

Findings and challenges in understanding the impact of human-induced trampling on Antarctic edaphic microbial communities and their recovery potential

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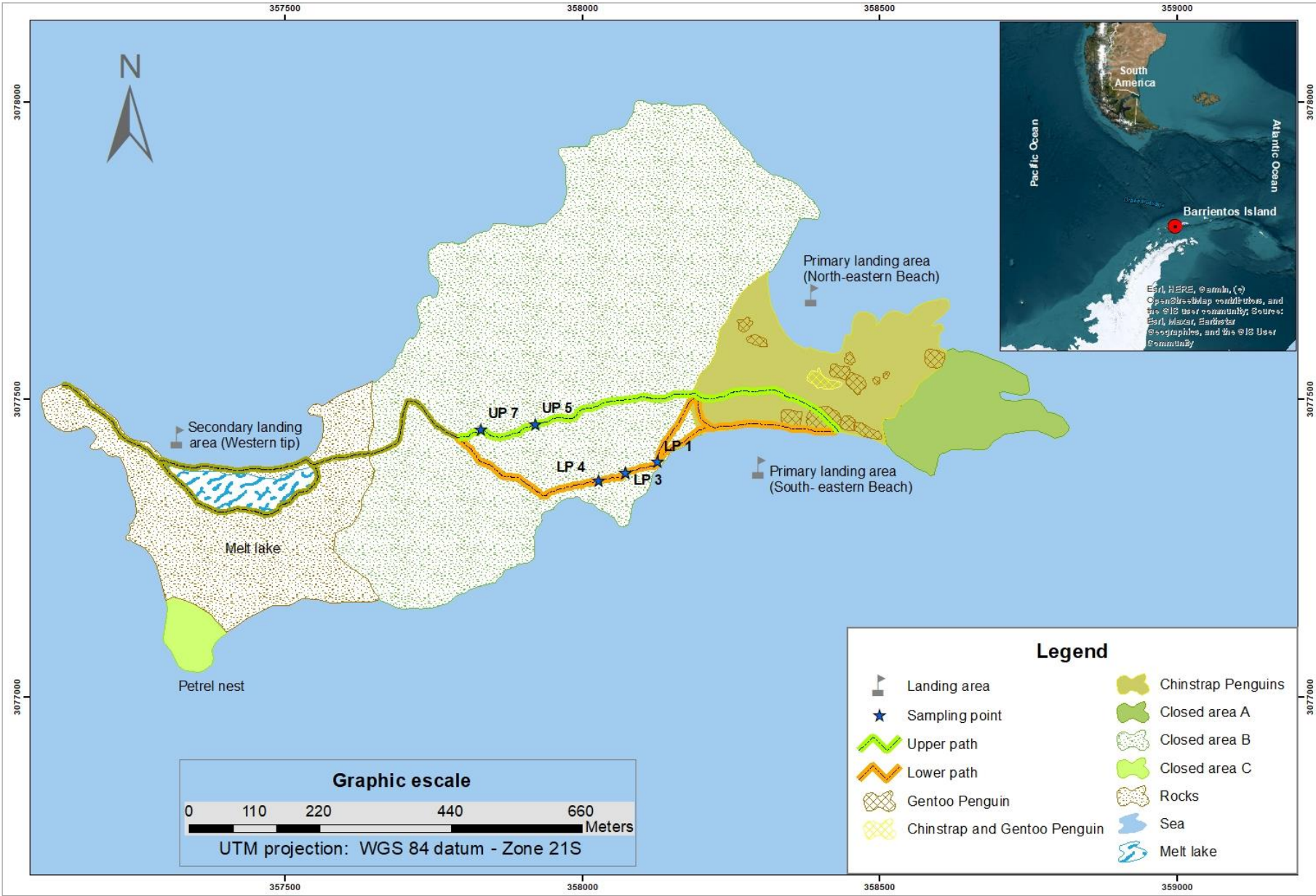
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Supporting Information

Supporting information 1: Map of Barrientos Island showing the two paths formerly established along it and the location of the sampling points. UP: Upper path; LP: Lower path



Supporting information 2: Samples information table, including physical and chemical data.

Path	Sample ID	Longitude (WGS 84)	Latitude (WGS 84)	Altitude (m.a.s.l.)	Temperature (°C)		Humidity (m ³ +m ⁻³)	Water FV*	Soil CE (S/m)**	Soil TC-EC (S/m)***	Pore water CE (S/m)****
					5cm	10cm					
Lower path (LP)	LP1U	6224436	5944901	3m	1.1	3.5	0.314	0.205	0.012	0.017	0.07912
	LP1T				1	2.4					
	LP3U	6224459	5944810	-1m	1	2.5	0.624	0.254	0.038	0.056	0.08674
	LP3T				1.2	2.3					
	LP4U	6224464	5944808	1m	1	2.2	0.658	0.306	0.049	0.073	0.1057
	LP4T				1.3	2.2					
Upper path (UP)	UP5U	6224425	5945030	12m	0.7	2.5	0.242	0.118	0.005	0.007	0.09817
	UP5T				0.6	2.5					
	UP7U	6224421	5945103	11m	0.4	2.8	0.209	0.189	0.007	0.01	0.08655
	UP7T				0.4	2.7					

*Water fraction per volume (water content; Water FV)

**Soil electric conductivity (S/m).

***Soil electric conductivity with corrected temperature (TC-EC, S/m)

****Pore water electric conductivity (S/m)

Supporting information 3: DNA extraction, sequencing and bioinformatic analyses

Total DNA was extracted from soil samples by means of QIAGEN DNeasy Powersoil Pro kit, following standard protocols. DNA extracts were subsequently subjected to quality controls and *16S rRNA* gene V3-V4 regions was sequenced by means of Illumina MiSeq sequencer (Illumina Inc., San Diego, CA, USA) at Arizona State University (Tempe, AZ, USA). Obtained reads were analyzed on ‘DADA2’ package (Callahan et al., 2016) for R programming language (R Core Team, 2021) to cluster the *16S rRNA* gene sequences in Amplicon Single Variants (ASVs). Taxonomic adscriptions were made by comparing ASVs sequences with SILVA database (v. r138.1, Quast et al., 2013). Statistical analyses and plotting were carried out employing ‘vegan’, ‘phyloseq’ and ‘ggplot2’ packages (McMurdie and Holmes, 2013; Oksanen et al., 2016; Wickham, 2016) for R programming language.

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Supporting information 5

Table showing SIMPER analysis results

ASVs	Family	Genus	Contribution (%)	Cumulative contribution (%)
ASV2	Chthoniobacteraceae	<i>Candidatus Udaeobacter</i>	2.727	2.727
ASV1	Gemmatimonadaceae	*	2.559	5.285
ASV11	Chitinophagaceae	*	1.435	6.72
ASV3	Bacteroidetes vadinHA17	*	1.388	8.108
ASV12	Bacteroidetes vadinHA17	*	0.995	9.093
ASV8	Chitinophagaceae	*	0.984	10.08
ASV26	Ktedonobacteraceae	*	0.903	11.92
ASV18	Chitinophagaceae	<i>Puia</i>	0.884	12.8
ASV19	Chthoniobacteraceae	<i>Chthoniobacter</i>	0.841	13.64
ASV17	Gemmatimonadaceae	<i>Puia</i>	0.793	14.44
ASV6	Chitinophagaceae	<i>Gemmatimonas</i>	0.766	15.2
ASV10	Chitinophagaceae	*	0.7	15.9

*ASV genus adscription non defined

Table showing the alpha diversity indices results

Sample ID	no. of sequences*	Richness (S)	Shannon (H)	Simpson (D)	Pielou's Phylogenetic diversity (PD)
LP1U	61182	1132	6.0176	0.993	2307.084
LP1T	46150	1028	6.1233	0.9953	2155.682
LP3U	71962	1248	6.1146	0.9945	2418.704
LP3T	67679	1032	5.6479	0.9882	2147.55
LP4U	101851	1648	6.574	0.9971	2901.562
LP4T	94714	1446	6.3037	0.9958	2696.216
UP5U	60071	888	5.4839	0.9828	1939.901
UP5T	82071	1014	5.7518	0.9912	2133.243
UP7U	48121	867	5.724	0.9922	1912.656
UP7T	85778	1220	6.084	0.9948	2374.081

*Number of sequences that passed the quality control bioinformatic processes

Supporting information 5 (cont.)

Table showing Bray-Curtis distances among sampling points based on microbial community composition (beta diversity, microbial community composition turnover)

Samples	LP1U	LP1T	LP3U	LP3T	LP4U	LP4T	UP5U	UP5T	UP7U
LP1T	0.908								
LP3U	0.602	0.851							
LP3T	0.903	0.598	0.85						
LP4U	0.63	0.85	0.526	0.851					
LP4T	0.841	0.642	0.747	0.574	0.762				
UP5U	0.675	0.952	0.803	0.953	0.838	0.927			
UP5T	0.722	0.907	0.776	0.923	0.814	0.877	0.495		
UP7U	0.57	0.943	0.693	0.936	0.777	0.907	0.465	0.58	
UP7T	0.649	0.931	0.728	0.934	0.773	0.89	0.527	0.473	0.424