

The author's response to the third reviewing process (the associate Editor initial decision) on the manuscript entitled "Microbial colonisation in diverse surface soil types in Surtsey and diversity analysis of its subsurface microbiota". Enclosed are the response to the comments, questions and suggestions from the three referee's.

The authors are thankful for the kind general and specific comments on the paper given by the associate Editor. Enclosed, we have addressed his comments and questions by detailed point-by-point response.

We have made some additional corrections in the manuscript as following:

Change 182 to "181", page 1, line 21

Added "with its drill hole", page 2, line 65

Added (Figure 1 and Figure 2) page 3, line 94

Change methonogens to "methanogens" page 15, line 483

Added "surface" page 16, line 490

Added "higher" page 16, line 492

Other minor changes have possibly be made with track changes.

Associate Editor: 1) The general and specific comments of Anonymous Referee #1 are dealt with rather loosely and should be given further consideration and responded to more positively, e.g. with regards to improvements on the discussion, general description and background information about the island, borhole history and what did it initially reveal (bellshape of temp. curve, what causes it?).

Author's response: We have addressed these comments by rewritten the discussion chapter (see track changes), page 12-14.

"We have added some background information about the borehole history by following: (Pórarinsson, 1967, 1968; Pórarinsson, 1965). The eruption was thoroughly documented from the first plume of ash until the end of the lava flow in June 1967 and then a 181 m deep hole was drilled in 1979 to investigate the substructure of the volcano as well as the nature of the hydrothermal system (Jakobsson & Moore 1979). Consequently, the island of Surtsey provides....." page 2 line 59.

We have changed the graph in figure 5, page 26 and also the text for the figure.

Associate Editor: 2) The graphic presentations and explanations given are rather poor and confusing at times, major improvements necessary.

- Figure 1. There were 44 surface samples taken on the island, only 41 are found on the overview map of the island! You have to show all the samples on the map. If they are in the same spot as other nearby samples, that has to be shown. Also you need to make a reference to Figure 1 earlier in the main text, e.g. in section 2.1. (page 3 of the manuscript), where you describe the surface sampling and study sites.

Author's response: We have added the missing samples to figure 1, SJ11, SR2 and SR7. Figure 1, page 22

Author's response: We have included reference of Figure 1 page 3, line 91

Associate Editor:- Figure 2. No reference is made to this figure in the main text!! Should also be done in section 2.1. on page 3. The order of sample types in the figure text should be the same as on the photos above. You start with bare sand without droppings, i.e. SJ samples, etc. The last sentence of the figure text is irrelevant and should be taken out, the tracks after the respiration chambers are hardly noticeable on photos C and D.

Author's response: The order of sample types in the figure text has been changed in the same order as on the photos above. See following text with figure 2:

Figure 2. Pictures of the sample types. Samples were divided into three types: a) SJ samples (Barren sand or pumice without bird droppings, see SJ-5), b) SS samples (Barren sand or pumice with bird droppings, see SS-8) and SR samples c) (partly vegetated surfaces, see SR6) and d) (totally vegetated surfaces see SR-15).

Author's response: The last sentence of the figure text has been omitted.

Associate Editor:- Figure 3. The HAE has earlier raised questions on the presentation of data in the figure. a) what determines the order of samples? b) it is, in my mind, not logical to use a line diagram here as the samples are not connected, a scatter plot or even a table would be more appropriate, c) three types of surface samples were taken on the island (SJ, SS and SR), in Fig. 3 you, on the other hand, present four types of samples, which is confusing. d) on the x-axis of the graph a separation should be made between the different categories and the samples lying on the borderlines. The vertical lines should be drawn between the samples (e.g. SR-8 and SJ-9) etc.

Author's response: We have addressed the comments a) to c) by rewrite the text for figure 3 as following:

Figure 3. Total bacterial counts with plate count agar method at 22°C. The blue diamond shaped dots shows the total environmental bacterial counts with plate count agar method and the red squares shaped dots the numbers grown on R2A media at 22°C. The number of *Enterobacteriaceae* in the soil samples are showed with green triangle shaped dots. The samples order is oriented according to the appearance of the surface soil, starting with pure sand or pumice and in some cases with tiny vegetation (SR9, SR6) or bird droppings (SS9, SS8, SR8) including sample SR8 (A), partly vegetation including sample SR13 (B), total

vegetation with bird droppings including sample SS5 (C) and sand with bird droppings including sample SS10 (D). See Figure 1 for locations.

d) Has been addressed by drawing lines between samples differently as shown on figure 3

Associate Editor: - - Figure 4. Part of the sample labels on the left side of the graph are unreadable, as they overlap with each other, they need to be moved about and made readable.

Author's response: Unfortunately the program used does not allow movement of the sample label. However, the purpose of the figure is to represent the general differences and similarities between the four sample types. We could therefore omit the sample labels for clarity as shown in Figure 4 page 25.

Associate Editor: - - Figure 5. The text "Depth fo(?)temp. data logger" should be deleted from the diagram, can be indicated by e.g. TL, after the sample numbers. Note also the figure text. What was the main purpose of this exercise? Was it to remeasure the temperature in the hole or was it to take samples at different depths and temperature regimes. You have to mention the samples taken in the text. The last sentence of the figure text should be taken out, has already been explained in the main text.

Author's response: A new figure has been designed with new figure text. The text is as follows: Figure 5. Temperature measurements along the drill hole. Temperatures were measured along the drill hole at 5 m interval from the surface down to the bottom at 178 m with a borehole temperature meter. The circles show the depth and temperature of the SB samples.

Associate Editor: In the main text, section 3.1.5, p. 11, you make a reference to figures 5a and 5b. There is only one Figure 5 in the manuscript.

Author's response: We have addressed this comments, a in figure 5a has been omitted, page 11, line 315.

Figur 5b has been omitted, page 11, line 318

Associate Editor: - Figure 6. In the figure text you should mention that the SB samples are from the Surtsey borehole.

Author's response: We have addressed this comments by adding "from the borehole" in figure 6 text, page 27.

Associate Editor: - Figure 7. The presentation of the data would be more logical in a table than this columnar digrams. a) In Figure 7.a. your legend shows 18 different types, however, only about 6 are visible on the two diagrams. You do not give a legend for something you cannot see or identify. The same applies to Fig. 7.b., there you see less than half of the types indicated by the legend. b) Also note the order on the two figures, on Fig. a, you start with the larges class at the top, but on Fig. 7b., it is the opposite.

Author's response:

Author's response: a) We cannot agree to this comment as presentation of such data is often showed in this way and it shows very well the dominance of few taxa in the borehole, which is the purpose her. Moreover, this presentation shows within the legends more types than can be seen on the diagrams, as the method is highly specific by detecting less than 0,1% of taxa. These figures are presented on the legends, although not show in the diagrams, to show the complete diversity. This means that although graphically, the taxon are small, they are present and therefore their names and quantity is given in the legend. It can be identified by such deep sequence method.

The columns and the colours show the % of each major taxon (see text and % of each taxon on the right side for major and minor taxon).

We have address this comment in the Figure 7 text by include "minor" and "major" in the tex:

Figure 7. Sequencing results of the 16S rRNA gene with next generation sequencing method.

a) The longer reads of the v4-v6 regions with Titanium pyrosequencing on samples SB-5 and SB-6 (from the borehole). b) pyrosequencing of short fragment of the v6 region of the SB-4 (from the borehole) v6 library. The columns and the colours show the % of each major taxon (see text and % of each taxon on the right side for major and minor taxon).

b) We have addressed this comments by changing the fig. b in a similar way as fig a by stating with dominance class.