Response to reviewer comments on manuscript bg-2018-488: "From substrate to soil in a pristine environment – pedochemical, micromorphological and microbiological properties from soils on James Ross Island, Antarctica"

We would like to thank the referees for their helpful and constructive comments, which greatly helped to improve our manuscript. We have prepared a response where we account for all points raised by the referees, as described below. We show the referees' comments in grey text, while our responses are formatted as standard text. Line indications refer to the changes in the revised manuscript.

Anonymous Referee #2:

Before answering the individual comments, we would like to thank the referee for taking a constructive and critical look at our manuscript.

However, I have a problem that the intention of the manuscript is not clearly presented. From the introduction, one may understand that the manuscript is devoted to: - increase the general understanding of soils developed in the transitional zone of the eastern APR (l. 109-111), - add to the understanding of drivers of soil microbial diversity in high latitude soils (l. 125-126), - perfrom micromorphological studies on soils of the eastern APR (l. 132-134).

At the end of the introduction it appears that it is all a little bit (l. 139-143). Further, the mentioned goals are not embedded into a theoretical framework. This makes it a bit hard to prepare the potential reader of what can be learned by reading the manuscript, which goes beyond a list of microorganisms. Here, the authors may consider reworking the introduction incl. the objectives chapter.

Many thanks for your comment. We completely rewrote the introduction according to you comment and changed almost the full introduction as follows:

"Therefore, soil scientific investigations became a relevant topic in Antarctic research, proving that there are actually soils in Antarctica (Jensen, 1916) and identifying soil forming processes (Ugolini, 1964)." (L. 87-89)

"However, diverse microbial communities thrive in a variety of Antarctic habitats, such as permafrost soils (Cowan et al., 2014)." (L. 97-99)

“Local conditions determine nutrient availability in Antarctic soils (Prietzel et al., 2019). Ca, Mg, K and P contents are generally high in igneous and volcanic rocks, whereas P and N contents are highest in ornithogenic soils. Ornithogenic soils are well known in Antarctica. The World Reference Base for Soil Resources (WRB, 2014) defines ornithogenic material (from Greek ornithos, bird, and genesis, origin) as material, which is characterized by penguin deposits mainly consisting of guano and often containing a high content of gravel transported by birds (cf. Ugolini, 1972).” (L. 103-109)

“At the microscale, microbial activity such as photosynthesis and nitrogen fixation has a distinct influence on soil chemical parameters, e.g. the increase of carbon and nitrogen contents in oligotrophic soils (Ganzert et al., 2011; Cowan et al., 2011; Niederberger et al., 2015). In return, these changes in soil characteristics affect microbial community composition.” (L. 132-136)

“Since most of the non-lichenized Antarctic fungi are known to be decomposers and their abundance and distribution is limited by plant derived nutrients, and bio-available Carbon (Arenz et al., 2011), the focus of this study lies on the prokaryotic interplay with soil characteristics and soil formation.” (L 137-140)

“We selected two different soils, representing coastal soils and inland soils of James Ross Island, developed on similar substrate and at similar topographic positions, but differing in local climate conditions and nutrient contents due to their relative position towards the mainly SW-winds. The western study site (Brandy Bay –BB) is located in a windward position and is highly influenced by sea spray, while the eastern study site (Santa Martha Cove – SMC), located behind a mountain range, is located in a leeward position (Prietzel et al., 2019). This setting enables an investigation of interdependencies particularly between prokaryotic life and soil properties, since the selected soils are not influenced by vascular plants, sulfides, and penguin rookeries.
With this, the main goal of our study is to identify major soil and microbiological properties in an extreme environment by combining pedochemical and micromorphological methods with microbial community studies based on high throughput sequence analyses. Thus, we will gain a better general understanding of (i) the main soil forming processes and (ii) the drivers of soil microbial diversity community structure in the eastern APR. This addresses also the question, if the

variance of pedogenic and microbiological properties are larger between depth increments within one profile (e.g. with different distances to the permafrost table) or between different soil profiles, i.e. due to different local environmental conditions.” (L. 151-167)

A further problem that I encounter is that only two profiles are compared. I understand that at such regions of the world, it is often not possible to carry out a longer-term field study. But one must be aware that this is not a very solid basis for identifying cause-and-effect relations between the soil environment and the microbiota. Multivariate statistics could be performed, because the soil increments were considered as being independent form each other (if I understand the Bray-Curtis dissimilarity right). But at the other hand the authors also reported of water and solute flow through the profiles, thus linking the different horizons. But I think that this problem can be solved by a more careful discussion.

Of course, we agree that the inclusion of additional soil profiles would increase the (statistical) power of our analysis. However, since this is not possible, at least for this paper, we followed your advice and rephrased the parts in the discussion based on our multivariate statistics and observations in a more careful fashion.

Following changes were made:

“In case of the pedogenic oxide ratios, 12.5% of the total compositional variation could be explained, which indicates a correlation between the microbial community structure and weathering at this very initial stage of soil formation.” (L. 595-597)

“For example, the amount and size of microaggregates have been shown to be important regarding prokaryotic colonization, leading to genetically distinct communities as well as cell densities in different size classes of aggregates (Ranjard et al., 2000). Thus, in addition to chemophysical environmental parameters, which shape the overall prokaryotic community, the microstructure of the initial soils could have a substantial influence on species distribution.” (L. 664-668)

We also added the following paragraph to better explain how we applied statistics:

“Multivariate statistics were performed for soil depth increments, which we considered to be independent. However, when processes are discussed that link soil horizons, e.g. water and solute flow through the profiles, we account for the limited number of two soil profiles with great care. We could not detect any environmental factors that increase or decrease the correlation between the chosen depth increments" (L. 628-632)

Also in the Abstract the goal of the study is written only in a quite vague manner. It is not clear, how the lee and luv position should impact the soil development? Was it the different input of salts with sea spray? Also the rest of the abstract is quite vague. E.g., what are the changes in soil microstructure below 20 cm depth and what is the potential impact on water availability and matter fluxes.

Many thanks for this comment. We rewrote the abstract as follows:

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

l. 53: Is it fair to say that the soils are dominated by bacterial taxa, when obviously no fungal taxa were investigated? But I believe as well that fungi most likely are of minor importance in these soils.

Most of non-lichenized Antarctic fungi are decomposers, and their abundance and distribution is limited by plant-derived nutrients and bio-available carbon (Arenz et al., 2011). Due to the absence of plants and lichens, and the overall low organic carbon contents, we assume that microbial communities are dominated by prokaryots and especially bacteria.

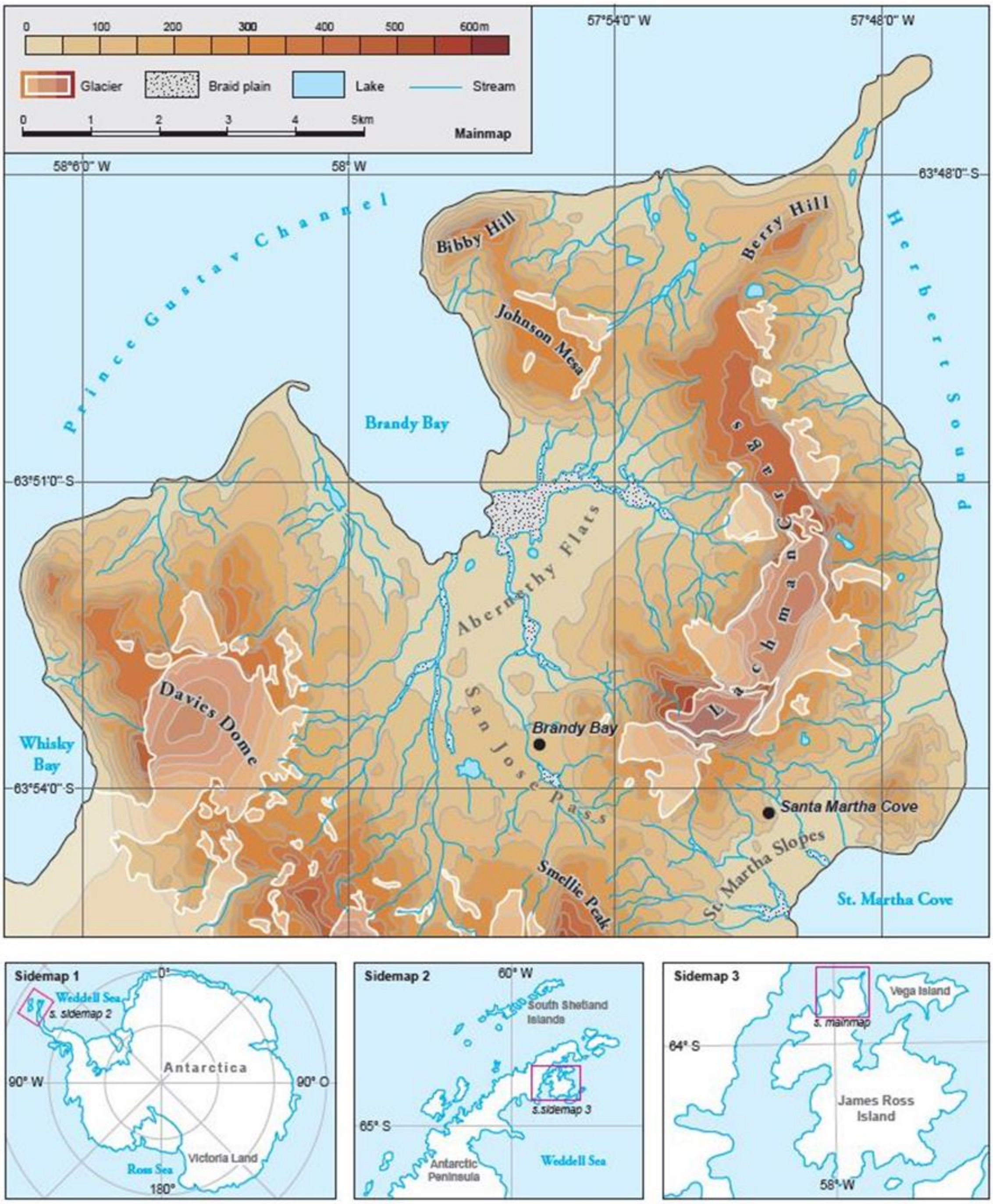
To clarify this, we changed the text as follows:

"In this study, two soil profiles from JRI (one at St. Martha Cove - SMC, and another at Brandy Bay - BB) were investigated, in order to gain information about the initial state of soil formation and its interplay with prokaryotic activity, by combining pedological, geochemical and microbiological methods. (L. 44-47)

"Since most of the non-lichenized Antarctic fungi are known to be decomposers and their abundance and distribution is limited by plant derived nutrients, and bio-available Carbon (Arenz et al., 2011), the focus of this studies lies on the prokaryotic interplay with soil characteristics and soil formation." (L. 137-140)

The introduction largely emphasis the different soil forming conditions, primarily related to climate, at different regions of Antarctica. Even though there are usually no figures in the introduction, here I would suggest to show a map of Antarctica highlighting the different areas that are mentioned in the discussion (it can be a slightly modified version of the present Fig. 1). But, of course, this also depends on whether the editors will accept this suggestion.

Many thanks for this remark. We replaced the satellite image of figure 1 with the following map. We suggest to mention figure 1 (L. 148) in the introduction and leave it in the methods section, because we describe there the study area more precisely.



0
100
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Mainmap
57°54'0" W
57°48'0" W
58°6'0" W
58° W
63°48'0" S
63°51'0" S
63°54'0" S
Prince Gustav Channel
Herbert Sound
Brandy Bay
Bibby Hill
Johnson Mesa
Berry Hill
Abernethy Flats
Lachman Crags
Whisky Bay
Davies Dome
San Jose Pass
Brandy Bay
Santa Martha Cove
St. Martha Slopes
St. Martha Cove
Smellie Peak
Sidemap 1
0°
Weddell Sea
s. sidemap 2
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Antarctica
90° O
Ross Sea
Victoria Land
180°
Sidemap 2
60° W
South Shetland Islands
65° S
s.sidemap 3
Antarctic Peninsula
Weddell Sea
Sidemap 3
s. mainmap
Vega Island
64° S
James Ross Island
58° W

l. 123-125: This sentence is not clear, actually sating that the microbial activity has an influence on the microbial composition . . . Please, rephrase.

We agree and rephrased this part as follows:

"At the microscale, microbial activity such as photosynthesis and nitrogen fixation has a distinct influence on soil chemical parameters, e.g. the increase of carbon and nitrogen contents in oligotrophic soils (Ganzert et al., 2011; Cowan et al., 2011; Niederberger et al., 2015). In return, these changes in soil characteristics affect microbial community composition." (L. 132-136)

Regional setting of James Ross Island, maritime Antarctica

Can be first subchapter of Material and Methods.

We moved the chapter "Regional setting of James Ross Island, maritime Antarctica" now as a new subchapter into the "Material and Methods" section.

l. 221: Please, indicate in what solution pH was measured.

EC and pH were measured in deionized water. Probably the wording was misleading. Therefore, we substituted the word "solution" by "water".

We changed the sentence as follows:

"Values of pH and electric conductivity were measured from bulk samples < 2mm in deionized water with a sample to water ratio of 1:2.5." (L. 235-237).

l. 223-228: I do not understand how Cinorg (the abbreviation has not been introduced) can be measured by dry combustion after fumigation of the carbonates with HCl. I rather assume that Corg was measured and Cinorg was calculated by difference of Ctot and Corg. Otherwise, methods are properly described.

Thank you for the important remark. We replaced "C_{inorg}" with the more common term "TIC". We also changed this part of the material and methods chapter to clarify this procedure:

"Carbon (C) and nitrogen (N) contents of the bulk soils were analyzed by dry combustion (Elementar CNS Vario Max Cube). 300 to 500mg per sample were analyzed in duplicate. In Order to distinguish between the total organic carbon (TOC) content and the total inorganic carbon (TIC), TIC was removed by acid fumigation after Ramnarine et al. (2011). 100 mg of the milled bulk soil samples were moistened with

20 to 40 µl of deionized water and put into a desiccator together with 100ml of 37% HCl. Afterwards, the samples were dried at 40°C. Finally, the samples were measured again by dry combustion (EuroVector EuroEA3000 Elemental Analyser) to obtain the TOC content. TIC content was calculated: TIC =C_{tot} – TOC." (L. 238-245)

l. 347: Why "virtually" unvegetated?

Many thanks, we deleted "virtually". The sentence was changed as follows:
"Both sites were unvegetated by cryptogamic or vascular plants." (L. 366-367)

l. 357-360: Since this property was not identified in the field I would shift this paragraph to the presentation of the micromorphological features.

We moved this paragraph as suggested:

l. 375-376: Present the TIC content as mg g-1. How can a TIC content transform to a TOC content? Consider rewording.

We changed the units in mg g^{-1} for TOC and TIC. "Transform" is a wrong word; we rewrote this sentence:

"The TIC content was low in both soils ranging between 0.1 and 0.3 mg g^{-1} in SMC and between 0.7 and 2.0 mg g^{-1} in BB. The TOC content ranges from 0.8-0.9 mg g^{-1} for SMC and from 1.4 and 2.6 mg g^{-1} for BB and increased there slightly with depth." (L. 391-393)

l. 378-380: Is there any explanation for the very low C/N ratios, most often much lower than in microbial biomass?

Long periods of atmospheric deposition of salts in soil surfaces, the lack of leaching in arid areas and insignificant biological turnover may lead to comparably high nitrogen contents (Bockheim, 1997; Barrett et al., 2007). In combination with the generally low C contents, these relative high N contents might lead to C/N ratios that depart from biological stoichiometry. Similar C/N ratios have been observed in other Antarctic soil habitats (e.g. Ganzert et al., 2011, Arenz et al., 2011, Barrett et al., 2007), which indicates this to be a common observation in such environments.

l. 395: Move this sentence to the beginning of the paragraph.

We moved the sentence as suggested.

In l. 192 a strong wind ablation was mentioned at BB. What is the role of the stronger ablation of fine material at BB on the chemical soil parameters? Can the selective erosion of a particular particle size blurr the results of the different weathering indices?

Many thanks for your questions. We assume that the enrichment of pebbles at BB protect the finer material beneath them. However, a selective erosion of distinct grains sizes cannot be excluded, at least before the enrichment of coarser pebbles at the soil surface took place. The effect of selective erosion of fine particles is shown by the weathering indices, with lower CIA values in the top centimeters of both soil profiles. At BB, the influence of salts from sea spray is pronounced, with highest Na and Mg contents in the topsoils. We discussed this result as a rejuvenation effect of the weathering indices by salt input (L 534-536).

Further, we added the following sentence to the results section:

"The amount of coarse material > 2mm was larger at the profile BB. Deflation processes led to a residual enrichment of larger grains and pebbles at the soil surface of both profiles. The permafrost table was not reached in both soil profiles, but ground ice was visible in a depth of 85cm at SMC." (L. 362-366)

l. 499-501: I would rewrite the sentence "Due to the absence of vascular plants, the ice-free area of JRI is a pristine laboratory and offers the exceptional opportunity to improve our understanding of the interrelations between soil formation and microbiological properties" as "The JRI offers an exceptional opportunity to improve our understanding of the interrelations between soil formation and microbiological properties in the absence of plants".

Thank you very much, we follow your suggestion and wrote:

"James Ross Island offers an exceptional opportunity to improve our understanding of the interrelations between soil formation and microbiological properties in the absence of plants." (L. 524-526)

l. 512-513: Present TOC and N contents as mg g-1.

We changed the units as follows:

“The examined soils on JRI were characterized by low TOC (0.9-2.6mg g^{-1}) and low total nitrogen contents (approx. 0.4mg g^{-1}), which is common for Antarctic soil environments (e.g. Cannone et al., 2008), and relative high pH values (7.4- 8.6).” (L. 536-538)

l. 516-517: If low P contents refer to total P, then this cannot be taken to indicate a relative juvenility of the soils. Soils rather loose P with development than they gain. In the soils under study, there is no P input by birds and I assume that also the atmospheric P input is negligible.

Many thanks for this remark. We omitted “and P”. (L 541)

l. 557-561: Here, I do not understand the line of argumentation.

To clarifiy our line of argumentation, we rephrased the paragraph as follows:

“Interestingly, the relative abundances of these taxa changed according to the degree of weathering. This could indicate a possible interrelation between the occurrence of these potential weathering-related organisms and the degree of weathering of Antarctic soils. (L. 583-585)

l. 562-567: This part is quite speculative, but could have been easily proven. Why has Na not been leached before the total elemental analysis of the soil minerals? I cannot imagine the formation of stable secondary mineral phases entrapping Na.

Thanks for this comment. We conduct the XRF analyses generally with the total soil material. Leaching in advance of this analysis might leach also other elements than Na and change the results in an incalculable way. We added the results for Na from ion chromatography to Table 1. The results show that the amount of Na is significantly higher in BB, which is most likely because of Na input by sea spray. Regardless of its origin, Na is detected by XRF and therefore taken into account for the calculation of the CIA. For this reason, we cannot rule out the possibility that the CIA values for the BB location may be underestimated.

We adjusted the following sentences:

"Ion Chromatography results show that the Na content is significantly higher at BB. This difference is most likely caused by the increased input of salts due to sea spray, which is known to carry high amounts of Na (Udisti et al., 2012). Since the calculation of the CIA takes Na into account (Nesbitt & Young, 1982), the CIA values would be significantly higher if the additional input of sea salts could be excluded." (L. 587-591)

l. 572-577: This is an important finding.

Many thanks.

l. 585-609: Nice discussion based on micromorphology.

Thank you very much as well.

l. 610-674: The discussion on the different taxa is well written, and it is a good message that this initial stage of soil development, chemolithoautotrophic lifestyles plays an important role for the generation of biomass and initial accumulation of soil organic carbon and nitrogen (even though this finding is not really new). But might be this offers also a good opportunity for an introduction, in order to base it better on a conceptional background.

Many thanks. At your advice, we added this to the introduction as follows:

"At the microscale, microbial activity such as photosynthesis and nitrogen fixation has a distinct influence on soil chemical parameters, e.g. the increase of carbon and nitrogen contents in oligotrophic soils (Ganzert et al., 2011; Cowan et al., 2011; Niederberger et al., 2015). In return, these changes in soil characteristics affect microbial community composition." (L. 132-136)

Additional Literature

Arenz, B., and Blanchette, R.: Distribution and Abundance of Soil Fungi in Antarctica at Sites on the Peninsula, Ross Sea Region and Mcmurdo Dry Valleys, Soil Biology and Biochemistry, 43, 308-315, 2011.

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