiome was explained by spatial differences within mountain ranges. When accounting for environmental variation we found that only a small fraction of the spatial variation was shared with environmental conditions at each scale (among and within mountain ranges: 8% and 3.8%) while the remaining pure spatial effects explained a larger fraction of the variation among and within mountain ranges (among: 8.7% and within: 10.7%). Less than 5% of the total variation in GFS microbiome (3.6%) was explained by environmental conditions not spatially structured.

Similarly, the total variation in the core microbiome was mainly driven by spatial processes, including the spatial trend (16.3%, $F=9.8$, $p=0.001$), the among and within mountain range components (among - 26.4%, $F=7.7$, $p=0.001$; within 20.8%, $F=2.2$, $p=0.001$). Yet, we found that the spatial structuring of the environment explained a larger variation among mountain ranges (among: 18.3% and within: 8.8%) than pure spatial effects (among: 8% and within: 12%). Pure environmental conditions explained a slightly greater variation 6.6% of the total variation as compared to the entire microbiome.

Partitioning the variance for the endemic ASV dataset demonstrated strong effects of spatial processes (linear trend: 6.6%, $F=1.23$, $p=0.001$; among mountain range: 11.5% $F=3$, $p=0.001$; within mountain range: 11.3%, $F=1.3$, $p=0.001$), and specifically of pure spatial processes (among: 8.8% and within: 9.6%) on GFS microbiome composition. Accordingly, only 2.7% and 1.7% of the variance was attributable to spatially structured environmental variation among and within mountain ranges, while only 1.5% of the total variation was explained by pure environmental variation.