1　**Title: Small-scale soil microbial community heterogeneity linked to landforms on**

2　**King George Island, maritime Antarctica**

3

4　**Running title: Landform influence on microbial biogeography**

5

6　Yumin Zhang[a], Lu Lu[a], Xulu Chang[a], Fan Jiang[a], Xiangdong Gao[b], Fang Peng[a#]

7

8　[a]China Center for Type Culture Collection (CCTCC), College of Life Sciences, Wuhan

9　University, Wuhan 430072, China

10　[b]College of Life Sciences, Wuhan University, Wuhan 430072, China

11

12　#Corresponding author. Email: pf-cctcc@whu.edu.cn; Tel: (+86) 02768752056; Fax:

13　(+86) 02768754982; Address: China Center for Type Culture Collection (CCTCC),

14　College of Life Sciences, Wuhan University, Bayi Road, Wuhan 430072, China.

15

ABSTRACT

We analysed soil-borne microbial (bacterial, archaeal, and fungal) communities around the Fildes Region of King George Island, maritime Antarctica, which were divided into two groups according to soil elemental compositions and environmental attributes (soil chemical parameters and vegetation conditions) located in Holocene raised beach and Tertiary volcanic stratigraphy. Prokaryotic communities of the two groups were well separated; they predominantly correlated with soil elemental compositions, and were secondly correlated with environmental attributes (e.g., soil pH, total organic carbon, NO_3^-, and vegetation coverage; Pearson test, r = 0.59 vs. 0.52, both P < 0.01). The relatively high abundance of P, S, Cl, and Br in Group 1 was likely due to landform uplift. Lithophile-elements (Si, Al, Ca, Sr, Ti, V, and Fe) correlated with prokaryotic communities in Group 2 may originate from weathering of Tertiary volcanic rock. The elements and nutrients accumulated during formation of different landforms influenced the development of soils, plant growth, and microbial communities, and resulted in small-scale spatially heterogeneous biological distributions. We propose that the geological evolution of the Fildes Region was crucial to its microbial community development.

IMPORTANCE

This current study analyzed soil-borne microbial communities around the Fildes Region of King George Island, maritime Antarctica, which were divided into two groups according to soil elemental compositions and environmental attributes. We

38   provide new evidence for the crucial influence of landforms on small-scale structures

39   and spatial heterogeneity of soil microbial communities.

40

41   **KEYWORDS**

42   Soil-borne microbial community, small-scale spatial heterogeneity, landform,

43   maritime Antarctica, pyrosequencing, phospholipid fatty acid

44

45   **Introduction**

46   Investigating microbial communities at different spatial scales, and the factors that

47   affect microorganism distributions, are fundamental aspects of microbial

48   biogeography (39, 42). In many terrestrial ecosystems, bacterial, fungal, and archaeal

49   communities are distributed along soil parameter gradients (e.g., temperature, pH,

50   water content, salinity, and nutrition; (35, 37, 48, 60, 71). Meanwhile, plants and

51   animals that depend on the soil ecosystem may also have significant influences on

52   microorganisms (22, 55, 75). It is therefore difficult to determine the most sensitive

53   factors influencing microbial communities. Nonetheless, in most distinct terrestrial

54   areas with special environments, distribution trends in the microbial community are

55   dominantly shaped by environmental factors that limit or prevent cell growth (33, 39).

56   Historical contingencies are also important for microbial community distribution on

57   spatial scales of one to tens of thousands of kilometres, but such effects may be

58   overwhelmed at small and intermediate scales (100m-1000km), including across

59   Antarctica (12, 42). Therefore, microbial spatial structures can be used to partly

60 reflect the external or intrinsic drivers of microbial population development and

61 activities (72).

62

63 The extreme conditions of Antarctica, such as low temperatures, low nutrient

64 availability, high UV radiation, and frequent freeze-thaw (14, 65), result in relatively

65 simple ecosystems. Hence, the relatively uncomplicated food-web structure of

66 Antarctic terrestrial habitats provides an appropriately manageable system to

67 investigate the drivers of soil microbial diversity and composition (70). Unsurprisingly,

68 spatial microbial community patterns have been observed here, ranging from

69 site-specific regions to large regional scale (11, 63, 69). However, to the best of our

70 knowledge the effects of historical contingency on microbial community distribution

71 at small scale spatial have not been observed yet.

72

73 In this study, we used Illumina Miseq pyrosequencing and the phospholipid fatty

74 acids (PLFA) method to survey the diversity and structure of prokaryotic and fungal

75 communities in 12 quadrat plots around the Fildes Region, King George Island. Fildes

76 is one of the largest ice-free regions in maritime Antarctica, and has higher

77 biodiversity than continental Antarctica. This typical small-scale spatial region

78 includes two Antarctic Special Protected Areas (ASPAs), covering approximately 30

79 km^2 of the Fildes Peninsula, Ardley Island, and adjacent islands (8). Nevertheless,

80 after the last glacial maximum, this region experienced multiple geologic and glacial

81 events, including deglaciation (8400~5500 BP), glacial re-advance (after 6000 BP),

4

82   Holocene glacio-isostatic and tectonic uplift during the glacial erosive phase, and

83   glacier retraction (30, 44). Glacial activities and past sea level changes were the key

84   drivers of landform and soil development across the Fildes region (5, 62), and their

85   effects on terrestrial microbial communities should not be ignored (59).

86

87   The 12 permanent quadrats analysed in our study have been established since 2013.

88   Their primary aim was to evaluate long-term ecosystem evolution according to

89   biomass and diversity under climate change conditions, and to build a

90   comprehensive research platform for multi-disciplinary study, including botany,

91   microbiology, ecology, and environmental science (68). For these purposes, all

92   selected quadrats must: (a) include Antarctic hairgrass (*Deschampsia antarctica*), the

93   only advanced plant discovered in the Fildes region, associated with moss and lichen;

94   (b) have stable soil and vegetation for long term monitoring; and (c) be protected

95   from human disturbance (mostly scientific explorers) and animal activity as much as

96   possible. Soil maturation and vegetation colonization takes a very long time under

97   unfavourable conditions, so the established quadrats represented natural and stable

98   habitats around the region. Recently, based on 454 pyrosequencing data, Wang et

99   al.(61) found that the diversity and structure of soil bacterial communities in four

100   sites of the Fildes region were significantly affected by pH, phosphate phosphorus,

101   organic carbon, and organic nitrogen. The relationships between microbial

102   communities, geological factors, and landform development have not been studied.

103   In this study, we attempt to answer three questions: (i) what is the microbial

104   community structure in this small-scale region of maritime Antarctica; (ii) which

105   factors affect the distribution of microbial communities; and (iii) do microbial

106   communities reflect different landform development in history. In order to explore

107   these issues, a number of soil chemical properties and vegetation attributes were

108   measured, representing conventional environmental factors; the soil element

109   composition was determined by X-ray fluorescence spectrometer, which is an

110   acceptable proxy for soil or sediment erosion and development in different

111   landforms (4, 28, 66) , and combining geological literature related to the Fildes region

112   to represent landform types. We aim to improve understanding of terrestrial

113   microbial communities in maritime Antarctic ice-free areas, and contribute to a new

114   perspective on small-scale microbial biogeography.

115

116   **Results**

117   *Soil elemental compositions and environmental attributes of quadrats*

118   A total of 20 elements in the sample soils were detected by X-Ray fluorescence

119   spectrometer, and 11 environmental attributes were measured (Table S1). Principle

120   component analysis with normalized whole soil elemental compositions and

121   environmental attribute data showed that 36 samples were well separated by the

122   original point of the PC1 coordinate axis (Fig.1a). Hence the quadrat plots could be

123   divided into two groups reflecting different soil types and environmental conditions,

124   in which Group 1 included quadrat plots Q2, Q3, Q6, and Q7, and Group 2 included

125   all other quadrat plots (Q1, Q4, Q5, Q9, Q10, Q11, Q12, and Q13). The heatmap

126   cluster analysis supported the grouping suggested by whole element and

127   environmental data (Fig. 1b).

128

129   The soil element profile revealed that lithophile elements (Si, Al, Ca, Mg, Fe)

130   occupied the major portions. Twelve elements (Al, Ca, Cu, Fe, K, Mg, Mn, Si, Sr, Ti, V,

131   and Zn) were more abundant in Group 2, and four elements (P, S, Cl, and Br) were

132   significantly more abundant in Group 1 (Wilcoxon test, P < 0.05, Table S1).

133   PERMANOVA analysis revealed a highly significant difference in soil element

134   composition between the two sample groups (*Pseudo*-F = 17.74, P < 0.01). Pairwise

135   correlative comparisons between elements demonstrated that P, S, Cl, and Br were

136   positively correlated with each other, and negatively correlated with Mg, Al, Si, Ca,

137   Mn, Zn, Sr, V, Fe and Ti, which also had positive correlations with each other (Fig. S1).

138   A significant difference was also observed in the environmental attributes between

139   Group 1 and Group 2 (PERMANOVA test, *Pseudo*-F = 15.17, P < 0.01), consisting of

140   lower soil pH and higher total organic carbon (TOC, NH_4^+, NO_3^-) and moisture

141   contents in Group 1. In addition, vegetation properties such as hairgrass coverage

142   (DAC) and total vegetation coverage (VC) were also higher in Group 1 plots (Wilcoxon

143   test, P < 0.05, Table S1).

144

145   *Diversity and composition of microbial communities in quadrats*

146   After sequence-quality filtering, we obtained a total of 2,389,662 high-quality

147   bacterial 16S rRNA gene reads, 1,423,619 archaea 16S rRNA gene reads, and

7

148   1,953,908 ITS reads. These reads constituted 98,887, 49,000, and 8,464 operational

149   taxonomic units (OTUs) at a 0.03 discrepancy (97% identity) for bacterial, archaeal,

150   and fungal taxa, respectively. The OTUs diversities of Shannon, Chao 1, and ACE index

151   of bacteria, archaea, and fungi did not differ between Group 1 and Group 2

152   (Wilcoxon test, P > 0.05, Table S2). For bacteria, 20 phyla and some unidentified

153   bacteria (0~0.8%) were detected, and the OTU sequences of most quadrat soils were

154   dominated by *Actinobacteria* (24.2%), *Acidobacteria* (14.7%), *Proteobacteria* (15.1%),

155   *Chloroflexi* (12.3%), and *Gemmatimonadetes* (7.2%) (Fig. S2). For archaea, a

156   number of OTU sequences (74.6%, maximum) were not assigned to any taxon; the

157   remainder was dominated by *Crenarchaeota* (94.5%) and *Euryarchaeota* (5.5%), and

158   the dominant classes of the phyla were *Thaumarchaeota* and *Thermoplasmata,*

159   respectively. For fungi, six phyla were detected, and all quadrat plot soils were

160   dominated by *Ascomycota* (69.1%), *Basidiomycota* (17.6%), and *Zygomycota* (4.5%).

161   The percentage of unassigned OTUs and unidentified fungi were 6.8%–64.3% and

162   0.4%–7.7%, respectively. PERMANOVA tests of Bray-Curtis distances revealed

163   significant differences between the two groups for prokaryotic 16S rRNA genes

164   (integrated data normalized by the bacterial and archaeal OTU data set, Pseudo-F =

165   3.1, P = 0.0002), but not for fungal ITS genes (Pseudo-F = 1.3, P = 0.196). This was

166   consistent with the grouping result of nonmetric multidimensional scaling (NMDS)

167   analysis with the bacterial and archaeal OTU dataset (Fig. 2).

168

169   *Links among microbial composition, soil elemental composition, and environmental*

170   *attributes*

171   The prokaryotic OTU composition showed a strong and significant correlation with

172   soil elements ($r = 0.59$, $P < 0.01$, Pearson test), and a less but still significant

173   correlation with environmental attributes ($r = 0.52$, $P < 0.01$, Pearson test; Fig. 3a).

174   Fungal community composition did not show strong correlations with either soil

175   elements or environmental attributes ($P > 0.05$; Fig. 3b). The soil elements and

176   environmental attributes in canonical correspondence analysis (CCA) were selected

177   by variation inflation test (see the Methods for details). CCA results for bacterial and

178   archaeal community composition and soil elements, with significant models at the

179   confidence level (both $P < 0.01$), indicated that the 11 soil elements are important

180   factors controlling bacterial and archaeal community structures, and explain 60.0%

181   and 47.3% of their variations, respectively (Fig. 4a and b).

182

183   Among these elements, P and Br were important elements controlling microbial

184   community structures in Group 1, and other lithophile and metal elements

185   controlled Group 2. The importance of these soil elements was verified by a Monte

186   Carlo test ($P < 0.05$, 999 permutations) with prokaryotic community data comprising

187   bacterial and archaeal community compositions (Table 1). For fungal communities,

188   CCA analysis showed that the two groups were not well separated from the others,

189   and the model was not significant within the confidence level (P > 0.05). Only 37.3%

190   of fungal community variations could be explained by the 11 soil elements (Fig. 4c).

191   Considering the relationships between microbial communities and environmental

9

192   attributes, the same analysis was made by CCA (Fig. 4d, e, and f). The models were

193   significant between both bacterial ($P < 0.01$, 56.2% explained) and archaeal ($P < 0.05$,

194   43.6% explained) community structures and environmental attributes. The Monte

195   Carlo test (999) revealed that total organic C (TOC), soil pH, moisture, site altitude,

196   hairgrass coverage (DAC), and total vegetation coverage (VC) showed strong effects

197   on prokaryotic communities. For fungal communities, the model was not significant

198   within the confidence level ($P > 0.05$, 34.3% explained); however, soil pH, site

199   altitude, moss species amount (MS), and lichen species amount (LS) were the

200   environmental attributes affecting fungal community composition (Monte Carlo test,

201    $P < 0.05$, 999 permutations; Table 1). For the mantel test of microbial community

202   structures, including all factors investigated in this study, see Table S3.

203

204   *Microbial biomass and microbial diversity determined by the phospholipid fatty acids*

205   *(PLFA) method*

206   The total amounts of PLFA (totPLFA) of Group 1 were significantly higher than those

207   of Group 2 (Wilcoxon test, $P < 0.05$; Fig. S3). CCA analysis of the individual relative

208   concentration (mol%) of the 45 most common PLFAs showed that, on the whole, the

209   11 soil elements and the 11 environmental attributes were all important factors

210   controlling soil PLFA patterns (Fig. S4, $P < 0.01$), with 47.5% and 47.0% of the

211   variations explained, respectively. Among these factors, each of the 11 elements, and

212   pH, moisture, total organic C (TOC), *Deschampsia antarctica* coverage (DOC), and

213   total vegetation coverage (VC) of the environmental attributes had significant effects

214   on soil PLFA composition (Table 1). Microorganism categories including bacteria,

215   fungi and protozoa were classified by indictor PLFAs according to microbial

216   identification systems (MIDI). The relative abundance of AM fungi, actinomycetes,

217   and anaerobes were higher in Group 1, and Gram-negative bacteria was higher in

218   Group 2 (Fig. S5; Welch's t-test, two-sided, $P < 0.05$).

219

220   *Differences of microbial community composition between the two groups*

221   In our analysis, the classified mode of the Random forests machine learning

222   technique (9, 16) could be accepted if the ratio of the baseline error to the observed

223   error was greater than 2, and we considered an OTU to be highly predictive if its

224   importance score was at least 0.001. For bacteria, random forest analysis revealed

225   that 58 OTUs distinguished the two groups, *Acidobacteria* were overrepresented in

226   Group 1, and the OTUs assigned to the *Thermoleophilia* class of the phylum

227   *Actinobacteria*, and genus *Geobacillus* of phylum *Firmicutes,* were overrepresented

228   in Group 2. For archaea, 38 OTUs distinguished the two groups, except for 12 OTUs

229   with no assigned taxa. Some 11 OTUs were overrepresented in Group 1 and 17 OTUs

230   were overrepresented in Group 2; all were assigned to genus *Candidatus*

231   *Nitrososphaera* of phylum *Crenarchaeota*. As the ratio of the baseline error to the

232   observed error of the random forest analysis with fungal OTUs was less than 2, we

233   considered that the non-obvious classified result suggested that there was no

234   credible difference in fungal community composition between the two groups (Table

235   S4).

236

Discussion

238 The prokaryotic community composition of the quadrats can be divided into two

239 groups that correlate with soil element compositions and environmental attributes.

240 Interestingly, a published geologic map of the Fildes region and previously reported

241 literature (38, 44) showed that the quadrats in Group 2 were located in Tertiary

242 volcanic stratigraphy, and those in Group 1 were found on a Holocene raised beach

243 (Fig. 5). We suggest that there is a potential relationship between microbial

244 communities and the development of landforms at this small spatial scale.

245

246 In this area, a series of geological events, including volcanic activity, glacial erosion

247 and retraction, isostatic uplift, and sea level change, created rich landform types.

248 According to geomorphological and sedimentary evidence, relative sea level (RSL)

249 gradually fell to < 14.5 m between 7000 and 4750 cal a BP as a consequence of

250 isostatic uplift in response to regional deglaciation (30, 62). During landform

251 formation, rich marine elements and nutrients were transferred to the land (5, 44).

252 Moreover, from approximately 2500 years ago, mammals, especially penguins, began

253 to colonize the newly uplifted beaches until at least ~500 years ago when the raised

254 beaches were abandoned (according to chronological research of abandoned

255 rookeries on King George Island (54)). These abandoned penguin rookeries are

256 indicators of Holocene paleoclimate and also accumulated rich nutrients during the

257 period (3). These input elements, nutrients, and marine microorganisms clearly

258   promoted the development of soil and plant growth, and influenced patterns of

259   microbial community formation.

260

261   Soil elemental profiles can be seen as proxy indicators of soil types and landforms

262   with strong soil-landform relationships (20, 34). In previous studies, soil compositions

263   tested by X-ray fluorescence spectrometer have been revealed to be the key factors

264   for the distribution of bacterial and fungal communities in some field sites,

265   sediments, glacier forefields, and deserts (27, 29, 43, 52). In this study, the CCA

266   analysis and similarity test showed that both environmental attributes and soil

267   element compositions could influence the microbial structure and biomass. However,

268   compared with environmental attributes, the relationship between soil element

269   composition and prokaryotic community was stronger (Fig 4; Fig 5). Mantel analysis

270   revealed that the relative abundance of almost every element was important for

271   shaping prokaryotic compositions (Table S3). The quadrat plots located on the

272   Holocene raised beach landform showed relatively high abundances of P, S, Cl, and Br,

273   which were more correlated to marine environments and organisms. These elements

274   are readily absorbed by vegetation and microorganisms, and presumably resulted in

275   the development of microbial community structures in Group 1 (CCA analysis; Fig. 4).

276   The accumulation of elements P and S may represent marine input but also mammal

277   and bird excrement that accumulated in these raised beaches during the early stage

278   of uplifted landform formation (54). Meanwhile, the halogen elements Cl and Br in

279   island coastal soil likely derived mostly from seawater (49).

280

281 Conversely, the bacterial and archaeal community compositions of Group 2 were

282 more correlated with lithophile-elements (Si, Al, Ca, Sr, Ti, V, and Fe, CCA analysis),

283 and the landforms were almost completely isolated from the external environment

284 until the icecap retreated ~11000–7500 cal a BP (62). This suggests that soil in

285 quadrats located in tertiary volcanic stratigraphy mainly developed from the

286 chemical and biological weathering of volcanic rock generated by Tertiary volcanism,

287 and underwent paraglacial and periglacial processes. That may explain the lower soil

288 biomass, nutrition, and vegetation coverage, as compared with Group 1; the limited

289 nutrient input distinguished the prokaryote community composition from that of the

290 nutrient-rich soil of Group 1. Therefore, we believe that the element composition of

291 the soil associated with these landforms reveals geological background and historic

292 effects.

293

294 In Group 1, the soil contained high contents of TOC, NH_4^+, NO_3^-, and vegetation

295 coverage, which also correlates with prokaryotic community. The relatively low pH

296 values (Table S1) may be a result of higher vegetation coverage with more humus and

297 fulvic acids produced by moss and lichen (21). As the rich nutrients and elements

298 transferred from Holocene raised beach marine environments could promote soil

299 development and plant growth, these environmental attributes seem to be a

300 secondary factor affecting the prokaryotic community when compared to soil

301 element compositions. Unlike Group 1, very small amounts of nutrients in the soil

302   samples of Group 2 were more likely caused by current precipitation, snowfall, and

303   animal activity. In keeping with reported studies (19, 23, 35), pH is one of the most

304   influential factors affecting the distribution of microbial communities in this study.

305

306   Interestingly, both prokaryotes and fungi communities were significantly correlated

307   to the altitude of the sample location. Despite the slightly different altitudes (ranging

308   from 11–56 m), they do not lead to significant changes in temperature, oxygen

309   content, etc., which seems to suggest that geological uplift had an impact on

310   microbial communities. In addition, moss and lichen species were significantly

311   associated with fungal communities. It was previously reported that some fungal

312   species coexist with moss and lichen in Antarctica (36, 58). We also noted that the

313   soil element compositions and environmental attributes of ancient landforms

314   investigated in our study were relatively stable, while those of younger landforms

315   were more volatile (from Euclidean distances computed between samples from the

316   PCA analysis in Fig 1a, Table S5). This suggests that the quadrat plots of Group 1 may

317   be in an unstable new geological layer within a transboundary ecological stage from

318   ocean to land, and disturbance from the new terrestrial environment may increase

319   the heterogeneity of the geomorphic ecology.

320

321   As the quadrats in our study all had hairgrass growth, vegetation may be one of the

322   main causes of this difference. Thus, our results were similar to bacterial community

323   compositions in other vegetated parts of Antarctica (36, 58), with relatively high

324   abundances of *Chloroflexi* and *Gemmatimonadetes,* which have strong reported

325   relationships with plants (2, 10, 17). Previous studies of Antarctic archaeal

326   communities were mostly concentrated in marine and lake environments (13, 18, 31,

327   32, 45), and in this study, only two archaeal phyla (*Crenarchaeota* and *Euryarchaeota*)

328   were detected, with *Crenarchaeota* representing the overwhelming majority (> 90%)

329   of archaeal communities. This was consistent with other terrestrial archaeal

330   structures of Antarctica derived using other investigated methods (e.g., clone

331   libraries of rRNA genes and microarray (1, 70).

332

333   Random forest analysis revealed that OTUs belonging to *Alphaproteobacteria*,

334   *Acidobacteria,* and *Bacteroidetes* were mostly overrepresented in Group 1. These

335   phyla have shown positive correlations with vegetation and the rhizosphere in

336   farmland, arctic glacier moraines, and the Brazilian Antarctic Station (41, 55, 57). The

337   results of our study showed different patterns at the family level, with

338   *Acidobacteriaceae*, *Koribacteraceae*, *Chitinophagaceae,* and *Rhodospirillaceae* the

339   most overrepresented families in Group 1. The family level differences from our

340   study could be due to the locations of sampling points and the diverse sequencing

341   methods. Conversely, in Group 2, the major overrepresented OTUs were class

342   *Thermoleophilia* of phylum *Actinobacteria*. *Thermoleophilia* is a newly proposed class

343   of phylum *Actinobacteria* that was created from the splitting of *Rubrobacteridae* (40),

344   and its ecological position is not well understood. However, *Thermoleophilia* is

345   abundant in deserts and glacier forelands (15, 74), and some isolated parts could be

346   cultured in low nutritional media during long incubation periods. Thus, it is

347   reasonable that this class is found in the quadrats located in volcanic stratigraphy

348   with high proportions of lithospheric elements and low nutrition conditions. In

349   addition, we also found that five OTU sequences affiliated to *Flavobacteriaceae*

350   extracted from Group 1 were clustered in marine clades, and no marine clade OTUs

351   of *Flavobacteriaceae* were found in Group 2 (Fig. S6). Members of the family

352   *Flavobacteriaceae* are among the most abundant picoplankton in coastal and polar

353   oceans, and a number of genera have potential evolutionary sources from the ocean

354   (7). Regarding genus *Candidatus Nitrososphaera,* the vital ammonia-oxidizing

355   archaea (51, 53) was the overrepresented archaeal OTU in both groups. The

356   uncultivable species *s_Ca.* N. SCA1170 was a major genus in Group 2 but did not

357   appear in Group 1. This, along with evidence from NMDS analysis, implies that the

358   two different landforms have diverse archaeal communities. Those suggested that

359   spatial constraints for microorganisms also occur at small spatial scales.

360

361   The importance of geological factors, such as the landform and lithology, on

362   microbial structure is less well understood (59). Locations with distinct geologic

363   factors generally exhibit geographical isolation; hence, they are mostly distributed at

364   large and global scales. Limited research has shown that different landforms and soil

365   profiles are also important drivers of bacterial diversity at the regional scale (>1000

366   km distance), and their impacts are more significant than contemporary

367   environmental factors (25, 47). Interestingly, we found that, on such a small spatial

17

368   scale, prokaryotic communities also showed a landform-governed distribution trend,

369   and the microbial community structure is expected to be an indicator of the

370   formation of the landform. The role of geological evolution in microbial distribution

371   can be highlighted in this study because: (i) a clear effect of the geological evolution

372   of the Fildes region in maritime Antarctica. Glacial activity, sea level changes, and

373   tectonic uplift due to climate change after LGM have all resulted in landform

374   heterogeneity at a small spatial scale; (ii) seasonal freezing-thawing cycles in the area

375   have enhanced soil development, and promoted soil particle and nutrition migration

376   to upper and surface soil layers; and (iii) the low activity of microorganisms under the

377   cold climate, and less human disturbance where the quadrats were established,

378   maintained relatively stable microbial community diversity for a long time after the

379   geological changes. However, in contrast to other research, we did not attempt to

380   classify the common environmental factors measured in this study as 'contemporary

381   environmental' factors because those representing soil nutritional conditions were

382   considered to be the consequence of landform development (44, 54), especially in

383   Group 1. In our study, the environmental factors had strong influences on bacterial

384   and archaeal community structures. Nonetheless, these were likely to play

385   subsequently important roles in the distribution of microbial communities,

386   predominantly driven by landforms and soil element compositions.

387

388   In conclusion, this study provides evidence for the influence of geological evolution

389   on the small-scale distribution of microbial communities. As a result, microbial

18

390   community structure is proposed as an indicator of the two different landforms in

391   the Fildes region, King George Island. In addition, other locations in Antarctica

392   experience the same type of glacial activity and isostatic uplift as the coastal ice-free

393   areas around King George Island, maritime Antarctica, and Prince Charles Mountains

394   area, East Antarctica (64), implying that microbial communities may also be diverse

395   and influenced by different geological evolution events at small to moderate spatial

396   scales in these areas. Continued research already, in progress, will verify whether

397   microbial communities can be used as indicators of different landforms in other,

398   similar geological areas in maritime Antarctica. This will contribute to finding

399   different microbial communities in limited spatial regions based on geological

400   research, and will examine different types of geological heterogeneity according to

401   microbial communities.

402

403   **Materials and Methods**

404   *Quadrat plot description, soil sampling, and sample preparation*

405   The Fildes region is the largest ice-free area on King George Island, with a humid and

406   relatively mild sub-Antarctic maritime climate. The mean annual temperature and

407   precipitation are −2.4ºC and over 500 mm, respectively (26). The 12 permanent

408   quadrat plots (1.5 m × 1.0 m each) investigated in this study had been established on

409   Fildes Peninsula and Ardley Island between 2013 and 2015. For plot characteristics,

410   please see the Introduction for details. Each quadrat plot was fenced to minimize

411   disturbance. GPS coordinates, vegetation characteristics, and the landscape of

412   quadrat locations are shown in Table 2 and Fig. S7. The distance between quadrat

413   plots ranges from approximately 1.6 to 8.2 km. Sampling occurred during China's

414   33rd Antarctic expedition in January 2017. Soils were sampled from the A-horizon (10

415   cm), at an internal distance of approximately 3–5 m, in triplicate around each

416   quadrat plot. Soil samples collected for each replicate were taken from five soil cores

417   (5 cm diameter) and mixed thoroughly. A total of 36 soil samples were placed in

418   sterile plastic bags, and soil DNA was extracted within 2 h in the laboratory of the

419   Great Wall Station. The remaining soils were stored in the freezer until further soil

420   physic-chemical property analysis.

421

422   *DNA extraction, PCR, pyrosequencing, and pyrosequencing data treatment*

423   Genomic DNA was extracted using a PowerSoil DNA Isolation Kit (MoBio, Carlsbad,

424   CA, USA) according to the manufacturer's instructions. Duplicate DNA extraction was

425   performed for each sampling plot, and all duplicated DNA products were pooled to

426   reduce potential DNA extraction bias. Afterwards, DNA concentration was measured

427   by UV spectrophotometer (Eppendorf, Bio Photometer), and its molecular size was

428   estimated by 0.8% agarose gel electrophoresis. Details of pyrosequencing and

429   pyrosequencing data treatment are described in Appendix S1. These sequence data

430   have been submitted to the DDBJ/EMBL/GenBank databases (SRA) under accession

431   no. SRP132288, accession no.SRP132345 and accession no.SRP132350.

432

433   *PLFA analysis*

434   Phospholipid fatty acids (PLFAs) from soil samples were extracted, fractionated,

435   quantified, and analysed using the protocol described by (6). In brief, 2.0 g of soil (dry

436   weight) was extracted with a chloroform-methanol-citrate buffer mixture (1:2:0.8)

437   and fractionated into neutral lipids, glycolipids, and phospholipids on a silicic acid

438   column (Agilent Technologies, Sillic Box, CA, USA). Phospholipids were subjected to

439   mild alkaline methanolysis, after separating out fatty acid methyl esters on an Agilent

440   6890N gas chromatograph equipped with a flame ionization detector and an HP-1

441   Ultra 2 capillary column (Agilent Technologies, Santa Clara, CA, USA). Peak areas were

442   quantified by adding methyl non-adecanoate fatty acid (C19:0) (Sigma) as an internal

443   standard. The fatty acid methyl esters were prepared according to the MIDI protocol

444   and analysed using the MIDI Sherlock Microbial Identification System (MIDI, Newark,

445   DE). The fatty acids i14:0, i15:0, a15:0, i16:0, a16:0, i17:0, and a17:0 represented

446   gram-positive bacteria, 16:1ω9c, cy17:0, 18:1ω5c, 18:1ω7c, and cy19:0 represented

447   gram-negative bacteria, 10Me16:0 (24), 10Me17:0, and 10Me18:0 represented

448   Actinomycetea (73), branched monoenoic and mid-branched saturated fatty acid

449   PLFAs represented anaerobic microorganisms (76), and 16:1ω5 represented AM fungi

450   (46). PLFAs were categorized and calculated in the MIDI Sherlock Microbial

451   Identification System (MIDI, Newark, DE).

452

453   *Soil element composition determined by X-ray fluorescence spectrometer*

454   Soil samples were dried at 105ºC for 6 h and then ground into powder. The soil

455   powder was pressed in a 45 mm bore steel die under an approximately 20 t hydraulic

456   press. Every soil sample formed a stable soil pie of 45 mm diameter and 10 mm

457   height. These pies were generally analysed within a few hours. The elements within

458   soil samples were determined by X-ray fluorescence spectrometry (Bruker AXS,

459   Germany) using a standardless quantitative analysis method (29). We removed poor

460   quality elemental signals that rarely appeared (< 0.01%), generally in only one or two

461   samples.

462

463   *Soil parameters and vegetation attribute measurements*

464   Soil temperature was measured by a plug-type thermometer (ZD Instrument, China)

465   at depths of 15 cm during soil sampling. Soil pH was measured by adding 10 ml of

466   distilled water to 5 g of soil, and recording pH by a pH electrode (Mettler-Toledo,

467   Switzerland). Soil moisture was determined as the gravimetric weight loss after

468   drying the soil at 105ºC until reaching a constant weight. Analysis of total organic

469   carbon was performed using a TOC analyser (vario TOC, Elementar, Germany). To

470   measure NH_4^+ and NO_3^-, 10 g of soil was suspended in 50 ml of 2 mol/L KCl

471   solution and shaken at 25ºC for 1 h. Then, the soil solution mixture was centrifuged

472   for 5 min in 3000 g. Subsequently, clear supernatant was passed through a filter of

473   0.45 μm (Millipore, type GP), and analysed using a continuous flowing analyser

474   (FIAstar 5000, Foss, Denmark). Each quadrat of 1 × 1 m was selected to measure

475   vegetation attributes including moss species number (MS), lichen species number

476   (LS), hairgrass (*Deschampsia antarctica*) coverage (DAC), and total vegetation

477   coverage (VC) according to previous protocols (68).

478

479 *Data statistical analyses*

480 For estimating bacterial, archaeal, and fungal diversity, Operational Taxonomic Unit

481 (OTU) analysis including the Shannon, Chao1, and ACE indices was performed using

482 the Mothur v. 1.30.2 software package (50). The relationships between soil

483 elemental compositions and environmental attributes in the 32 soil samples were

484 analysed by principal component analysis and hierarchical clustering heatmap

485 analysis using the R v. 3.3.1 statistical software. The Wilcoxon test was performed for

486 the soil elements and environments to determine the level of significance with a

487 two-sided hypothesis using the Statistical Package for the Social Sciences software

488 (SPSS). Significant differences in soil elemental compositions, environmental

489 attributes, and microbial community structures between groups were determined by

490 permutational multivariate analysis of variance (PERMANOVA) on 999 permutations

491 of residuals under a reduced model using the R v. 3.3.1 statistical software. The

492 Bray-Curtis distance was used to obtain dissimilarity matrices in the PERMANOVA

493 test for microbial OTU data. The similarity test, Mantel test, and Canonical

494 correspondence analysis (CCA) were used to evaluate the linkages between microbial

495 community structures (general levels) and soil elemental compositions and

496 environmental attributes with the Vegan package (v. 2.4-1) in R v. 3.3.1 according to

497 the method described by Yang et al (67). Variation inflation factors were used to

498 select factors in CCA modelling, of which the variance of canonical coefficients was

499 not inflated by the presence of correlations with other factors, so that soil elements

23

500   and environmental attributes were removed if the variation inflation factor was

501   larger than 20. Variation partitioning analysis resulted in 11 soil elements (Si, Ca, Zn,

502   Fe, Al, Mn, V, Ti, Sr, P, Br) and 10 environmental attributes. The effect of factors on

503   microbial community structures and the PLFA profile was estimated by a Monte Carlo

504   permutation test (999 permutation). Differences in microbial categories marked by

505   PLFA was determined using Welch's t-test (two-sided) using the STAMP software (v.

506   2.1.3) package.

507

508   **Acknowledgements**

References:

515

516   1.   Ayton, J., J. Aislabie, G. M. Barker, D. Saul, and S. Turner. 2010. Crenarchaeota

517   affiliated with group 1.1b are prevalent in coastal mineral soils of the Ross Sea region

518   of Antarctica. Environ Microbiol 12:689-703.

519   2.   Banning, N. C., D. B. Gleeson, A. H. Grigg, C. D. Grant, G. L. Andersen, E. L. Brodie,

520   and D. V. Murphy. 2011. Soil Microbial Community Successional Patterns during

521   Forest Ecosystem Restoration. Appl Environ Microb 77:6158-6164.

522   3.   Baroni, C., and G. Orombelli. 1994. Abandoned penguin rookeries as holocene

523   paleoclimatic indicators in antarctica. Geology 22:23-26.

524   4.   Battaglia, S., L. Leoni, and F. Sartori. 2003. Mineralogical and grain size

525   composition of clays developing calanchi and biancane erosional landforms.

526   Geomorphology 49:153-170.

527   5.   Boelter, M. 2011. Soil development and soil biology on King George Island,

528   Maritime Antarctic. Pol Polar Res 32:105-116.

529   6.   Bossio, D. A., and K. M. Scow. 1995. Impact of carbon and flooding on the

530   metabolic diversity of microbial communities in soils. Appl Environ Microb

531   61:4043-4050.

532   7.   Bowman, J. P. 2006. The Marine Clade of the Family Flavobacteriaceae: The

533   Genera Aequorivita, Arenibacter, Cellulophaga, Croceibacter, Formosa, Gelidibacter,

534   Gillisia,   Maribacter,   Mesonia,   Muricauda,   Polaribacter,   Psychroflexus,

535   Psychroserpens,   Robiginitalea,   Salegentibacter,   Tenacibaculum,   Ulvibacter,

536   Vitellibacter and Zobellia, p. 677-694. *In* M. Dworkin, S. Falkow, E. Rosenberg, K.

537   Schleifer, and E. Stackebrandt (ed.), The Prokaryotes: Volume 7: Proteobacteria:

538   Delta, Epsilon Subclass. Springer New York, New York, NY.

539   8.   Braun, C., O. Mustafa, A. Nordt, S. Pfeiffer, and H. Peter. 2012. Environmental

540   monitoring and management proposals for the Fildes Region, King George Island,

541   Antarctica. Polar Res 31.

542   9.   Breiman, L. 2001. Random forests. Mach Learn 45:5-32.

543   10. Castro, H. F., A. T. Classen, E. E. Austin, R. J. Norby, and C. W. Schadt. 2010. Soil

544   Microbial Community Responses to Multiple Experimental Climate Change Drivers.

545   Appl Environ Microb 76:999-1007.

546   11. Chong, C. W., D. A. Pearce, P. Convey, G. Y. A. Tan, R. C. S. Wong, and I. K. P. Tan.

547   2010. High levels of spatial heterogeneity in the biodiversity of soil prokaryotes on

548   Signy Island, Antarctica. Soil Biol Biochem 42:601-610.

549   12. Chong, C., D. A. Pearce, and P. Convey. 2015. Emerging spatial patterns in

550   Antarctic prokaryotes. Front Microbiol 6.

551   13. Church, M. J., E. F. Delong, H. W. Ducklow, M. B. Karner, C. M. Preston, and D. M.

552   Karl. 2003. Abundance and distribution of planktonic Archaea and Bacteria in the

553   waters west of the Antarctic Peninsula. Limnol Oceanogr 48:1893-1902.

554   14. Convey, P. 1996. The influence of environmental characteristics on life history

555   attributes of Antarctic terrestrial biota. Biological reviews of the cambridge

556   philosophical society 71:191-225.

557   15. Crits-Christoph, A., C. K. Robinson, T. Barnum, W. F. Fricke, A. F. Davila, B.

558   Jedynak, C. P. Mckay, and J. Diruggiero. 2013. Colonization patterns of soil microbial

559   communities in the Atacama Desert. Microbiome 1.

560   16. Cutler, D. R., Jr. T. C. Edwards, K. H. Beard, A. Cutler, and K. T. Hess. 2007.

561   Random forests for classification in ecology. Ecology 88:2783-2792.

562   17. Debruyn, J. M., L. T. Nixon, M. N. Fawaz, A. M. Johnson, and M. Radosevich.

563   2011. Global Biogeography and Quantitative Seasonal Dynamics of

564   Gemmatimonadetes in Soil. Appl Environ Microb 77:6295-6300.

565   18. Delong, E. F., K. Y. Wu, B. B. Prezelin, and R. Jovine. 1994. High abundance of

566   archaea in antarctic marine picoplankton. Nature 371:695-697.

567   19. Dumbrell, A. J., M. Nelson, T. Helgason, C. Dytham, and A. H. Fitter. 2010.

568   Relative roles of niche and neutral processes in structuring a soil microbial

569   community. Isme J 4:337-345.

570   20. Evans, I. S. 2012. Geomorphometry and landform mapping: What is a landform?

571   Geomorphology 137:94-106.

572   21. Fabiszewski, J., and B. Wojtun. 1993. The maritime Antarctic coastal ecosystem

573   of Admiralty Bay.

574   22. Falush, D., T. Wirth, B. Linz, J. K. Pritchard, M. Stephens, M. Kidd, M. J. Blaser, D.

575   Y. Graham, S. Vacher, G. I. Perez-Perez, Y. Yamaoka, F. Megraud, K. Otto, U. Reichard,

576   E. Katzowitsch, X. Y. Wang, M. Achtman, and S. Suerbaum. 2003. Traces of human

577   migrations in Helicobacter pylori populations. Science 299:1582-1585.

578   23. Fierer, N., and R. B. Jackson. 2006. The diversity and biogeography of soil

579   bacterial communities. P Natl Acad Sci Usa 103:626-631.

580   24. Frostegard, A., and E. Baath. 1996. The use of phospholipid fatty acid analysis to

581   estimate bacterial and fungal biomass in soil. Biol Fert Soils 22:59-65.

582   25. Ge, Y., J. He, Y. Zhu, J. Zhang, Z. Xu, L. Zhang, and Y. Zheng. 2008. Differences in

583   soil bacterial diversity: driven by contemporary disturbances or historical

584   contingencies? Isme J 2:254-264.

585   26. Gerighausen, U., K. Brautigam, O. Mustafa, and H. U. Peter. 2003. Expansion of

586   vascular plants on an Antarctic island a consequence of climate change? p. 79-83. *In*

587   A. Huiskes, W. Gieskes, J. Rozema, R. Schorno, S. M. VanDerVies, and W. J. Wolff

588   (ed.).

589   27. Goncalves, V. N., C. L. Cantrell, D. E. Wedge, M. C. Ferreira, M. A. Soares, M. R.

590   Jacob, F. S. Oliveira, D. Galante, F. Rodrigues, T. M. A. Alves, C. L. Zani, P. A. S. Junior,

591   S. Murta, A. J. Romanha, E. C. Barbosa, E. G. Kroon, J. G. Oliveira, B. Gomez-Silva, A.

592   Galetovic, C. A. Rosa, and L. H. Rosa. 2016. Fungi associated with rocks of the

593   Atacama Desert: taxonomy, distribution, diversity, ecology and bioprospection for

594   bioactive compounds. Environ Microbiol 18:232-245.

595   28. Guilizzoni, P., A. Marchetto, A. Lami, A. Brauer, L. Vigliotti, S. Musazzi, L. Langone,

596   M. Manca, F. Lucchini, N. Calanchi, E. Dinelli, and A. Mordenti. 2006. Records of

597   environmental and climatic changes during the late Holocene from Svalbard:

598   palaeolimnology of Kongressvatnet. J Paleolimnol 36:325-351.

599   29. Handley, K. M., C. Boothman, R. A. Mills, R. D. Pancost, and J. R. Lloyd. 2010.

600   Functional diversity of bacteria in a ferruginous hydrothermal sediment. Isme J

601   4:1193-1205.

602   30. Ingolfsson, O., C. Hjort, P. A. Berkman, S. Bjorck, E. Colhoun, I. D. Goodwin, B.

603   Hall, K. Hirakawa, M. Melles, P. Moller, and M. L. Prentice. 1998. Antarctic glacial

604   history since the Last Glacial Maximum: an overview of the record on land. Antarct

605   Sci 10:326-344.

606   31. Kalanetra, K. M., N. Bano, and J. T. Hollibaugh. 2009. Ammonia-oxidizing

607   Archaea in the Arctic Ocean and Antarctic coastal waters. Environ Microbiol

608   11:2434-2445.

609   32. Karr, E. A., J. M. Ng, S. M. Belchik, W. M. Sattley, M. T. Madigan, and L. A.

610   Achenbach. 2006. Biodiversity of methanogenic and other Archaea in the

611   permanently frozen Lake Fryxell, Antarctica. Appl Environ Microb 72:1663-1666.

612   33. Kuang, J., L. Huang, L. Chen, Z. Hua, S. Li, M. Hu, J. Li, and W. Shu. 2013.

613   Contemporary environmental variation determines microbial diversity patterns in

614   acid mine drainage. Isme J 7:1038-1050.

615   34. Lark, R. M. 1999. Soil-landform relationships at within-field scales: an

616   investigation using continuous classification. Geoderma 92:141-165.

617   35. Lauber, C. L., M. Hamady, R. Knight, and N. Fierer. 2009. Pyrosequencing-Based

618   Assessment of Soil pH as a Predictor of Soil Bacterial Community Structure at the

619   Continental Scale. Appl Environ Microb 75:5111-5120.

620   36. Lee, J. S., H. K. Lee, J. Hur, M. Andreev, and S. G. Hong. 2008. Diversity of the

621   lichenized fungi in King George Island, Antarctica, revealed by phylogenetic analysis

622   of partial large subunit rDNA sequences. J Microbiol Biotechn 18:1016-1023.

623   37. Lin, W., Y. Wang, B. Li, and Y. Pan. 2012. A biogeographic distribution of

624   magnetotactic bacteria influenced by salinity. Isme J 6:475-479.

625   38. Liu, X., and X. Zheng. 1988. Geology of volcanic rocks on Fildes Peninsula, King

626   George Island, West Antarctica. Chinese Journal of Polar Research 1988-01.

627   39. Lozupone, C. A., and R. Knight. 2007. Global patterns in bacterial diversity. P Natl

628   Acad Sci Usa 104:11436-11440.

629   40. Ludwig, W., J. Euzéby, and W. B. Whitman. 2010. Road map of the phyla

630   Bacteroidetes, Spirochaetes, Tenericutes (Mollicutes), Acidobacteria, Fibrobacteres,

631   Fusobacteria, Dictyoglomi, Gemmatimonadetes, Lentisphaerae, Verrucomicrobia,

632   Chlamydiae, and Planctomycetes, p. 1-19. *In* N. R. Krieg, J. T. Staley, D. R. Brown, B. P.

633   Hedlund, B. J. Paster, N. L. Ward, W. Ludwig, and W. B. Whitman (ed.), Bergey's

634   Manual® of Systematic Bacteriology: Volume Four The Bacteroidetes, Spirochaetes,

635   Tenericutes (Mollicutes), Acidobacteria, Fibrobacteres, Fusobacteria, Dictyoglomi,

636   Gemmatimonadetes, Lentisphaerae, Verrucomicrobia, Chlamydiae, and

637   Planctomycetes. Springer New York, New York, NY.

638   41. Mapelli, F., R. Marasco, A. Rizzi, F. Baldi, S. Ventura, D. Daffonchio, and S. Borin.

639   2011. Bacterial Communities Involved in Soil Formation and Plant Establishment

640   Triggered by Pyrite Bioweathering on Arctic Moraines. Microb Ecol 61:438-447.

641   42. Martiny, J., B. Bohannan, J. H. Brown, R. K. Colwell, J. A. Fuhrman, J. L. Green, M.

642   C. Horner-Devine, M. Kane, J. A. Krumins, C. R. Kuske, P. J. Morin, S. Naeem, L.

643   Ovreas, A. L. Reysenbach, V. H. Smith, and J. T. Staley. 2006. Microbial biogeography:

644   putting microorganisms on the map. Nat Rev Microbiol 4:102-112.

645   43. Meola, M., A. Lazzaro, and J. Zeyer. 2014. Diversity, resistance and resilience of

646   the bacterial communities at two alpine glacier forefields after a reciprocal soil

647   transplantation. Environ Microbiol 16:1918-1934.

648   44. Michel, R. F. M., C. E. G. R. Schaefer, J. Lopez-Martinez, F. N. B. Simas, N. W.

649   Haus, E. Serrano, and J. G. Bockheim. 2014. Soils and landforms from Fildes

650   Peninsula and Ardley Island, Maritime Antarctica. Geomorphology 225:76-86.

651   45. Murray, A. E., C. M. Preston, R. Massana, L. T. Taylor, A. Blakis, K. Wu, and E. F.

652   Delong. 1998. Seasonal and spatial variability of bacterial and archaeal assemblages

653   in the coastal waters near Anvers Island, Antarctica. Appl Environ Microb

654   64:2585-2595.

655   46. Olsson, P. A., I. Thingstrup, I. Jakobsen, and E. Baath. 1999. Estimation of the

656   biomass of arbuscular mycorrhizal fungi in a linseed field. Soil Biol Biochem

657   31:1879-1887.

658   47. Reith, F., J. Brugger, C. M. Zammit, A. L. Gregg, K. C. Goldfarb, G. L. Andersen, T.

659   Z. Desantis, Y. M. Piceno, E. L. Brodie, Z. Lu, Z. He, J. Zhou, and S. A. Wakelin. 2012.

660   Influence of geogenic factors on microbial communities in metallogenic Australian

661   soils. Isme J 6:2107-2118.

662   48. Rousk, J., E. Baath, P. C. Brookes, C. L. Lauber, C. Lozupone, J. G. Caporaso, R.

663   Knight, and N. Fierer. 2010. Soil bacterial and fungal communities across a pH

664   gradient in an arable soil. Isme J 4:1340-1351.

665   49. Santos, I. R., D. I. T. Favaro, C. E. G. R. Schaefer, and E. V. Silva-Filho. 2007.

666   Sediment geochemistry in coastal maritime Antarctica (Admiralty Bay, King George

667   Island): Evidence from rare earths and other elements. Mar Chem 107:464-474.

668   50. Schloss, P. D., S. L. Westcott, T. Ryabin, J. R. Hall, M. Hartmann, E. B. Hollister, R.

669 A. Lesniewski, B. B. Oakley, D. H. Parks, C. J. Robinson, J. W. Sahl, B. Stres, G. G.

670 Thallinger, D. J. Van Horn, and C. F. Weber. 2009. Introducing mothur: Open-Source,

671 Platform-Independent, Community-Supported Software for Describing and

672 Comparing Microbial Communities. Appl Environ Microb 75:7537-7541.

673 51. Spang, A., A. Poehlein, P. Offre, S. Zumbraegel, S. Haider, N. Rychlik, B. Nowka, C.

674 Schmeisser, E. V. Lebedeva, T. Rattei, C. Boehm, M. Schmid, A. Galushko, R.

675 Hatzenpichler, T. Weinmaier, R. Daniel, C. Schleper, E. Spieck, W. Streit, and M.

676 Wagner. 2012. The genome of the ammonia-oxidizing Candidatus Nitrososphaera

677 gargensis: insights into metabolic versatility and environmental adaptations. Environ

678 Microbiol 14:3122-3145.

679 52. Summers, S., A. S. Whiteley, L. C. Kelly, and C. S. Cockell. 2013. Land coverage

680 influences the bacterial community composition in the critical zone of a sub-Arctic

681 basaltic environment. Fems Microbiol Ecol 86:381-393.

682 53. Swanson, C. A., and M. K. Sliwinski. 2013. Archaeal assemblages inhabiting

683 temperate mixed forest soil fluctuate in taxon composition and spatial distribution

684 over time. Archaea (Vancouver, B.C.) 2013:870825-870825.

685 54. Tatur, A., A. Myrcha, and J. Niegodzisz. 1997. Formation of abandoned penguin

686 rookery ecosystems in the maritime Antarctic. Polar Biol 17:405-417.

687 55. Teixeira, L. C. R. S., R. S. Peixoto, J. C. Cury, W. J. Sul, V. H. Pellizari, J. Tiedje, and

688 A. S. Rosado. 2010. Bacterial diversity in rhizosphere soil from Antarctic vascular

689 plants of Admiralty Bay, maritime Antarctica. Isme J 4:989-1001.

690 56. Teixeira, L. C. R. S., R. S. Peixoto, J. C. Cury, W. J. Sul, V. H. Pellizari, J. Tiedje, and

691   A. S. Rosado. 2010. Bacterial diversity in rhizosphere soil from Antarctic vascular

692   plants of Admiralty Bay, maritime Antarctica. Isme J 4:989-1001.

693   57. Thomson, B. C., N. Ostle, N. Mcnamara, M. J. Bailey, A. S. Whiteley, and R. I.

694   Griffiths. 2010. Vegetation Affects the Relative Abundances of Dominant Soil

695   Bacterial Taxa and Soil Respiration Rates in an Upland Grassland Soil. Microb Ecol

696   59:335-343.

697   58. Tosi, S., B. Casado, R. Gerdol, and G. Caretta. 2002. Fungi isolated from Antarctic

698   mosses. Polar Biol 25:262-268.

699   59. Viles, H. A. 2012. Microbial geomorphology: A neglected link between life and

700   landscape. Geomorphology 157:6-16.

701   60. Walsh, D. A., R. T. Papke, and W. F. Doolittle. 2005. Archaeal diversity along a

702   soil salinity gradient prone to disturbance. Environ Microbiol 7:1655-1666.

703   61. Wang, N. F., T. Zhang, F. Zhang, E. T. Wang, J. F. He, H. Ding, B. T. Zhang, J. Liu, X.

704   B. Ran, and J. Y. Zang. 2015. Diversity and structure of soil bacterial communities in

705   the Fildes Region (maritime Antarctica) as revealed by 454 pyrosequencing. Front

706   Microbiol 6.

707   62. Watcham, E. P., M. J. Bentley, D. A. Hodgson, S. J. Roberts, P. T. Fretwell, J. M.

708   Lloyd, R. D. Larter, P. L. Whitehouse, M. J. Leng, P. Monien, and S. G. Moreton. 2011.

709   A new Holocene relative sea level curve for the South Shetland Islands, Antarctica.

710   Quaternary Sci Rev 30:3152-3170.

711   63. Webb, C. O., D. D. Ackerly, M. A. Mcpeek, and M. J. Donoghue. 2002.

712   Phylogenies and community ecology. Annual Review of Ecology and Systematics

713   33:475-505.

714   64. Wellman, P., and R. J. Tingey. 1981. Glaciation, erosion and uplift over part of

715   east antarctica. Nature 291:142-144.

716   65. Wynnwilliams, D. D. 1990. Ecological aspects of antarctic microbiology.

717   Advances in Microbial Ecology 11:71-146.

718   66. Xie, X. J., and T. X. Ren. 1993. National Geochemical Mapping And Environmental

719   Geochemistry - Progress In China. J Geochem Explor 49:15-34.

720   67. Yang, Y., Y. Gao, S. Wang, D. Xu, H. Yu, L. Wu, Q. Lin, Y. Hu, X. Li, Z. He, Y. Deng,

721   and J. Zhou. 2014. The microbial gene diversity along an elevation gradient of the

722   Tibetan grassland. Isme J 8:430-440.

723   68. Yao, Y., X. Wang, J. Li, J. Yang, S. Cao, F. Peng, L. Kurbatova, H. Peter, C. Braun,

724   and C. Li. 2017. A network for long-term monitoring of vegetation in the area of

725   Fildes Peninsula,King George Island. Advances in Polar Science:23-28.

726   69. Yergeau, E., K. K. Newsham, D. A. Pearce, and G. A. Kowalchuk. 2007. Patterns of

727   bacterial diversity across a range of Antarctic terrestrial habitats. Environ Microbiol

728   9:2670-82.

729   70. Yergeau, E., S. A. Schoondermark-Stolk, E. L. Brodie, S. Dejean, T. Z. Desantis, O.

730   Goncalves, Y. M. Piceno, G. L. Andersen, and G. A. Kowalchuk. 2009. Environmental

731   microarray analyses of Antarctic soil microbial communities. Isme J 3:340-351.

732   71. Yergeau, E., S. Bokhorst, A. H. L. Huiskes, H. T. S. Boschker, R. Aerts, and G. A.

733   Kowalchuk. 2007. Size and structure of bacterial, fungal and nematode communities

734   along an Antarctic environmental gradient. Fems Microbiol Ecol 59:436-451.

72. Young, I. M., J. W. Crawford, N. Nunan, W. Otten, and A. Spiers. 2008. Microbial distribution in soils: physics and scaling, p. 81-121. *In* D. L. Sparks (ed.), Advances in Agronomy, vol. 100.

73. Zelles, L. 1997. Phospholipid fatty acid profiles in selected members of soil microbial communities. Chemosphere 35:275-294.

74. Zhang, B., X. Wu, G. Zhang, S. Li, W. Zhang, X. Chen, L. Sun, B. Zhang, G. Liu, and T. Chen. 2016. The diversity and biogeography of the communities of Actinobacteria in the forelands of glaciers at a continental scale. Environ Res Lett 11.

75. Zhang, N., and M. Blackwell. 2002. Population structure of dogwood anthracnose fungus. Phytopathology 92:1276-1283.

76. Zhong, W., T. Gu, W. Wang, B. Zhang, X. Lin, Q. Huang, and W. Shen. 2010. The effects of mineral fertilizer and organic manure on soil microbial community and diversity. Plant Soil 326:511-522.

748

(a)   (b)

749
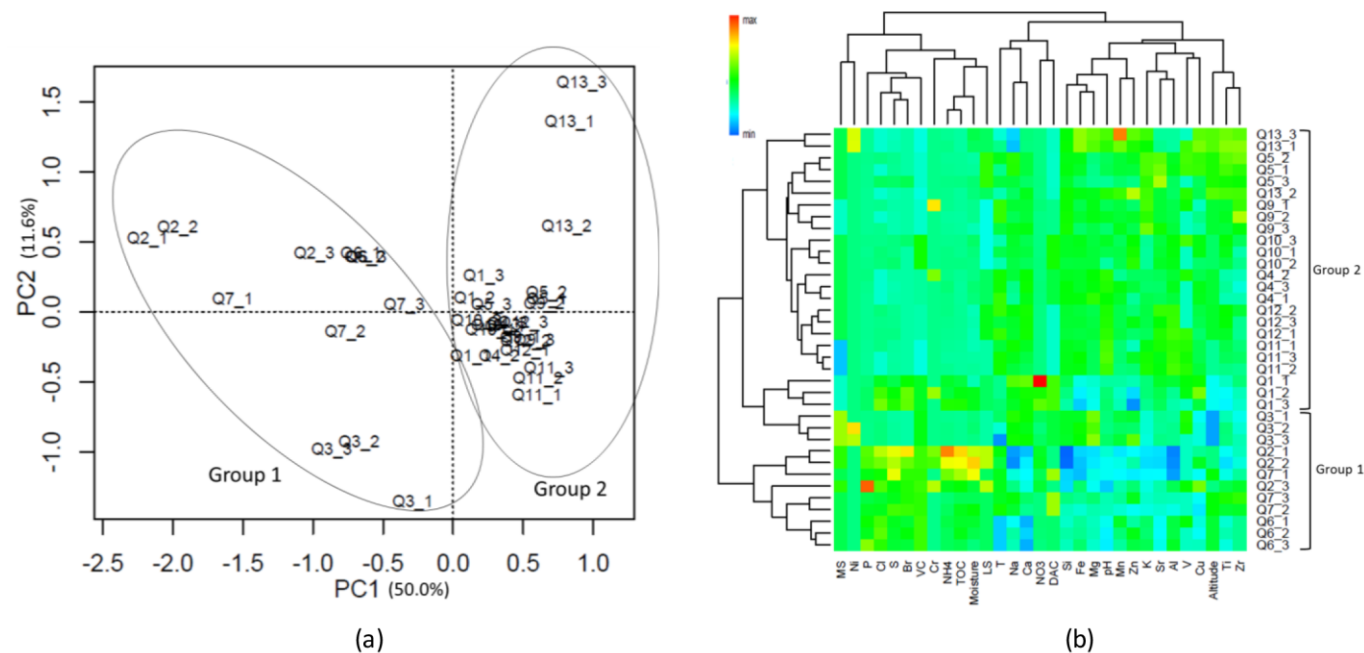
Fig. 1 (a) Principle component analysis (PCA) and (b) heatmap cluster analysis of the normalized soil elemental compositions and environmental attribute data. The values of PC1 and 2 are percentages of total variations that can be attributed to the corresponding axis. Abbreviations: T, temperature; TOC, total organic carbon; MS, moss species amount; LS, lichen species amount; DAC, hairgrass (*Deschampsia antarctica*) coverage; and VC, total vegetation coverage.

755
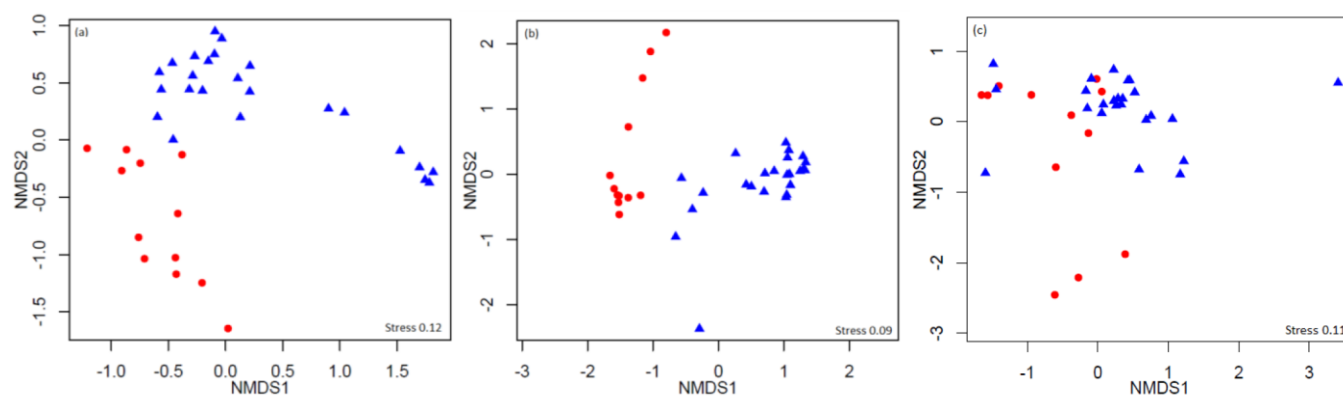
Fig. 2 Nonmetric multidimensional scaling (NMDS) analysis of the (a) bacterial, (b) archaeal, and (c) fungal Operational Taxonomic Unit (OTU) datasets. Circle = Group 1; triangle = Group 2.

37
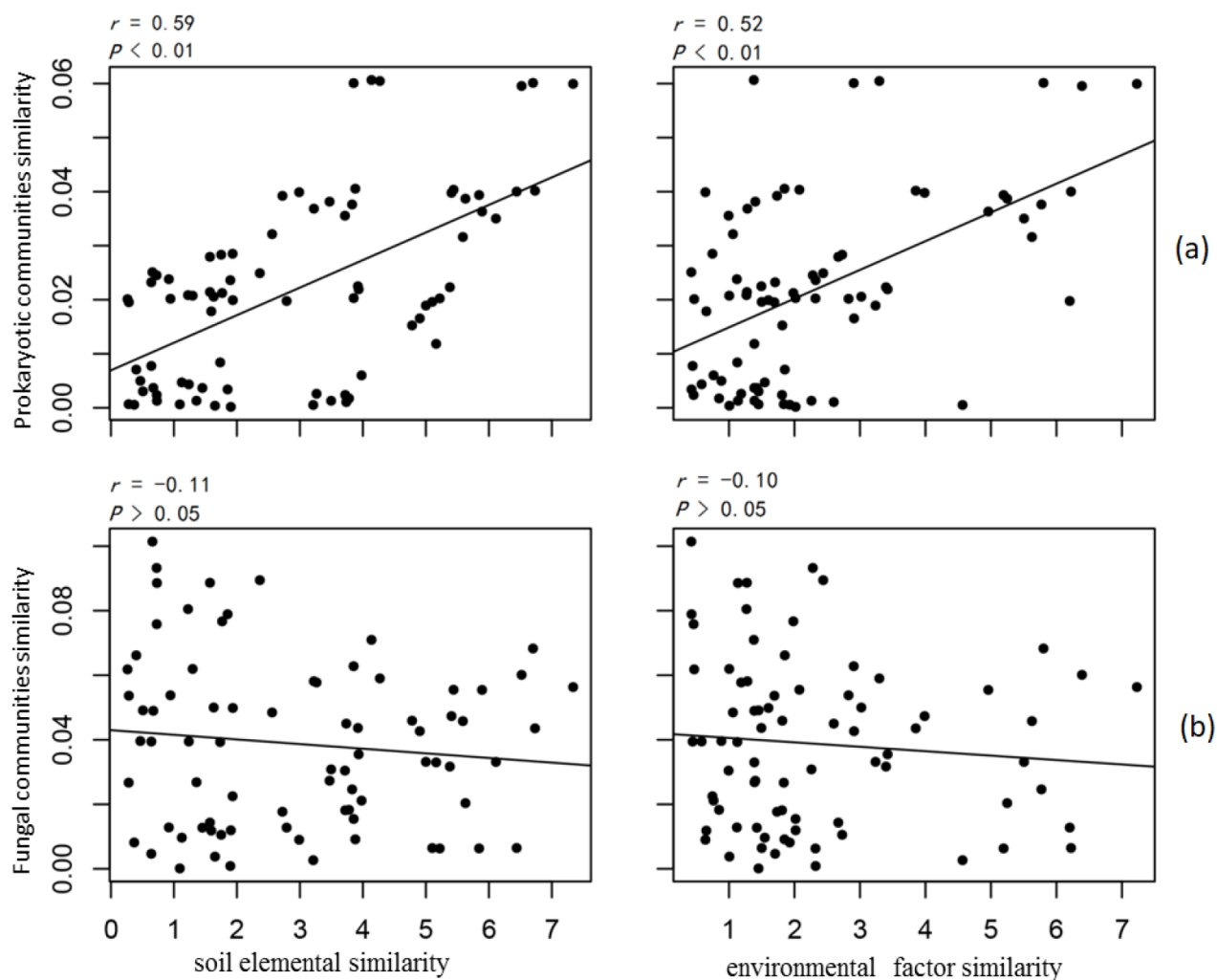
Fig. 3 Pearson correlations between (a) the prokaryotic community and (b) the fungal community with soil elemental compositions and environmental attributes. Similarity values are directly indicated by calculated pairwise Euclid distances between samples.
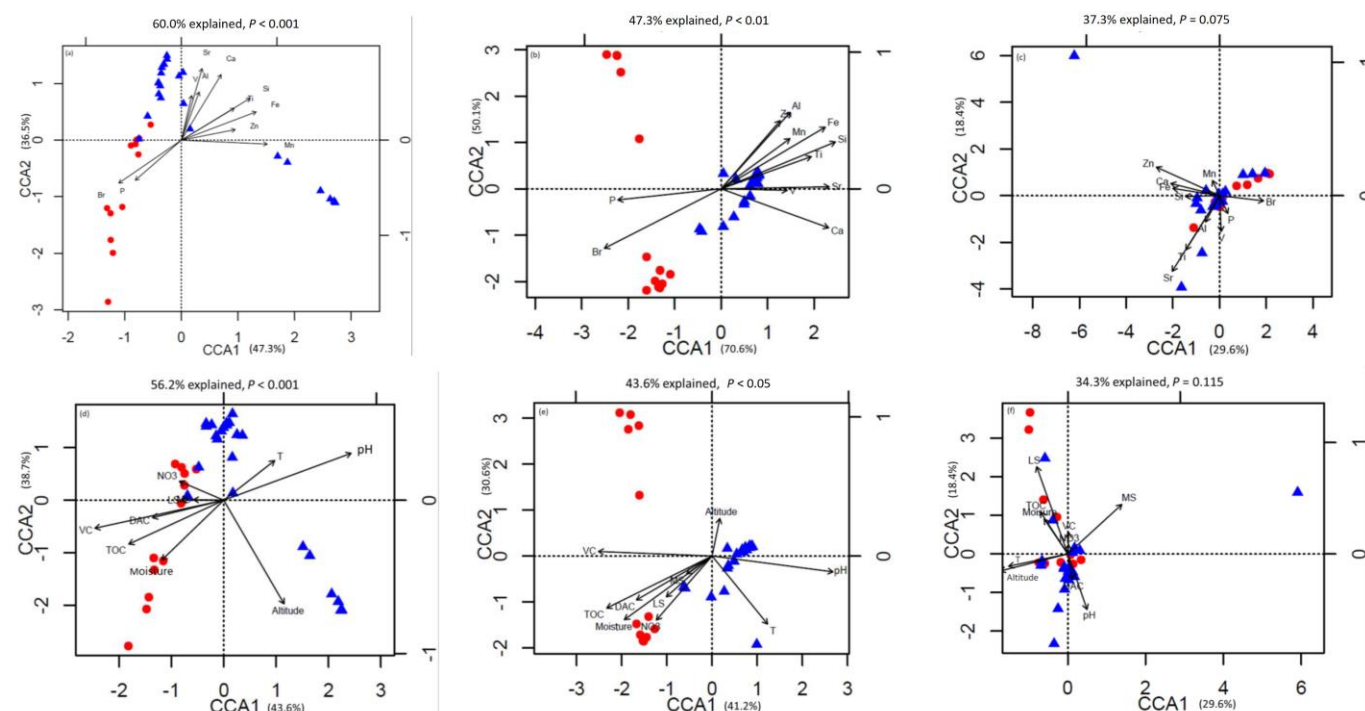
Fig. 4 Canonical correspondence analysis (CCA) of (a) Bacterial Operational Taxonomic Unit (OTU) data and elemental compositions; (b) archaeal OTU data and elemental compositions; (c) fungal OTU data and elemental compositions; (d) bacterial OTU data and environmental attributes; (e) archaeal OTU data and environmental attributes; (f) fungal OTU data and environmental attributes.
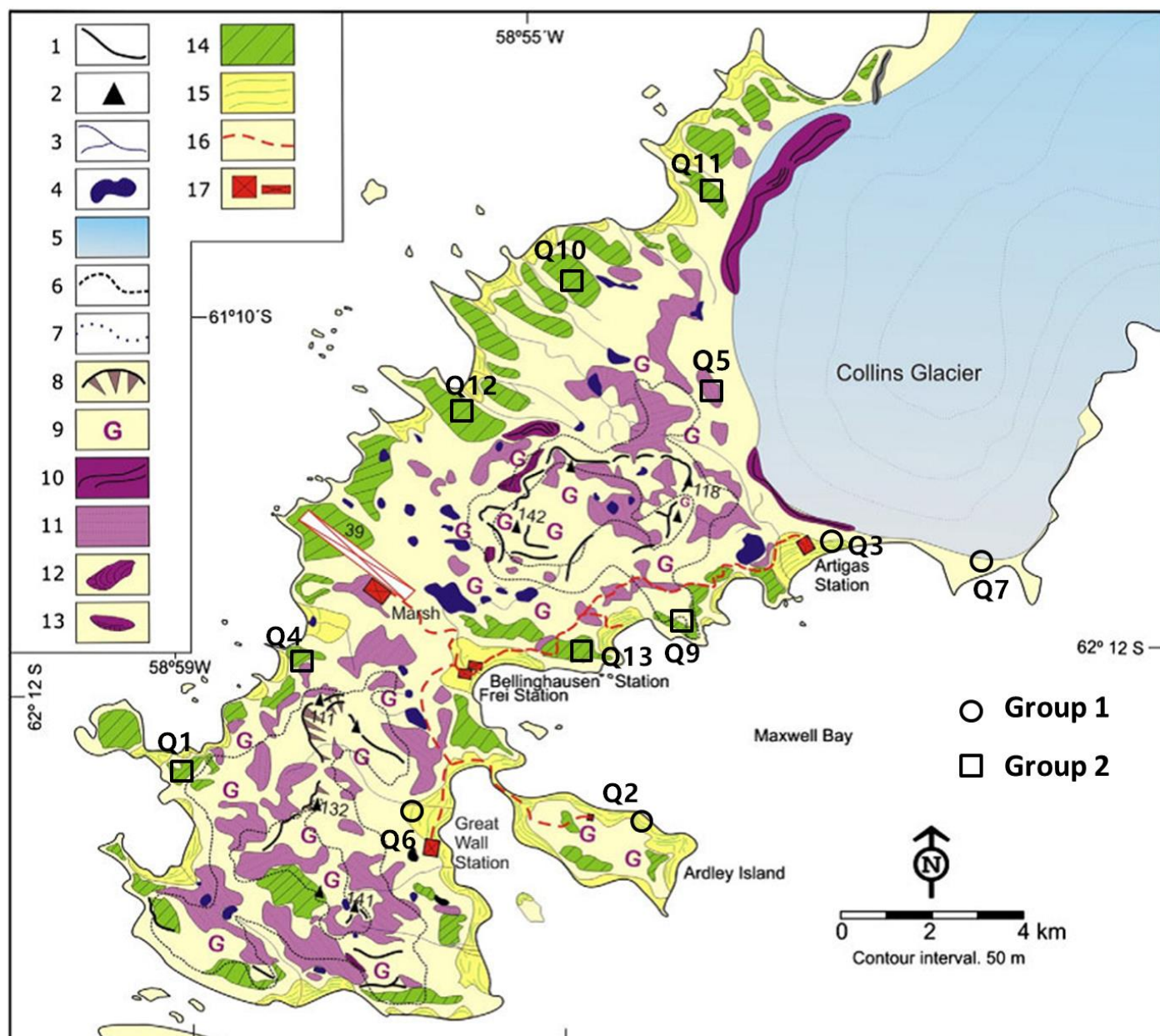
Fig. 5 Geomorphological map of the Fildes region, derived from Michel et al (44). Quadrats of Group 1 and Group 2 were located at Holocene raised beaches (No. 15) and marine platforms (periglacial landforms belonging to Tertiary volcanic stratigraphy, No. 14). The landform type of quadrat Q7 can be deemed as part of the Holocene raised beaches because it suffered recent glacio-isostatic uplift but was still covered by ice during that uplift (personal communication; Michel, 2018). Please refer to original literature for landforms marked by other numbers.

Soil elements	Prokaryote		Fungi		PLFA	
	r2	*P*-value	r2	*P*-value	r2	*P*-value
Al	0.2547	0.002**	0.0014	0.979	0.4648	0.001***
Ca	0.5647	0.001***	0.0012	0.981	0.3276	0.003**
Mn	0.5235	0.001***	0.0018	0.965	0.3859	0.001***
P	0.3372	0.005**	0.0488	0.378	0.2883	0.012*
Si	0.5621	0.001***	0.0299	0.609	0.6624	0.001***
Sr	0.5332	0.001***	0.2784	0.007*	0.4546	0.001***
Ti	0.3316	0.003**	0.2702	0.010*	0.3503	0.001***
V	0.2005	0.020*	0.0408	0.697	0.1828	0.041*
Zn	0.2309	0.009**	0.1120	0.216	0.3289	0.003**
Br	0.5059	0.001***	0.0369	0.558	0.6223	0.001***
Fe	0.5210	0.001***	0.0165	0.761	0.6978	0.001***
Environmental factors	Prokaryote		Fungi		PLFA	
	r2	*P*-value	r2	*P*-value	r2	*P*-value
TOC	0.4139	0.001***	0.1748	0.099	0.6776	0.001***
NO3	0.0667	0.286	0.0055	0.792	0.5661	0.120
T	0.1531	0.068	0.1867	0.055	0.1042	0.176
pH	0.6958	0.001***	0.2385	0.007**	0.4830	0.001***
Moisture	0.2644	0.012*	0.1231	0.128	0.5844	0.001***
Altitude	0.3003	0.002**	0.2463	0.024*	0.0065	0.912
DAC	0.2007	0.030*	0.0454	0.516	0.3487	0.003**
MS	0.0320	0.557	0.2723	0.007**	0.0505	0.430
LS	0.0624	0.325	0.5927	0.001***	0.1367	0.103
VC	0.6465	0.001***	0.0298	0.779	0.4422	0.001***

Table 1 Monte Carlo test of the factors (soil elemental compositions and environmental attributes) and compositions of microbial communities and phospholipid fatty acids (PLFA). Significant differences (P < 0.05) are indicated in bold. ***$P < 0.001$, **$P < 0.01$, *$P < 0.05$. *P*-values based on 999 permutations.

803

804

Quadrate code	Coordinates	Elevation /(m.a.s.l)	Aspect*	Number of grass tufts*	Grass cover/%*	Moss cover/%*	Lichen cover/%*
Q1	62°12'39''S 59°00'49''W	11	NW	26	20.75	55	10
Q2	62°12'39.6''S 58°55'35.9''W	34	N	30	11	46	43
Q3	62°11'05.1''S 58°52'37.3''W	22	NE	>50	37.50	56	6
Q4	62°12'00''S 58°59'40''W	42	NW	46	20	15	7
Q5	62°10'13''S 58°55'26''W	50	NW	1	1.75	35	5
Q6	62°13'00''S 58°57'52''W	42	NE	4	14	40	45
Q7	62°11'00.4''S 58°51'28.6''W	47	NE	>100	50	40	10
Q9	62°11'20''S 58°55'10''W	42	NW	24	10	20	10
Q10	62°09'09.1''S 58°55'44.2''W	37	NW	17	31	60	1
Q11	62°09'57.4''S 58°57'59.4''W	32	NW	2	1.50	-	20
Q12	62°10'33''S 58°58'16''W	43	NW	1	1.50	10	30
Q13	62°11'45.6''S 58°56'21.1''W	56	NE	1	2.50	5	25

805   *Data from the previous study (Yao *et al.*, 2017)

806   **Table 2** Locations and partial vegetation properties of the 12 soil quadrats.

807