

1 **Pedogenic and microbial interrelation in initial soils under**
2 **semiarid climate on James Ross Island, Antarctic Peninsula**
3 **Region**

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42 **Abstract.** James Ross Island (JRI) offers the exceptional opportunity to study microbial driven
43 pedogenesis without the influence of vascular plants or faunal activities (e.g. penguin
44 rookeries). In this study, two soil profiles from JRI (one at St. Martha Cove - SMC, and another
45 at Brandy Bay - BB) were investigated, in order to gain information about the initial state of
46 soil formation and its interplay with prokaryotic activity, by combining pedological,
47 geochemical and microbiological methods. The soil profiles are similar in respect to
48 topographic position and parent material but are spatially separated by an orographic barrier
49 and therefore represent windward and leeward locations towards the mainly south-westerly
50 winds. These different positions result in differences in electric conductivity of the soils caused
51 by additional input of bases by sea spray at the windward site, and opposing trends in the depth
52 functions of soil pH and electric conductivity. Both soils are classified as Cryosols, dominated
53 by bacterial taxa such as Actinobacteria, Proteobacteria, Acidobacteria, Gemmatimonadates
54 and Chloroflexi. A shift in the dominant taxa was observed below 20 cm in both soils as well
55 as an increased abundance of multiple operational taxonomic units (OTUs) related to potential
56 chemolithoautotrophic Acidiferrobacteraceae. This shift is coupled by a change in
57 microstructure. While single/pellicular grain microstructure (SMC) and platy microstructure
58 (BB) is dominant above 20 cm, lenticular microstructure is dominant below 20 cm at both soils.
59 The change in microstructure is caused by frequent freeze-thaw cycles and a relative high water
60 content and goes along with a development of the pore spacing and is accompanied by a change
61 in nutrient content. Multivariate statistics revealed the influence of soil parameters such as
62 chloride, sulfate, calcium and organic carbon contents, grain size distribution, and pedogenic
63 oxide ratios on the overall microbial community structure and explained 49.9% of its variation.
64 The correlation of the POR with the compositional distribution of microorganisms as well as
65 the relative abundance certain microorganisms such as potentially chemolithotrophic
66 Acidiferrobacteraceae-related OTUs could hint on an interplay between soil forming processes
67 and microorganisms.

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78 **1 Introduction**

79 In extreme environments, like Antarctica, local climatic conditions such as low temperatures,
80 precipitation or irradiance are important and often limiting factors for soil formation. Even
81 though soils in Antarctica are often poorly developed, they can be highly diverse (Michel et al.,
82 2014; Simas et al., 2008; Bockheim et al., 2015). Therefore, soil scientific investigations
83 became a relevant topic in Antarctic research, proving that there are actually soils in Antarctica
84 (Jensen, 1916) and identifying soil forming processes (Ugolini, 1964). Antarctic soil research
85 is mostly located in Victoria Land, continental Antarctica, especially in the McMurdo Dry
86 Valleys (Michel et al., 2014; Ugolini and Bockheim, 2008), in the South Shetlands, maritime
87 Antarctica (Simas et al., 2015) and the western Antarctic Peninsula Region (APR) (Haus et al.,
88 2015; Hrbáček et al., 2017b; Schaefer et al., 2017; Souza et al., 2014; Pereira et al., 2017).

89 Soils on continental Antarctica are often saline with thick salt horizons (Souza et al., 2014).
90 Due to environmental stressors such as very low temperatures, low water availability, frequent
91 freeze-thaw cycles and limited organic nutrient contents, soils from continental Antarctica show
92 limiting conditions for higher organisms (Cary et al., 2010). However, diverse microbial
93 communities thrive in a variety of Antarctic habitats, such as permafrost soils (Cowan et al.,
94 2014).

95 Soils in maritime Antarctica and western APR differ from soils in continental Antarctica
96 according to their stage of development (Balks et al., 2013; Blume et al., 2004; Parnikoza et al.,
97 2017). They show extensive cryoturbation processes with occasional salt crusts at the soil
98 surface (Balks et al., 2013; Bockheim, 1997). Local conditions determine nutrient availability
99 in Antarctic soils (Prietz et al., 2019). Ca, Mg, K and P contents are generally high in igneous
100 and volcanic rocks, whereas P and N contents are highest in ornithogenic soils. Ornithogenic
101 soils are well known in Antarctica. The World Reference Base for Soil Resources (WRB, 2014)
102 defines ornithogenic material (from Greek ornithos, bird, and genesis, origin) as material, which
103 is characterized by penguin deposits mainly consisting of guano and often containing a high
104 content of gravel transported by birds (cf. Ugolini, 1972). Soils from the eastern part of the APR
105 (also called Weddell Sea sector) are different, since they are associated with a dry climatic
106 transitional zone between the wet, warmer maritime Antarctica and colder, arid continental
107 Antarctica. Mean temperatures are below 0°C and liquid water supply is sufficient to allow soil
108 forming processes (Souza et al., 2014). Souza et al. (2014) also showed that cryoturbation is
109 less pronounced in the eastern APR than in the South Shetlands. The base saturation (>50%)
110 and electric conductivity (EC) are generally high whereas the amount of total organic carbon
111 (TOC) is substantially low. Regarding cryoturbation, active layer depth, chemical weathering

112 and soil organic C-content, soils from the eastern APR are comparable to soils from inland areas
113 of the Ross Sea Region (Balks et al., 2013), though they are formed on different parent material
114 (Daher et al., 2018). In comparison, eastern APR with its semiarid soils remains one of the least
115 studied areas in Antarctica (Souza et al., 2014; Daher et al., 2018).

116 Since Microorganisms in Antarctica show a broad diversity as revealed by recent molecular
117 phylogenetic and metagenomic methods (Cowan et al., 2014) and contribute to the weathering
118 of minerals in soils (Uroz et al., 2009), they are pivotal to understand initial soil formation. The
119 bacterial phyla Proteobacteria, Acidobacteria, Actinobacteria, Bacteroidetes, Firmicutes and
120 Gemmatimonadates, commonly found in temperate soils, also dominate the microbial
121 communities observed in Antarctic habitats (e.g. Bajerski and Wagner, 2013; Cary et al., 2010;
122 Pearce et al., 2012; Chong et al., 2012). The microbial community structure is influenced by
123 local soil chemical parameters, especially pH (e.g. Chong et al., 2010, Siciliano et al., 2014),
124 but also by soil physical parameters such as grain size distribution and soil moisture (Ganzert
125 et al., 2011). Chong et al. (2015) proposed, however, that historical contingency and dispersal
126 limitations could have a stronger influence on differences in community distributions at a
127 regional scale (>1000km). At the microscale, microbial activity such as photosynthesis and
128 nitrogen fixation has a distinct influence on soil chemical parameters, e.g. the increase of carbon
129 and nitrogen contents in oligotrophic soils (Ganzert et al., 2011; Cowan et al., 2011;
130 Niederberger et al., 2015). In return, these changes in soil characteristics affect microbial
131 community composition. Conflicting results illustrate the lack in the understanding of drivers
132 of soil microbial diversity in high latitude soils (Cowan et al., 2014). Since most of the non-
133 lichenized Antarctic fungi are known to be decomposers and their abundance and distribution
134 is limited by plant derived nutrients, and bio-available Carbon (Arenz et al., 2011), the focus of
135 this study lies on the prokaryotic interplay with soil characteristics and soil formation.

136 Micromorphological studies in the maritime Antarctica and the western APR described
137 sulphurization and phosphatization in ornithogenic soils and mineral transformation on
138 volcanic rocks (Pereira et al., 2013; Schaefer et al., 2008); and paleosols (Kirshner and
139 Anderson, 2011; Spinola et al., 2017). Even though micromorphology offers the opportunity to
140 study constituents of soil and their mutual relations in space and time and to identify soil
141 forming processes in an undisturbed state (Stoops, 2003), so far no micromorphological study
142 has been published about soil forming processes in the eastern APR that are influenced neither
143 by sulfates nor by birds.

144 Our study sites are located on James Ross Island in the eastern APR (Fig.1) and therefore offer
145 a unique setting to study soil formation and microbial communities in a transitional Antarctic

146 landscape between the wet maritime and dry, colder continental Antarctica. We selected two
147 different soils, representing coastal soils and inland soils of James Ross Island, developed on
148 similar substrate and at similar topographic positions, but differing in local climate conditions
149 and nutrient contents due to their relative position towards the mainly SW-winds. The western
150 study site (Brandy Bay –BB) is located in a windward position and is highly influenced by sea
151 spray, while the eastern study site (Santa Martha Cove – SMC), located behind a mountain
152 range, is located in a leeward position (Prietz et al., 2019). This setting enables an
153 investigation of interdependencies particularly between prokaryotic life and soil properties,
154 since the selected soils are not influenced by vascular plants, sulfides, and penguin rookeries.
155 With this, the main goal of our study is to identify major soil and microbiological properties in
156 an extreme environment by combining pedochemical and micromorphological methods with
157 microbial community studies based on high throughput sequence analyses. Thus, we will gain
158 a better general understanding of (i) the main soil forming processes and (ii) the drivers of soil
159 microbial diversity community structure in the eastern APR. This addresses also the question,
160 if the variance of pedogenic and microbiological properties are larger between depth increments
161 within one profile (e.g. with different distances to the permafrost table) or between different
162 soil profiles, i.e. due to different local environmental conditions.

163 **2. Material and Methods**

164 **2.1. Regional setting of James Ross Island, maritime Antarctica**

165 **[Figure 1]**

166
167 James Ross Island is situated east of the Antarctic Peninsula and is the largest island in the
168 western Weddell Sea sector (Hjort et al., 1997). The study area is located on Ulu Peninsula in
169 the northern part of JRI (Fig. 1). It represents one of the largest ice-free areas of the APR
170 (Nedbalová et al., 2013; Hrbáček et al., 2017b) with the beginning of its deglaciation 12.9 ± 1.2
171 ka ago (Nývlt et al., 2014). More than 300 km² of the JRI lowlands are currently ice-free, except
172 for a few glaciers (Engel et al., 2012).

173 The climate on JRI is semi-arid polar-continental (Martin and Peel, 1978). The precipitation,
174 mostly snow, ranges between 200 to 500 mm of water equivalent per year with the major share
175 during winter (Davies et al., 2013; Zvěřina et al., 2014). The thickness of the snow cover does
176 not exceed 30 cm, but varies due to strong winds (Hrbáček et al., 2017b; Hrbáček et al., 2016a).
177 The annual air temperature ranges between +10 °C and -30 °C on Ulu Peninsula (Hrbáček et
178 al., 2016a; Láska et al., 2011). The year 2015 marked the warmest summer ever measured on

179 Ulu Peninsula, having a mean seasonal summer temperature (MSST) of 0.0 °C and a maximum
180 air temperature of 13.3 °C (Hrbáček et al., 2017a); even though the mean annual air temperature
181 (MAAT) decreased slightly from -6.8 °C in 2011 to -7°C in 2015 (Hrbáček et al., 2016b; Láska
182 et al., 2012).

183 The two study sites are located at Brandy Bay (BB) near the western coast and at St. Martha
184 Cove (SMC) at the eastern coast of Ulu Peninsula. Both sites are located at similar topographic
185 positions (small plateaus) and elevation (80 m a.s.l.) with no visible vegetation (Fig. 2 and Fig.
186 3).

187

188 [**Figure 2**]

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190 BB is located windward towards the mainly south-westerly winds (Hrbáček et al., 2016c; Nývlt
191 et al., 2016), whereas SMC is located leeward, shielded by the Lachman Crags from the stronger
192 winds. This results in less precipitation in the eastern part of JRI (Davies et al., 2013). Therefore,
193 BB can be considered as a characteristic wind-exposed coastal site with high influence of sea
194 spray, whereas SMC represents a characteristic soil of an inland site with less influence of sea
195 spray.

196 The substrate of both study sites is basically composed of coarse-grained cretaceous sandstones
197 and siltstones of the Alpha Member of the Santa Martha Formation (Hrbáček et al., 2017b).

198 The land surface is generally covered by a debris layer of gravels and large clasts mixed with
199 loose sandy regolith, mostly derived from James Ross Volcanic Group basalts, which were
200 deposited as debris flows containing mainly basalt and hyaloclastite breccia and palagonite
201 (Davies et al., 2013; Hrbáček et al., 2017b; Salzmann et al., 2011). No nesting birds are found
202 on JRI.

203 The continuous permafrost on James Ross Island shows an active layer thickness ranging
204 between 40 and 107 cm related to the topographic position on Ulu Peninsula (Bockheim et al.,
205 2013; Borzotta and Trombotto, 2004).

206 **2.2 Soil sampling**

207 During the austral summer period in 2016 soil samples from BB and SMC (Fig. 4 and Fig. 5)
208 were taken. Both profiles were dug until a layer of coarse gravel was found. Bulk samples of
209 both profiles were taken in depth increments (0-5cm, 5-10cm, 10-20cm, 20-50cm, >50cm) and
210 were placed into sterile plastic bags, which were frozen immediately. Continuous cooling at -
211 20°C was ensured by a transfer with the research vessels *RV Polarstern* to Germany. For
212 micromorphological analyses, undisturbed and oriented samples were taken in modified

213 Kubiena boxes (10cm x 6cm x 5cm). Samples for micromorphology were taken at depth of 0-
214 10cm, 10-20cm, 30-40cm, 50-60cm and 80-90cm at SMC. BB samples represent the depth of
215 10-20cm, 20-30cm und 40-50cm. Soils were described according to Food and Agriculture
216 Organization of the United Nations (FAO) (2006) and classified according to the World
217 Reference Base for Soil Resources (WRB; IUSS Working Group WRB, 2015).

218 **2.3 Soil physical and chemical analysis**

219 **2.3.1 Grain size distribution**

220 The samples were saturated (100ml of deionized water) and sonicated (800J ml⁻¹). Coarse-
221 medium sand (>200µm), fine sand (63-200µm) and coarse silt (20-63µm) were obtained by wet
222 sieving. The smaller fractions, including medium silt (6.3-20µm), fine silt (2-6.3µm) and clay
223 (<2µm), were separated by sedimentation. Fractions >20µm were dried at 45°C and weighed
224 afterwards. The fractions <20µm were freeze-dried before weighing. The different procedures
225 were chosen due to practical reasons: freeze-drying allows submitting the finer fractions to
226 further analyses (particularly carbon and nitrogen content) immediately, while the coarser
227 fractions need milling anyway.

228 **2.3.2 pH, EC, C&N contents, major elements and pedogenic oxides**

229 The pH value was obtained using a pH meter (ph197i, WTW, Germany). Electrical conductivity
230 was measured with a conductivity meter (LE703, Mettler-Toledo, USA). Values of pH and
231 electric conductivity were measured from bulk samples < 2mm in deionized water with a
232 sample to water ratio of 1:2.5.

233 Carbon (C) and nitrogen (N) contents of the bulk soils were analyzed by dry combustion
234 (Elementar CNS Vario Max Cube). 300 to 500mg per sample were analyzed in duplicate. In
235 Order to distinguish between the total organic carbon (TOC) content and the total inorganic
236 carbon (TIC), TIC was removed by acid fumigation after Ramnarine et al. (2011). 100 mg of
237 the milled bulk soil samples were moistened with 20 to 40 µl of deionized water and put into a
238 desiccator together with 100ml of 37% HCl. Afterwards, the samples were dried at 40°C.
239 Finally, the samples were measured again by dry combustion (EuroVector EuroEA3000
240 Elemental Analyser) to obtain the TOC content. TIC content was calculated: $TIC = C_{tot} - TOC$
241 Major elements were analysed with a wavelength dispersive XRF device (AXS S4 Pioneer,
242 Bruker, USA). Prior to preparation, the samples (ratio Li-metaborate to soil 1:5) were ground
243 with an agate mill for 12 minutes. Major elements were used for the calculation of weathering
244 indices.

245 Pedogenic iron-oxides (Fe_d) were determined by dithionite-citrate-hydrogen carbonate
246 extraction (Holmgren, 1967). Poorly to non crystallised Fe-oxides (Fe_o) were determined by
247 acid ammonium extraction (Schwertmann (1964). The extractions were analysed at a
248 wavelength of 238.204 nm by an inductively coupled plasma optical emission spectrometer
249 (Vista Pro CCD Simultaneous ICP-OES, Varian, USA).

250 **2.3.3 Ion chromatography**

251 The initial water content in the investigated soil material was too low to extract sufficient
252 amounts of pore water for ion chromatography. Hence, the soil samples were leached, according
253 to Blume et al. (2011). Five grams of soil material were suspended in 25ml deionized water,
254 shaken for 90 minutes and centrifuged at 9000rpm to separate the soil material from the soil
255 solution and sterile filtered through a 0.22 μ m PES filter (Sartorius AG, Germany).

256 The ion concentrations in leached water samples were analysed by using two ion
257 chromatography (IC) systems (SYKAM Chromatographie Vertriebs GmbH, Germany). For
258 cations, the IC system consisted of a 4.6 x 200 mm Reprosil CAT column (Dr. Maisch HPLC
259 GmbH, Germany), an S5300 sample injector and an S3115 conductivity detector (both SYKAM
260 Chromatographie Vertriebs GmbH, Germany), 175mg L⁻¹ 18-Crone-6 and 120 μ L
261 methanesulfonic acid served as the eluent with a set flow rate of 1.2mL min⁻¹. The injection
262 volume was 50 μ L. The column oven temperature was set at 30°C. The Cation Multi-Element
263 IC-standard (Carl Roth GmbH + Co. KG, Germany) containing NH_4^+ , Ca^{2+} , K^+ , Li^+ , Mg^{2+} , Na^+
264 was measured before every replication series. For anions, the IC system consisted of a SeQuant
265 SAMS anion IC suppressor (Merck KGaA, Germany), an S5200 sample injector, a 3.0 x
266 150mm Sykrogel A 01 column and an S3115 conductivity detector (all SYKAM
267 Chromatographie Vertriebs GmbH, Germany). 6mM Na_2CO_3 with 90 μ M sodium thiocyanate
268 served as the eluent with a set flow rate of 1 ml min⁻¹ and a column oven temperature of 50°C.
269 The injection volume was 50 μ L. The multi-element anion standard containing F^- , Cl^- , Br^- , NO_2^-
270 , NO_3^- , PO_4^{3-} and SO_4^{2-} was measured before every replication series. The standards and
271 samples were measured in triplicates.

272 **2.3.4 Weathering indices and pedogenic oxide ratios**

273 The KN Index A ($(SiO_2+CaO+K_2O+Na_2O)/(Al_2O_3+SiO_2+CaO+K_2O+Na_2O)$) was calculated
274 after Kronberg and Nesbitt (1981). The index is based on the relative enrichment of the Al and
275 Si oxide phase and the leaching of Na, K and Ca. It ranges between 0 (prevailing chemical
276 weathering) and 1 (prevailing physical weathering). To get more precise information on the

277 ongoing chemical weathering, the chemical index of alteration (CIA)
278 $[(Al_2O_3/(Al_2O_3+Na_2O+CaO^*+K_2O)) \times 100]$ after Nesbitt and Young (1982), in which CaO*
279 represents the amount of silicate-bound CaO, was calculated. The CIA is frequently used as a
280 quantitative measure of feldspar breakdown, assuming that feldspar represents the most
281 abundant and reactive mineral. Higher values indicate increasing weathering intensity.
282 Additionally, the degree of iron release (Fe_d/Fe_t) after Blume and Schwertmann (1969) was
283 calculated, which gives information on the iron release from primary Fe-bearing mineral
284 weathering: a longer or more intensive weathering process is indicated by a higher ratio
285 (Baumann et al., 2014; Mirabella and Carnicelli, 1992).

286 **2.4 Micromorphology**

287 Samples for thin section preparation were air dried and afterwards embedded with a mixture of
288 resin (Viscovoss N55 S, Vosschemie, Germany), stabilized Styrene (Merck KGaA, Germany)
289 and hardener (MEKP 505 F, Vosschemie, Germany). After hardening, the samples were
290 formatted into plane-parallel blocks and halved in the middle using a saw (Woco Top 250 A1,
291 Uniprec Maschinenbau GmbH, Germany), and then one half was ground with the grinding
292 machine (MPS-RC Vacuum, G&N GmbH, Germany) and mounted onto a glass carrier. Then
293 the mounted samples was sawed into slices of about 150 μ m thickness. Finally, these slices were
294 ground to a thickness of 25 μ m. The preparation followed the instructions given by Kühn et al.
295 (2017). Afterwards, they were analyzed by using a polarizing microscope (ZEISS Axio
296 Imager.A2m, Software AxioVision 4.7.2, Carl Zeiss Microscopy GmbH, Germany) and
297 described following the terminology of Stoops (2003).

298 **2.5 Microbial community analysis**

299 **2.5.1 Nucleic acids extraction**

300 For each soil sample (maximum amount of 0.5g per sample), triplicates of total genomic DNA
301 were extracted using the FastDNATM Spin Kit for Soil (MO BIO Laboratories Inc., USA). The
302 extracted DNA was stored at -20°C and used as a template for the enumeration of target genes
303 by quantitative PCR (qPCR) and next-generation sequencing (Illumina HiSeq).

304 **2.5.2 Quantification of bacterial 16S rRNA gene copy numbers**

305 qPCR was used to quantify total bacterial abundances. All qPCR assays were performed in
306 triplicates on a CFX96 Real-time thermal cycler (Bio-Rad Laboratories Inc., CA, USA) and
307 contained 10 μ l SensiFAST SYBR Mix (Bioline GmbH, Germany), 5.92 μ l PCR water, 0.04 μ l

308 of forward and reverse primer (100 μ M) and 4 μ l template. The quantification of the bacterial
309 16S rRNA gene was based on the primers 341F (5'-CCTACGGGAGGCAGCAG-3') and 534R
310 (5'-ATTACCGCGGCTGCTGG-3') according to Muyzer et al., 1993. After an initial
311 denaturing phase of 3 minutes at 95°C, the cyclor included 35 cycles of 3 seconds at 95°C, 20
312 seconds at 60°C and 60 seconds at 72°C plus the plate read. All cycling programs included a
313 melting curve from 60°C to 95°C with 0.5°C steps per plate read. The analysis of quantification
314 data was performed with the CFX Manager™ Software (Bio-Rad Laboratories Inc., CA, USA).

315 **2.5.3 Illumina HiSeq-Sequencing**

316 Unique combinations of tagged 515F (5'-GTGCCAGCMGCCGCGGTAA-3') and 806R (5'-
317 GGACTACHVGGGTWTCTAAT-3') (Caporaso et al., 2010) primers were assigned to each
318 sample (Tab. S1, S2). For each sample, two PCR reactions were prepared and the PCR product
319 pooled after PCR reduce PCR variability. The PCR was performed on a T100™ Thermal Cycler
320 (Bio-Rad Laboratories Inc., CA, USA) in 25 μ l reactions, containing 0.125 μ l OptiTaq DNA
321 Polymerase and 2.5 10x Pol Buffer B (Roboklon GmbH, Germany), 1 μ l MgCl₂ (25mM), 1 μ l
322 dNTP Mix (5mM), 16.625 μ l PCR water, each 0.625 μ l of forward and reverse primer (20 μ M)
323 and 2.5 μ l genomic DNA. The following cyclor program was used: Initial denaturing step for 3
324 minutes at 95°C followed by 10 cycles of 1 minute at 94°C, 1 minute at 53°C (-0.2°C/cycle)
325 and 1 minute at 72°C, followed by 20 cycles of 1 minute at 94°C, 1 minute at 50°C and 1 minute
326 at 72°C, followed by a final extension step for 10 minutes at 72°C. All barcoded samples were
327 pooled into a single sequencing library by adding an equal amount of DNA (60ng DNA per
328 sample). Subsequently, a purification of the PCR product pool was achieved by using the
329 Agencourt AMPure XP – PCR Purification (Beckman Coulter, Inc., CA, USA). The Illumina
330 HiSeq-sequencing was performed by GATC Biotech AG, Germany.

331 **2.5.4 Bioinformatics and statistical analysis**

332 Sequencing was performed on an Illumina HiSeq (2 x 300 bp). Dual-indexed reads were
333 demultiplexed using CutAdapt (options: e0.1; trim-n; Martin, 2011). Barcode base pairs were
334 required to have a phred quality score of Q25 and no mismatches were allowed. Read pairs
335 were merged using PEAR (options: Q25; p10⁻⁴; o20; Zhang et al., 2013). The orientation of all
336 sequences were standardized by an own script using the information from demultiplexing.
337 Sequences containing low-quality base pairs were trimmed and filtered using Trimmomatic
338 (quality score of at least Q25 for trailing and leading base pairs, sliding window length of 5
339 basepairs, minimum sequence length of 200; Bolger et al. 2014). QIIME (version 1.9.1)

340 (Caporaso et al., 2010) was employed for microbiome analysis. USEARCH 6.1 (Edgar, 2010)
341 was used for the detection and removal of chimeric sequences. The SILVA database (version
342 128) (DeSantis et al., 2006) was utilized for the clustering of operational taxonomic units
343 (OTUs) (97% sequence similarity) and their taxonomic assignments. Singletons, OTUs
344 assigned to chloroplasts and mitochondria as well as rare OTUs (relative abundance of <0.1%
345 within each sample) were removed. Sample triplicates were merged by the mean value of their
346 relative abundance before visualization of the sequencing data and before analysis of correlating
347 environmental factors. For the processing and visualization of the obtained OTU table, R and
348 PAST3 (Hammer et al., 2001) were used. The hierarchical clustering of the samples using the
349 average linkage method was based on the Bray-Curtis dissimilarity. CANOCO5 (Šmilauer and
350 Lepš, 2014) was used for the canonical correlation analysis (CCA). If the Bonferroni corrected
351 *p*-value was <0.05, a given environmental parameter was included. Demultiplexed raw
352 sequencing data were submitted to the European Nucleotide Archive
353 (<http://www.ebi.ac.uk/ena>) under accession number: PRJEB29853.

354 **3 Results**

355 **3.1 Field properties and soil classification**

356 Both soils derived from coarse-grained marine sand- and siltstones, which were covered with
357 volcanic clasts. There was a higher contribution of volcanic material in BB than in SMC. The
358 amount of coarse material > 2mm was larger at the profile BB. Deflation processes led to a
359 residual enrichment of larger grains and pebbles at the soil surface of both profiles. The
360 permafrost table was not reached in both soil profiles, but ground ice was visible in a depth of
361 85cm at SMC. Neither SMC nor BB showed any ornithogenic influence. Both sites were
362 unvegetated by cryptogamic or vascular plants. The C-horizon was the only distinct soil horizon
363 occurring at SMC, whereas BB shows two changes within horizontal structures by abrupt
364 textural change below 10 cm and 20 cm. The textural change below 20 cm goes along with a
365 change in textural class; SCL (Sand: 52.5%, Silt: 21.9% and Clay: 25.6%) - CL (Sand: 44%,
366 Silt: 27.2% and Clay: 28.8%). Different from macroscopic features of the soil profiles, both
367 soils showed evidences of a downward transport and accumulation of particles and nutrients,
368 e.g. soluble products most likely originating from sea spray (Tab. 1). Accumulation starts at a
369 depth of 50cm at SMC and below 20cm at BB. Soil color did not change through the profiles.
370 SMC was brown to yellowish brown and BB was brownish yellow.

371 Both soils were classified as Cryosols (eutric, loamic) according to the WRB (IUSS Working
372 Group WRB, 2015).

373 **3.2 Grain size distribution and soil chemistry**

374 SMC had higher sand contents (mean value 61.7%, Table 1), while BB was characterized by
375 lower sand contents (mean value 47.4%) and higher silt and clay contents (mean values 25.3%
376 and 27.2% respectively). The grain size distribution varied only slightly with depth and similar
377 clay and silt contents were demonstrated for both soils.

378 The pH was slightly to moderately alkaline in both profiles and highly alkaline only in the upper
379 5cm of BB. The pH values followed opposing trends with depth, increasing in SMC from 7.7
380 to 8.1 and decreasing in BB from 8.6 to 7.4. The EC ranged between 50-60 $\mu\text{S cm}^{-1}$ in SMC and
381 was substantially higher in BB with a minimum of 350-450 $\mu\text{S cm}^{-1}$ within 5-50cm and its
382 highest values around 900 $\mu\text{S cm}^{-1}$ between 0-5cm and from 50cm downwards. According to
383 the EC values, SMC and the middle part of BB can be considered as being salt-free, whereas
384 the salt content in the upper and lowermost part of BB was low (Food and Agriculture
385 Organization of the United Nations (FAO), 2006).

386 The total inorganic carbon (TIC) content was low in both soils ranging between 0.1 and 0.3mg
387 g^{-1} in SMC and between 0.7 and 2.0mg g^{-1} in BB. The TOC content ranges from 0.8-0.9mg g^{-1}
388 for SMC and from 1.4 and 2.6mg g^{-1} for BB and increased there slightly with depth. The N
389 content was around 0.4mg g^{-1} across both soil profiles. The C/N ratio was generally low with
390 values below 7.5 in both soils, it decreased with depth in SMC (2.6 – 2.1) and increased with
391 depth in BB (4.0-7.4).

392 Ion concentrations (Tab. 1) were parallel to the depth function of the conductivity in both soils;
393 e.g. higher EC and ion concentration characterized BB. Cl^{-} concentrations decreased with depth
394 in SMC from 20.5 to 3.5 $\mu\text{mol g}^{-1}$ soil as well as in BB from 4,522 to 231 $\mu\text{mol g}^{-1}$ soil. The
395 highest SO_4^{2-} concentrations were observed in the shallow (SMC: 9.6 $\mu\text{mol g}^{-1}$ soil; BB:
396 621 $\mu\text{mol g}^{-1}$ soil) and deepest (SMC: 15.3 $\mu\text{mol g}^{-1}$ soil; BB: 451 $\mu\text{mol g}^{-1}$ soil) samples. K^{+} ,
397 Mg^{+} and Ca^{+} concentrations followed the same trend as SO_4^{2-} . Br^{-} , NO_2^{-} , NO_3^{-} and PO_4^{3-} . Li^{+}
398 and NH_4^{+} concentrations were below the detection limit.

399

400 [Table 1]

401

402 **3.3 Weathering indices and pedogenic oxide ratios**

403 Weathering indices were calculated according to the major element contents (Table 3). The KN
404 Index A was at 0.91-0.92 in SMC and only slightly lower with 0.89 - 0.90 in BB (Table 2). The
405 CIA varied between 53.9 and 54.8 in SMC and between 56.9 and 58.8 in BB. Both indices
406 indicated weak chemical weathering with a slightly higher weathering intensity in BB.

407

408 [**Table 2**]

409

410 The Fe_d/Fe_t ratio showed a decreasing trend from 0.18 to 0.11 with depth in SMC indicating a
411 decreasing intensity of pedogenic processes with depth. No particular trend was found in BB;
412 but the Fe_d/Fe_t ratio is – similar to the CIA - generally higher around 0.20 except for 0.16 in the
413 upper 5cm.

414 **3.4 Micromorphology**

415 Formation of platy and lenticular aggregates due to repeated freezing and thawing processes
416 was detected. Neither platy and lenticular platy structures nor the results of translocation
417 (eluviation) processes were observed during fieldwork, but could be confirmed later using
418 micromorphology.

419 SMC had a weak to moderately developed pedality and a weak to moderate degree of separation
420 (Table 3). Both, pedality and degree of separation are well developed at a depth of 50-60cm
421 and were lowest developed close to the surface and at the bottom of the profile. In contrast, BB
422 had a well-developed pedality and a moderate to high degree of separation with its maximum
423 development close to the bottom of the profile.

424

425 [**Table 3**]

426

427 Lenticular and subangular blocky microstructures were present in both profiles, whereas
428 lenticular microstructure was dominant in SMC and subangular blocky microstructure was
429 dominant in BB. Lenticular shaped aggregates were first observed at a depth of 10cm in profile
430 BB, and at 30cm in SMC (Figures 3a and 3b).

431

432 [**Figure 3**]

433

434 Translocations features, like cappings consisting of clay and silt particles welded together with
435 sand-sized quartz grains were present in the upper part of both profiles. Link cappings occurred

436 in the lower part of both profiles, with lesser and smaller cappings in BB (Fig. 3d). Link
437 cappings were very rare and occurred only where coarse rock fragments were located close to
438 each other. Dusty silt and clay pendants occurred only in the lower part of BB (20-50cm) (Fig.
439 3e). The sphericity of mineral grains was smooth in both profiles. The minerals were slightly
440 better rounded in BB (subangular to round) than in SMC (subangular to subrounded).
441 Weathering processes were identified by pellicular and dotted alteration patterns on rock
442 fragments (mostly in sandstone fragments) in both profiles with a higher number of fragments
443 with dotted alteration patterns than with pellicular alteration patterns. The quantity and intensity
444 of dotted alteration patterns decreased with depth. Larger rock fragments were often strongly
445 weathered, so that mainly quartz-minerals were still preserved (Fig. 3f). Besides quartz,
446 glauconite is the main mineral component in the unweathered sandstone fragments. In addition,
447 feldspars and micas occur to a very small extent. The sandstones cemented by fine material and
448 faint Fe coatings are visible around quartz grains. Pellicular alteration pattern was found
449 exclusively on volcanic rock fragments, and only in the uppermost thin section (0-10cm) of
450 SMC (Fig. 3g). Fragments showing pellicular alteration patterns occurred in 10-30cm of BB.
451 Even though the number of weathered fragments decreased, pellicular patterns were slightly
452 thicker in slide BBII (20-30cm) than in BBI (10-20cm). However, pellicular alteration patterns
453 did not exceed the state of “pellicular” in any analyzed slide whereas dotted alteration patterns
454 often reach the state of “patchy cavernous residue” (Fig. 3e) and do occur also as dispersed
455 minute residues (Stoops, 2003).

456 **3.5 Microbial abundance and community structure**

457 The enumeration of the 16S rRNA gene revealed a similar trend for both soil profiles (Fig. 4).
458 The highest abundances with 6.6×10^8 copies g^{-1} soil (BB) and 1.7×10^8 copies g^{-1} soil (SMC)
459 were detected in the uppermost depth increment of both soil profiles. Both soils showed a
460 decrease in bacterial abundances with depth. The lowest bacterial abundances in SMC were
461 detected below 50cm depth with 3.7×10^5 copies g^{-1} soil, and in BB in 20-50cm depth with 1.7
462 $\times 10^6$ copies g^{-1} soil.

463 In total, 19,732,536 reads were obtained after merging the forward and reverse reads,
464 demultiplexing, filtering, and deletion of chimeric and singleton sequences. Additionally, reads
465 of chloroplast-associated OTUs (36,573), mitochondria-associated OTUs (1,117) as well as rare
466 OTUs (OTUs with a relative abundance of $<0.1\%$ in every sample; 4,287,382) were filtered,
467 resulting in 15,407,464 reads (Tab. S4). The number of reads per sample ranged from 54,122
468 to 916,583 with a mean value of 513,582. A total of 687 OTUs was clustered. After taxonomic

469 classification, 258 putative taxa were obtained. Shannon's H index was used to estimate and
470 compare the alpha diversity of the different depth increments interval of the soils (Tab. S5).
471 Both soils showed a similar Shannon's H index, which ranged from 3.7 to 4.7 not following
472 any specific trend.

473 Bacteria dominated the microbial community in both soil profiles (Fig. 4). Higher abundances
474 of Thaumarchaeota (7.2 - 12.9%) were found in the upper 10cm of the soil profile from SMC
475 (Tab. S4). On a phylum level, the soil profile of SMC was dominated by Proteobacteria (23.4 -
476 57.9%) and Actinobacteria (17.7-41.3%) but showed also relative high abundances of
477 Acidobacteria (3.9-14.1%). The microbial community in BB was also mainly composed of
478 Proteobacteria (28.2-30.8%), followed by Actinobacteria (27.6-46.6%), Gemmatimonadetes
479 (3.9-24.7%) and Chloroflexi (5.3-10.9%). Bacteroidetes were highly abundant (10.5%) in the
480 top 5 cm of BB. Regarding potential phototrophic organisms in the investigated soils, the
481 amount of chloroplast-related reads was relatively low (<0.2%) in each sample, except for SMC
482 >50 cm (0.03% - 1.30%) and BB 0 – 5 cm (0.87% - 1.01%). Cyanobacteria-related OTUs were
483 rare and only showed low relative abundances in SMC 5 – 10cm (0.06%), SMC 10 – 20cm (;
484 0.62%), SMC >50cm (0.04%).

485

486 [Figure 4]

487

488 The distribution of dominant OTUs was reflected by a cluster analysis based on the Bray-Curtis
489 dissimilarity of the investigated depth increments. Samples were clustered according to their
490 origin and depth. On a first level, samples grouped according to depth in upper (0-20cm) and
491 deeper (20-80cm) samples and within these groups they clustered according to location (BB vs.
492 SMC). An exception is the sample from BB from the depths 0-5cm which formed an own
493 cluster (Fig. 5). The deeper samples in both profiles (20-80cm depth) showed high relative
494 abundances of three OTUs related to Acidiferrobacteraceae(1, 2, 3) (SMC: 1.7-14.6%; BB: 2.2-
495 9.8%) and one OTU related to Gemmatimonadaceae(1) (SMC: 1.5-3.8%; BB: 14.1-20.3%).
496 High proportions of two OTUs related to Gammaproteobacteria(1, 2) (SMC: 2.8-11.4%; BB:
497 5.4-10.2%) and one OTU related to Gaiellales(2) (SMC: 3.7-5.7%; BB: 7.2-8.3%) were
498 observed in the shallow samples (0-20 cm). BB 0-5 cm was comprised of a strongly different
499 community. The most abundant taxa in this sample were related to *Thermomonas*(1) (6.4%),
500 *Sphingomonas* (3.7%) and *Solirubrobacterales*(1) (3.7%).

501

502 [Figure 5]

503

504 The relationship of OTU distribution and environmental parameters was examined by applying
505 a CCA (Fig. 6). Contents of chloride (18.5%), calcium (11.8%), sulfate (5.9%), silt (5.6%),
506 TOC (6%) and the Fe_d/Fe_t -ratio (12.5%) formed the optimal subset to explain variations in
507 community structure of the investigated soil profiles ($p < 0.05$). The adjusted explained
508 compositional variation was 49.9%. A strong correlation between the unique community of BB
509 0-5cm and the saline conditions was observed, mainly caused by high sulfate and chloride
510 concentrations. The remaining samples were arranged according to sample site and depth as
511 already observed in the cluster analysis above.

512

513 [**Figure 6**]

514 **4 Discussion**

515 The interaction of biotic and abiotic processes remains one of the fundamental questions in
516 ecosystem research and further the initial development of soils under harsh environmental
517 conditions, such as Antarctica. So far, only a few studies exist for polar environments that
518 integrate pedogenic and microbiological research (e.g. Aislabie et al. 2008, Cowan et al. 2014,
519 Ganzert et al. 2011; Bajerski and Wagner, 2013). James Ross Island offers an exceptional
520 opportunity to improve our understanding of the interrelations between soil formation and
521 microbiological properties in the absence of plants. The present interdisciplinary study gives
522 profound insights in the state of soil formation and microbial community structure in initial
523 soils in the transition zone between maritime and continental Antarctica.

524 James Ross Islands is located in the transition zone between warmer and wetter maritime
525 Antarctica and cold and dry continental Antarctica (Souza et al., 2014). In this area, we studied
526 two representative soils 16km apart, with different exposures to the dominant south-westerly
527 winds. The leeward position of SMC displays formation conditions of a typical inland soil,
528 while BB in its windward position represents coastal soils. As indicated by EC values, BB is
529 influenced by sea spray, while SMC, sheltered behind the Lachman Crags, does not show strong
530 input of soluble salts from sea spray.

531 The examined soils on JRI were characterized by low TOC ($0.9\text{-}2.6\text{mg g}^{-1}$) and low total
532 nitrogen contents (approx. 0.4mg g^{-1}), which is common for Antarctic soil environments (e.g.
533 Cannone et al., 2008), and relative high pH values (7.4- 8.6). The moderately to highly alkaline
534 pH in both soils cannot be explained by the occurrence of $CaCO_3$, because the soils have a
535 negligible amount with of TIC ($\leq 2\text{mg g}^{-1}$). Low C contents do not only show the missing
536 influence of penguins, but also indicate a relative juvenility of the soils: This indicates that no

537 cations have been leached from the topsoil, and therefore the pH remains neutral to basic
538 (Wilhelm et al., 2016). In addition, the content of basalt clasts in the parent material results in
539 increased soil pH values (Simas et al., 2002; Moura et al., 2012). The opposing trends in the
540 depth function of the pH values are caused by the input of soluble salts from sea spray: wind
541 can transport soluble salts from the sea causing an additional input of bases simultaneously
542 increasing the pH at BB, while SMC is not affected (Benassai et al., 2005; Russell et al., 2010;
543 Hara et al., 2004; Udisti et al., 2012). Since the substrate was not colonized by plants, lichens
544 or endolithic prokaryotes, and the taxonomic data revealed low abundances of phototrophic
545 organisms, the alkalization of the substrate by the release of hydroxyl ions in the course of
546 photosynthesis has a minor effect on soil pH. On the other hand, the neutral to basic pH does
547 not significantly affect the soil microbial community structure, which is in accordance with
548 observations in soils from Livingston Island (South Shetland Archipelago, maritime Antarctica)
549 by Ganzert et al. (2011). They explained it by the occurrence of a specific soil microbial
550 community, which thrives under low C and N conditions and is not depending on nutrient input.
551 Therefore, pH is mainly driven by the parent material composition combined with the input of
552 soluble salts in these young soils on JRI.

553 The additional input of airborne cations by sea spray led to higher sodium and calcium contents
554 and a rejuvenation of the affected depth increments of the soil profile, which can be seen in the
555 lower CIA values in 0-5 cm soil depth of both soils compared to the lower part of the profiles.
556 Ions, for instance sulfate accumulate close to the permafrost table, which acts as a barrier and
557 therefore explains increasing contents of sulfate with depth. The high amount of sulfate near
558 the surface is most likely caused by sea spray and precipitation, because they are known to carry
559 high amounts of sulfate in coastal areas (Blume et al., 2010).

560 Chemical weathering, as indicated by the KN-Index A (Kronberg and Nesbitt, 1981), is only of
561 minor importance whereas physical weathering is prevailing. The CIA and pedogenic oxide
562 ratios (POR) confirmed the low degree of soil formation. Pedogenic oxides with specific
563 degrees of crystallization relate to intensity and/or duration of pedogenic processes (Baumann
564 et al., 2014; Blume and Schwertmann, 1969; Mirabella and Carnicelli, 1992). The results show
565 that both CIA and both POR are slightly higher at BB compared to SMC. The KN-Index A and
566 the CIA showed a weak chemical weathering of these mineral soils (Michel et al., 2014). Both
567 indices indicated a more intensive chemical weathering at BB and, thus, indicate a slightly
568 stronger pedogenesis at BB than at SMC. This finding could be explained by the sea- and
569 windward position of BB, which results in an increased water availability and a slightly more
570 levelled microclimate. Since both soils are located in similar topographic positions and derived

571 from similar parent material, CIA and POR results allow the interpretation that soils influenced
572 by coastal conditions tend to be more weathered. Besides physical and chemical weathering,
573 microorganisms play an important role in mineral dissolution and oxidation. Adapted
574 microorganisms colonize minerals and are, depending on nutritional requirements, nutrient
575 availability and mineral type, potential contributors to the weathering of minerals (Uroz et al.,
576 2009). Taxonomical groups, which are usually connected to microbial weathering, are present
577 in the soils, such as *Massilia*, *Bacillus* (Ma et al., 2011) and *Polaromonas* (Frey et al., 2010).
578 Interestingly, the relative abundances of these taxa changed according to the degree of
579 weathering. This could indicate a possible interrelation between the occurrence of these
580 potential weathering-related organisms and the degree of weathering of Antarctic soils.

581 Evaluating weathering using the CIA, it must be noted that the value for BB is most likely
582 underestimated. Ion chromatography results show that Na-content is much higher at BB. The
583 high amount of Na is most likely caused by sea spray, which is known to carry high amounts
584 of Na (Udisti et al., 2012). Since the calculation of the CIA takes Na into account (Nesbitt &
585 Young, 1982), the CIA values would be significantly higher if the additional input of sea salts
586 could be excluded. It is very likely that the actual difference in state of weathering between
587 SMC and BB would be much higher. In conclusion, chemical weathering, even without
588 influence of guano deposits, is of higher importance for the current state of soil formation, than
589 the ongoing cryoturbation.

590 In case of the pedogenic oxide ratios, 12.5% of the total compositional variation could be
591 explained, which indicates a correlation between the microbial community structure and
592 weathering at a very initial stage of soil formation. The pedogenic oxide ratios correlate with
593 the compositional distribution of microorganisms in the investigated soils, and with the relative
594 abundances of one Acidiferrobacteraceae-related OTU. Microorganisms of this family are
595 described as autotrophic sulfur and iron oxidizers, which have the capacity to use ferrous iron,
596 thiosulfate, tetrathionate, sulfide and elemental sulfur as electron donors and oxygen or ferric
597 iron as terminal electron acceptor (Hallberg et al., 2011). The reactive iron could potentially be
598 used as terminal electron acceptor in the course of microbial iron cycling (Canfield, 1989).
599 Organic matter, a potential substrate for heterotrophic microbial processes, sorbs on mineral
600 surfaces (Kaiser and Guggenberger, 2000) and could be released in the course microbial
601 oxidation and reduction of reactive iron phases. In addition to the autotrophic processes, the
602 release of sorbed, organic matter from mineral surfaces could be an additional way to increase
603 the pool of biologically available carbon. The availability of such a mechanism potentially has

604 an influence on the microbial community structure and abundances in oligotrophic
605 environments.

606 Translocation features are common features in permafrost-affected soils. They often occur
607 together with platy rectangular or lenticular aggregates, caused by reoccurring freeze-thaw-
608 cycles (Van Vliet-Lanoë, 1985). Platy blocks and lenses dominated the microstructure in the
609 areas between 20 and 50cm of both profiles. They were absent near the surface of both profiles
610 and at the bottom of the profile SMC. These microstructures are known to occur in the transition
611 zone between permanently frozen and unfrozen soils (Shur et al., 2005; Van Vliet-Lanoë et al.,
612 2004). Here, the alternating temperature and soil moisture conditions additionally affect the
613 microbial community structure. The frequency of freeze-and-thaw cycles tends to be steady in
614 the middle part of a permafrost-affected soil, whereas weather shifts influence the surface,
615 causing several freeze-and-thaw events per day, which do not result in typical microstructure
616 formation due to insufficient water supply (Van Vliet-Lanoë, 1985). Aggregate formation by
617 reoccurring freeze-and-thaw cycles result in a change in pore shape and size (Van Vliet-Lanoë
618 et al., 2004). Especially during the summer season, intensive insolation causes high evaporation,
619 resulting in dry soil surfaces. Changes in pore space affects microbial habitats, due to larger
620 pores and a more sufficient water supply. This has a severe influence on matter fluxes and soil-
621 environmental conditions, which is reflected in a changing species distribution and, more
622 specifically, the occurrence of different clusters of highly abundant organisms in both soils.
623 Multivariate statistics were performed for soil depth increments considered as being
624 independent. However, when processes are discussed that link between soil horizons, e.g. water
625 and solute flow through the profiles, we account for the limited number of two soil profiles with
626 great care. We could not detect any environmental factors that increase or decrease the
627 correlation between the chosen depth increments. Nevertheless, freeze-and-thaw cycles
628 definitely also occur in the upper part of the profile, as indicated by the well sorted areas (Van
629 Vliet-Lanoë, 1985), which were described as single grain microstructure. Near the permafrost
630 table aggregates are often formed by frost desiccation and are hence poorly compacted what
631 makes them unstable upon moistening, which occurs during thawing events and explains the
632 missing platy microstructure at SMC near the bottom of the profile (Van Vliet-Lanoë, 2010).
633 The fact that lenticular shaped aggregates occur also in the lower part of the profile indicates
634 that the permafrost table is located underneath the layer of coarse gravel at BB.

635 Although the investigated soils were poorly developed, an abundant and diverse prokaryotic
636 community could be observed. Microbial abundances in both soils showed a decreasing trend
637 with depth. Values of up to 10^9 gene copies g^{-1} soil in the uppermost depth increments are

638 comparable to observed microbial abundances from other cold environments, such as alpine
639 glacial forelands (Sigler et al., 2002), permafrost-affected soils from arctic regions (Liebner et
640 al., 2008) and Antarctic glacier forefields (Bajerski and Wagner, 2013).

641 Both soils were characterized by a highly diverse community dominated by Proteobacteria,
642 Actinobacteria, Gemmatimonadetes, Acidobacteria and Chloroflexi, which is in accordance
643 with the observations in other continental and maritime Antarctic habitats (e.g. Yergeau et al.,
644 2007; Cary et al., 2010, Ganzert et al., 2011, Bajerski and Wagner 2013, Wang et al., 2016).
645 Substantial differences in geochemical parameters such as conductivity, the change of the
646 community structure on a phylum level were evident as well as the occurrence of depth-
647 dependent clusters (0-20 cm; >20 cm) of dominant OTUs (Fig. 8). Whereas the upper 20cm of
648 the soils were dominated by Gammaproteobacteria and Gaiellales, the deeper part of the soils
649 showed increased abundances of OTUs related to Acidiferrobacteraceae and
650 Gemmatimonadaceae. This distinct shift correlates with the occurrence of the microstructure
651 related to freezing and thawing and could be related to its changes of the pore space and the
652 availability of oxygen, water and nutrients. For instance, Gemmatimonadaceae were a common
653 observation in the soils and showed increased abundances in deeper parts of BB. These
654 organisms have a cosmopolitan distribution in terrestrial environments and depend on the soil
655 moisture condition of the respective soil and soil depth (DeBruyn et al., 2011; Bajerski and
656 Wagner, 2013). Only a few isolates have been described for this phylum (e.g. Zeng et al., 2015)
657 and their exact functions in soil ecosystems remain uncertain. The change in relative abundance
658 of these taxa with depth could be coupled to the changing availability of water, which depends
659 on the microstructure. For example, the amount and size of microaggregates have been shown
660 to be important regarding prokaryotic colonization, leading to genetically distinct communities
661 as well as cell densities in different size classes of aggregates (Ranjard et al., 2000). Thus, in
662 addition to environmental parameters, which shape the overall prokaryotic community, the
663 microstructure of the initial soils could have a substantial influence on species distribution.

664 Higher abundances of Bacteroidetes- and especially Flavobacteriaceae-related OTUs were
665 observed in the uppermost area of soil from BB, while only showing minor abundances in the
666 deeper soil areas. This area differed from the remaining soil in two regards, namely very high
667 chloride concentrations and a relative high content of coarse sandy material and could select
668 for adapted psychro- and halotolerant Bacteroidetes-related organisms, such as
669 Flavobacteriaceae (e.g. Bajerski et al., 2013a). Members of the Flavobacteriaceae family
670 detected in this area, for instance *Gillisia sp.*, were isolated from Antarctic habitats before and
671 were shown to be at least moderately tolerant to saline conditions (Bowman and Nichols, 2005).

672 Putative halotolerant or halophilic Flavobacteriaceae in this area could have a need for high
673 chloride contents. Chloride can be accumulated inside the cell to osmotically balance the
674 cytoplasm with the surrounding habitat (Oren et al., 2002; Müller and Oren, 2003).
675 Furthermore, the detected Bacteroidetes-related organisms could prefer the coarser, sandy
676 microstructure from this depth increment. The preference of microbial groups for certain grain-
677 size-dependent microenvironments, for instance the sand-sized fraction being preferred by
678 Bacteroidetes, was shown, e.g. in Typic Hapludalfs from central Denmark (Hemkemeyer et al.,
679 2018).

680 Both investigated soils were poor in soil organic C as well as N. Organisms with the ability to
681 use oxygenic photosynthesis to fixate CO₂, such as cyanobacteria, were nearly absent in the
682 investigated soils. Low abundances of Cyanobacteria are a common observation for Antarctic
683 soil habitats (Ji et al., 2016). Due to the lack of phototrophic organisms and organic carbon,
684 inorganic compounds and metabolic pathways utilizing those may have a more pronounced role
685 in sustaining the microbial ecosystem at this initial stage of the soils. Several of the most
686 abundant taxa observed in BB and SMC were putative chemoautotrophs involved in nitrogen,
687 iron and sulfur cycling, such as potential ammonia-oxidizing Thaumarchaeota or sulfur/iron-
688 oxidizing Acidiferrobacteraceae. Microorganisms can be seen as the primary pioneers of
689 nutrient-poor environments such as Antarctic soils, and were shown to have the genetic
690 potential to fix C and N (Cowan et al., 2011; Niederberger et al., 2015), thus increasing C and
691 N contents of these oligotrophic soils. The chemoautotrophic Thaumarchaeota oxidize
692 ammonia aerobically to nitrite (Brochier-Armanet et al., 2008; Vajrala et al., 2013) and were
693 observed in many studies located in Antarctica (Magalhães et al., 2014; Ayton et al., 2010).
694 However, ion chromatography showed that amounts of ammonia as well as nitrite and nitrate
695 were negligible in both soils. Ammonia originating from necromass and products in the course
696 of nitrification could be metabolized directly by the present community, so no accumulation of
697 the different intermediates containing nitrogen takes place. These organisms are reported to
698 have the genetic potential to use the hydroxypropionate/hydroxybutyrate pathway for CO₂
699 fixation, which is highly efficient and could provide an ecological advantage in oligotrophic
700 environments (Könneke et al., 2014). Further, a part of the community could use atmospheric
701 compounds as energy source. Atmospheric H₂, CO, and CO₂ are scavenged and used as an
702 energy source by microorganisms, especially organisms associated with the phyla
703 Actinobacteria, Chloroflexi, Acidobacteria, Planctomycetes, Verrucomicrobia, and
704 Proteobacteria (Greening et al., 2015; Ji et al., 2017). Operational taxonomic units related to
705 the phylum Actinobacteria and the associated orders Acidimicrobiales and Solirubrobacterales

706 were highly abundant in the investigated soils. Microorganisms in Antarctic soils, especially
707 bacteria related to the phyla Actinobacteria, AD3 and WPS-2, were shown to generate biomass
708 by consuming H₂, CO₂ and CO from the atmosphere (Ji et al., 2017). The gene for
709 chemosynthetic CO₂ fixation, *rbcLIE*, was found in multiple orders, including
710 Pseudonocardiales, Acidimicrobiales and Solirubrobacterales. Similar functional capabilities
711 could be present and active in the investigated soils. Our results show that, in this initial stage
712 of soil development, chemolithoautotrophic lifestyles plays an important role for the generation
713 of biomass and initial accumulation of soil organic carbon and nitrogen.

714 **5. Conclusion**

715 The presented soil and microbiological study on initial soils in the semiarid environment of
716 Antarctica shows the current state of soil formation indicated by main soil and microbiological
717 properties and their interplay. The results allow us to draw the following conclusions:

- 718 1. Despite similarities in topographic position and substrates, both profiles have distinct
719 differences in chemistry (content of salts indicated by EC, opposing trends in pH and
720 states of weathering, indicated by WI and POR) and microbiology (depth functions of
721 microbial abundances and diversity, e.g. Proteobacteria, Gemmatimonadetes and
722 Thaumarchaeota abundances), which are caused by the different local environmental
723 conditions at each site.
- 724 2. The EC values of the soils and the depth function of the pH values clearly showed
725 different conditions for soil formation at the two sites caused by the more exposed
726 location of BB towards the mainly south-westerly winds, resulting in a more intense
727 weathering and higher input of salt by sea spray.
- 728 3. Taking weathering and aggregation as indicators of soil formation, we conclude that
729 coastal conditions - in contrast to inland conditions - favor the formation of soils in
730 maritime Antarctica.
- 731 4. Despite different local environmental conditions at each site, the microbial communities
732 differ more distinctly between the depth increments in one profile than between the two
733 profiles. Therefore, we conclude that in this initial stage of soil formation factors such
734 as weathering and microstructure formation, as well as the resulting parameters (e.g.
735 water availability and matter fluxes), are more important drivers of soil microbial
736 community composition than chemical parameters such as EC and pH.
- 737 5. Assuming that prokaryotic life is highly affected by changes in soil structure and vice
738 versa, further investigations in this field should include analyses of (micro-) aggregates.

739

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741 Thomas Scholten and Carsten W. Mueller. Lars A. Meier and Carsten W. Mueller carried out
742 fieldwork during the PROANTAR fieldtrip led by Carlos E.G.R. Schaefer in 2016. Lars A.
743 Meier, Patryk Krauze, Isabel Prater and Fabian Horn did analyses and interpretation. Lars A.
744 Meier and Patryk Krauze prepared this manuscript with contributions from all co-authors.

745

746 *Competing interests.* The authors declare that they have no conflict of interests.

747

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1059 **Table 1: Soil properties of two soil profiles from St. Marta Cove (SMC) and Brandy Bay (BB) from James Ross Island,**
1060 **Antarctica.**

Sample	Depth [cm]	pH _{Hz20}	EC [μS cm ⁻¹]	TIC [mg g ⁻¹]	TOC [mg g ⁻¹]	N [mg g ⁻¹]	C/N	Na ⁺ [μmol g ⁻¹]	K ⁺ [μmol g ⁻¹]	Mg ⁺ [μmol g ⁻¹]	Ca ⁺ [μmol g ⁻¹]	Cl ⁻ [μmol g ⁻¹]	SO ₄ ²⁻ [μmol g ⁻¹]	Sand 63–2000 μm [%]	Silt 2–63 μm [%]	Clay <2 μm [%]
SMC 0-5	0-5	7.7	46	0.1	0.9	0.4	2.6	41.5	2.5	4	10.4	20.6	9.6	61.2	18.9	19.8
SMC 5-10	5-10	8	36	0.1	0.9	0.4	2.5	30.4	2.4	3.6	9.6	13.1	5.7	59.9	19.4	20.7
SMC 10-20	10-20	7.9	33	0.3	0.9	0.4	2.3	27.1	2	3.1	8.3	8.7	3.3	63.8	17.1	19.1
SMC 20-50	20-50	8	33	0.1	0.8	0.4	2.2	38.6	1.5	2.1	4.9	5.5	3	61.9	17.2	20.8
SMC >50	>50	8.1	65	0.2	0.9	0.4	2.1	91.5	2.7	3.1	6.3	3.5	15.3	61.7	20	18.3
BB 0-5	0-5	8.6	950	1.4	1.4	0.4	4	1590	23.4	84.6	151	4522	621	49.8	25.2	24.9
BB 5-10	5-10	8.1	561	1.2	2.1	0.4	5.6	470	16.3	57.4	108	702	123	46.4	25.7	27.9
BB 10-20	10-20	7.7	385	0.7	2	0.3	5.9	268	12.2	42.6	93	369	88	52.5	21.9	25.6
BB 20-50	20-50	7.6	505	2	2.5	0.4	6.7	191	18.3	79.8	173	386	163	44	27.2	28.8
BB >50	>50	7.4	965	1	2.6	0.4	7.4	149	23.9	140	297	231	451	44.3	26.8	28.9

1063 **Table 2: Weathering indices (WI) and pedogenic oxide ratios (POR) of two soil profiles from St. Marta Cove (SMC)**
 1064 **and Brandy Bay (BB) from James Ross Island, Antarctica. CIA = chemical index of alteration; KN-A = Kronberg**
 1065 **Nesbitt Index; Fe_d = dithionite-soluble iron; Fe_t = total iron; Fe_o = oxalate-soluble iron.**

Sample	Depth [cm]	WI		POR				
		CIA	KN-A	Fe _d /Fe _t	Fe _o /Fe _d	Fe _t [mg g ⁻¹]	Fe _d [mg g ⁻¹]	Fe _o [mg g ⁻¹]
SMC 0-5	0-5	53.9	0.92	0.18	0.56	45.57	7.99	4.48
SMC 5-10	5-10	54.2	0.91	0.18	0.45	44.71	7.83	3.56
SMC 10-20	10-20	54.8	0.91	0.16	0.53	40.74	6.61	3.48
SMC 20-50	20-50	54.3	0.91	0.15	0.59	40.76	5.96	3.53
SMC > 50	>50	54.1	0.92	0.11	1.72	42.25	4.83	8.3
BB 0-5	0-5	56.9	0.89	0.16	0.61	53.77	8.68	5.3
BB 5-10	5-10	58.5	0.89	0.21	0.57	44.09	9.08	5.19
BB 10-20	10-20	58.1	0.9	0.2	0.58	42.57	8.34	4.85
BB 20-50	20-50	58.8	0.9	0.21	0.56	39.82	8.43	4.68
BB > 50	>50	58.2	0.9	0.21	0.54	38.18	7.88	4.24

1066 **Table 3: Micromorphological features of two soil profiles from St. Marta Cove (SMC) and Brandy Bay (BB) from James**
 1067 **Ross Island, Antarctica**
 1068 **The micromorphological property is shown by the presence (cross) or absence (no cross). (x) = occasional occurring**
 1069 *** Microstructures separated by "/": two different microstructures were found. Microstructures separated by "()": one**
 1070 **ms shows partly features of another ms**
 1071 **** Degree of roundness and sphericity results separated by "/": two different degrees were mainly present ; measured**
 1072 **at 10x magnification.**

Slide	Depth [cm]	Aggregation		Voids				Micros *		Groundmass				Pedofeatures											
		wp	mp	hp	ds	spv	xpv	pl	vu	RS**	eff-related distribution		Micromass		Redoximorphic features		Translocation features								
										cm	cg	oe	ssee	chi	ce	color	u	gs	t	a	ro	li	cap	pen	ld
SMC I	0-10	x			w	(x)	x	(x)	fis /sgm	sub/su	x	(x)		(x)		gb	x		x	x	(x)	(x)	(x)		
SMC II	10-20		x		w		x	(x)	p gm	su	x					gb	x			x			x		
SMC III	30-40		(x)		w/m		x		wsl	sub/su	x			x	(x)	db	x		x		x		x		x
SMC IV	50-60		x		m/w		x	(x)	msl (hsp)	sub/su	x			x		db	x					x	(x)		x
SMC V	80-90		x		w		x	(x)	(fis) p gm	sub/su	x	(x)		x		db	x		(x)						
BB I	10-20		(x)		m		x	(x)	h-n sp	sub/su	(x)		x	(x)		gb	x	(x)				(x)			x
BB II	20-30		x		m		x	x	w-m sp (msl)	su-ro	(x)		x	(x)		gb	x	x		(x)	x		x		x
BB III	40-50			x	m/h		x	(x)	h-m sp (msl)	sub/ro			x			gb	x	x		x	x		x		x

Aggregation : hp = highly developed pedality, mp = moderately developed pedality, wp = weakly developed pedality
 : ds = degree of separation; h = highly separated, m = moderately separated, w = weakly separated
 : spv = simple packing voids, xpv = complex packing voids, pl = planes, vu = vughs

Voids : fis = fissure, sgm = single grain ms, p gm = pellicular grain ms, wsl = weakly separated lenticular ms, hsp = highly separated platy ms
 : msp = moderately separated platy ms, wsp = weakly separated platy ms, msl: moderately separated lenticular ms

Groundmass : sub = subrounded, su = subangular, ro = rounded, su-ro = subangular to rounded mineral grains
 and Sphericity** : cm = coarse monic, cg = chito-gefuric, oe = open equal enaulic, ssee = single spaced equal enaulic, chi = chitonic, ce = close enaulic
 (eff - R - Distr.) : t = typical, a = aggregate

Pedofeatures : u = undifferentiated, gs = granostriated
 color : gb = greyish brown, db = dark brown
 b - Fabric : t = typical, a = aggregate
 nodules : ro = redoximorphic hy poccoatings
 hp (hy poccoatings) : li = link cappings, cap = cappings, pen = pendent
 coatings : ld = loose discontinuous
 infillings

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1075 **Figures**

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1077 **Figure 1: Mainmap shows the regional setting of Ulu-Peninsula on James Ross Island, Maritime Antarctica. Black**
1078 **circles indicate the location of both study sites, Brandy Bay (BB) and St. Marta Cove (SMC). Sidemap 1-3 provide an**
1079 **additional overview over Antarctica, the Antarctic Peninsula Region and James Ross Island.**

1080 **Figure 2: Study sites and soil profiles on James Ross Island; a: St. Marta Cove (SMC). It is not covered with vegetation.**
1081 **A 90 cm deep soil profile was taken; b: soil profile St. Marta Cove (SMC). Scale of the tape measure is in cm; c: study**
1082 **site Brandy Bay (BB) is close to snowfield. It is not covered with vegetation. A 60cm soil profile was taken; d: Soil profile**
1083 **Brandy Bay (BB). Scale of the tape measure is in cm.**

1084 **Figure 3: Images of micromorphological features found at Brandy Bay (BB) and St. Marta Cove (SMC). Pictures were taking**
1085 **using plane polarized light (ppl) and crossed polarizers (xpl). (a) BB III: highly separated lenticular platy microstructure, platy**
1086 **aggregates are indicated by green dotted lines, lenticular ms is indicated by black dotted lines, 2.5x, ppl; (b) SMC IV:**
1087 **moderately separated lenticular platy microstructure, indicated by black dotted lines, 2.5x, ppl; (c) SMC I: coarse monic**
1088 **microstructure, 2.5x, ppl; (d) BB II: chitonic c/f-related distribution and thin link cappings (li) on quartz grains, 20x, ppl; (e)**
1089 **BB III: weathered rock fragment covered by silty capping (cap) and also showing a thick pendent (pen) consisting of silty**
1090 **material and mineral grains, 10x, ppl; (f) SMC I: strongly weathered sandstone fragment with former boundaries, indicated**
1091 **by red dotted line, still visible by capping (cap), 5x, ppl; (g) SMC I: weathered volcanic rock fragment with distinct pellicular**
1092 **alteration pattern, 5x, ppl; (h) BB II: weathered and broken volcanic rock fragment with internal volcanic glass and covered**
1093 **by a thin clay capping (cap),(110-120µm), 2.5x, ppl; (i) SMC I: weathered volcanic rock fragment with feldspar phenocrysts;**
1094 **covered by a dusty clay-silt capping (80-100 µm) (cap), 2.5x, ppl; (j) SMC I; usage of crossed polarizers makes it possible to**
1095 **tell external coating (cap) from altered internal material, border indicated by grey dotted line, 2.5x, xpl.**

1092 **Figure 4: Relative abundances of phyla and bacterial 16S rRNA qPCR gene abundances of soil profiles from Brandy**
1093 **Bay (BB) and St. Marta Cove (SMC) on James Ross Island, Antarctica. Triplicates are merged. Only phyla with a**
1094 **relative abundance of at least 5% at a given site are shown. The remaining phyla are summarized as “Others”.**

1095 **Figure 5: Heatmap based on the relative abundances of the observed operational taxonomic units (OTUs) in soil profiles**
1096 **from Brandy Bay (BB) and St. Marta Cove (SMC) on James Ross Island, Antarctica. Only OTUs with a relative**
1097 **abundance of at least 3% in a given sample were included. Samples as well as OTUs were clustered using average**
1098 **linkage hierarchical clustering.**

1099 **Figure 6: Canonical correlation analysis of the microbial composition of soil profiles from Brandy Bay (BB; black**
1100 **symbol) and St. Marta Cove (SMC; yello symbol) based on Bray-Curtis dissimilarities of the OTU data and its**
1101 **associated environmental parameters. If the Bonferroni corrected p-value was below 0.05, a given environmental**
1102 **parameter was included in the visualization. The amounts of chloride, sulfate, silt, Ca and TOC contents, and the Fe_d/Fe_e**
1103 **ratio explained 49.9% of the microbial community composition.**