

# 南極の砂礫における動物棲息地の菌叢に対する網羅的遺伝子解析

園田 史朗<sup>1,2</sup>、青木 弘太郎<sup>2</sup>、辻本 恵<sup>3</sup>、伊村 智<sup>3</sup>、渡邊 研太郎<sup>3</sup>、石井 良和<sup>2</sup>

<sup>1</sup> 東京医科歯科大学大学院医歯学総合研究科統合呼吸器病学

<sup>2</sup> 東邦大学医学部微生物感染症学

<sup>3</sup> 国立極地研究所

## Metagenomic analysis habitation of the animal on the Antarctica sand gravel

Shiro Sonoda<sup>1,2</sup>, Kotaro Aoki<sup>2</sup>, Megumu Tsujimoto<sup>3</sup>, Satoshi Imura<sup>3</sup>, Kentaro Watanabe<sup>3</sup>, Yoshikazu Ishii<sup>2</sup>

<sup>1</sup>*The Integrated Pulmonology, Tokyo Medical and Dental Univ., Tokyo, Japan*

<sup>2</sup>*Department of Microbiology and Infectious Disease, Toho Univ. Faculty of Medicine, Tokyo, Japan*

<sup>3</sup>*National Institute of Polar Research, Tokyo, Japan*

### Introduction

Several studies have shown that microbiome in the Antarctica is highly diverse via Next-generation, culture-independent sequencing. The strictly environment, cold and ultraviolet light, of the Antarctica causes mammals and birds living severe, and then the Antarctica harbors diverse, viable and metabolically active microbial populations that represent almost all the major phylogenetic groups. Many of microbial taxa are uncultured and taxonomically unique, and a community that seems to be structured solely by abiotic processes. Recently, it was reported of analyzing of subglacial aqueous environments. However, the different between the place the lives, mammals and birds, exists (called “the Exist Place”) and the place they don't or rare (called “the Not Place”), is not obviously. In this study, we investigated the sand gravel sample around the Showa-Station and compared the sand gravel acquired from the Exist Place with the Not Place.

### Materials and Methods

The sand gravel samples were acquired the point of the map (below). The points, where the samples 1 to 6 were acquired, are the Exist Place and 7 to 11 are the Not Place. The sand gravels were stocked in the 4°C, and DNA were extracted from 0.25g of them using PowerSoil DNA Isolation kit. For shotgun metagenome sequencing, DNA library were prepared with with Nextera XT DNA Preparation Kit and sequencing was performed using a MiSeq sequencer (Illumina) in a 2×300bp paired-end run. Obtained reads were trimmed of low-quality sequences (the Phred quality score < Q30). Subsequently, remaining DNA sequence reads were analyzed by MEGABLAST against the non-redundant nucleotides databases and whole-genome sequence database (<ftp://ftp.ncbi.nlm.nih.gov/>), followed by taxonomic classification using MEGAN ver. 6 software.

### Results

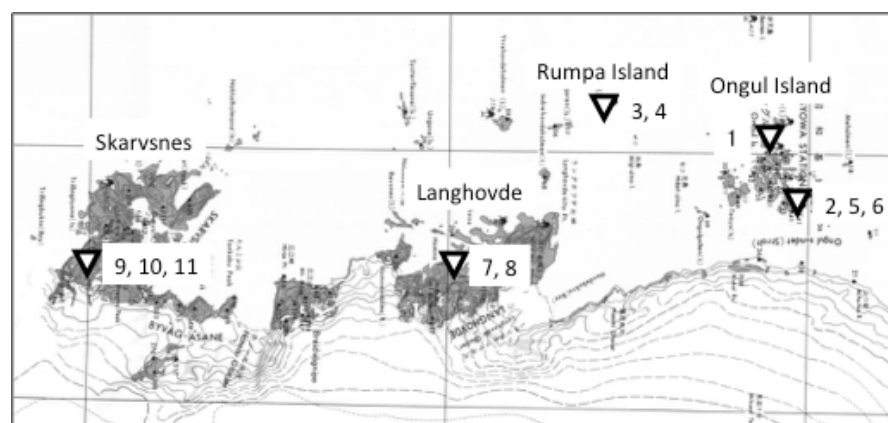
The reads number of  $13(\pm 3) \times 10^5$  were collected from each sample. At first, classified the kingdom, the most proportion (79.7±4.3%) of each sample is "No hits" (means not applicable among already known sequences), next to the bacteria (13.4±3.9%). The rest (the Archaea, Eukaryota and virus) is under 1% together. As analyzing to phylum among the Bacteria, the most phylum of the Exist Place is Proteobacteria (67.8±15.1%), on the other hand it of the Not Place is Actinobacteria (64.5±7.6%). The sample 4, which acquired from the place of rich fecas of mammals or birds (unknown in

detail), contained much of Bacteroidetes (32.4%) compared to others (0-10%). Besides, we investigated the phylum of bacteria related with causing diseases, there are a lot of Xanthomonadales and Burkholderiales in the Exist Place.

## Discussion

Our study showed the major phylum in the Exist Place is Proteobacteria, and in the Not Exist is Actinobacteria. It suggests that the lives make microbiome changed. Particularly, the more closely the lives related with, for example feces, the more microbiome seemed to be influenced. In addition, we confirmed the phylum related with diseases and the kinship existed more in the Exist Place than in the Not Place. It has not been clear that the phylum was originated from the Antarctica or came from our world with human. However, at least it suggests human is correlated to microbiome in the Antarctica.

Now as bacteria in only the Antarctica will be able to be empiric with global warming progressing and the field of activities of human expanding, the Antarctic bacteria is more and more important. Much of “No hits” seems to exist the unknown bacteria in the Antarctica. We planned the further analysis that bacterial species in detail and whether they have resistant factor to antibiotic.



## References

- Aislabie JM, Lau A, Dsouza M, Shepherd C, Rhodes P, Turner SJ. Bacterial composition of soils of the Lake Wellman area, Darwin Mountains, Antarctica. *Extremophiles : life under extreme conditions* 2013; 17:775-86.
- Barbosa A, Balague V, Valera F, et al. Age-Related Differences in the Gastrointestinal Microbiota of Chinstrap Penguins (*Pygoscelis antarctica*). *PloS one* 2016; 11:e0153215.
- Cary SC, McDonald IR, Barrett JE, Cowan DA. On the rocks: the microbiology of Antarctic Dry Valley soils. *Nature reviews Microbiology* 2010; 8:129-38.
- Christner BC, Priscu JC, Achberger AM, et al. A microbial ecosystem beneath the West Antarctic ice sheet. *Nature* 2014; 512:310-3.
- Dewar ML, Arnould JP, Krause L, Trathan P, Dann P, Smith SC. Influence of fasting during moult on the faecal microbiota of penguins. *PloS one* 2014; 9:e99996.
- Michaud L, Lo Giudice A, Mysara M, et al. Snow surface microbiome on the High Antarctic Plateau (DOME C). *PloS one* 2014; 9:e104505.
- Pessi IS, Osorio-Forero C, Galvez EJ, et al. Distinct composition signatures of archaeal and bacterial phylotypes in the Wanda Glacier forefield, Antarctic Peninsula. *FEMS microbiology ecology* 2015; 91:1-10.