

The 12<sup>th</sup> Symposium on Polar Science  
15 – 18 November 2021

National Institute of Polar Research  
Research Organization of Information and Systems

Session OB  
Polar Biology

Program and Abstracts

Convener : Nobuo Kokubun (NIPR)

## 【OB】 Polar Biology

### Scopes

This session covers the following research topics.

Polar marine ecosystem -biogeochemistry, environmental DNA, and ecology of phytoplankton to apex predators

Polar terrestrial ecosystem -environment, biodiversity, and biological responses

Convener : **Nobuo Kokubun (NIPR)**

### Real-time oral and poster presentations

**(09:55 – 11:30, 12:45 – 14:15, 14:30 - 16:00, 16:15 - 17:45)**

**Date: Wed. 17 November**

\* Invited oral presentation

09:55 - 10:00 Announcement Nobuo Kokubun (NIPR)			
Part1: Apex predators Chair: Nobuo Kokubun (NIPR)			
*OBo1	10:00 - 10:30	Inter-specific and intra-specific competition of two sympatrically breeding seabirds, Chinstrap and Gentoo penguins, at two neighboring colonies	*Won Young Lee (Korea Polar Research Institute), Seongseop Park (Korea Polar Research Institute & Incheon National Univ.), Kil Won Kim (Incheon National Univ.), Jeong-Hoon Kim (Korea Polar Research Institute), Jon-Ku Gal (Korea Polar Research Institute), <u>Hosung Chung (Korea Polar Research Institute)</u> *Kentaro Ito (NIPR), Akinori Takahashi (NIPR & SOKENDAI), Hina Watanabe (SOKENDAI), Junichi Takagi (NIPR), Nobuo Kokubun (NIPR & SOKENDAI), Yuuki Y. Watanabe (NIPR & SOKENDAI)
OBo2	10:30 - 10:50	Inter- and intra-colony food competition in foraging Adelie penguins	*Hina Watanabe (SOKENDAI), Junichi Takagi (NIPR), Akinori Takahashi (NIPR & SOKENDAI)
OBo3	10:50 - 11:10	3-D dive paths of foraging Adelie penguins under Antarctic fast sea-ice	*Bungo Nishizawa (NIPR), Takanori Sugawara (Nagoya Univ.), Jean Baptiste Thiebot (NIPR), Lindsay Young (Pacific Rim Conservation), Eric Vanderwerf (Pacific Rim Conservation), Fumio Sato (Yamashina Institute for Ornithology), Naoki Tomita (Yamashina Institute for Ornithology), Hiroshi Minami (National Research Institute of Far Seas Fisheries), Ken Yoda (Nagoya Univ.), Yutaka Watanuki (Hokkaido Univ.)
*OBo4	11:10 - 11:30	Seabird-fishery interactions revealed by bird-borne GPS and camera loggers: Interspecific comparisons of two albatross species	
Lunch			
Part 2: Marine ecosystem Dr. Masayoshi Sano (NIPR)			
*OBo5	12:45 - 13:15	Environmental DNA analysis for revealing fish community structure in the Arctic Ocean	*Tetsuya Kawakami (Hokkaido Univ.), Aya Yamazaki (Kyoto Univ.), Hiromichi Ueno (Hokkaido Univ.), Susumu Hyodo (AORI, Tokyo Univ.), Akihide Kasai (Hokkaido Univ.)
*OBo6	13:15 - 13:45	Environmental DNA reveals spatial distributions and trophic linkages of an Antarctic marine community	*Aiko Tachibana (TUMSAT), Masato Moteki (TUMSAT & NIPR), Ryosuke Makabe (NIPR & TUMSAT)
OBo7	13:45 - 14:15	Identification and phylogenetic analysis of fecal pellet-like dinoflagellates in the seasonal ice zone of the Southern Ocean	*Ryo Matsuda (Soka Univ.), Masayoshi Sano (NIPR), Shintaro Takao (NIES), Ryosuke Makabe (NIPR & SOKENDAI & TUMSAT), Masato Moteki (NIPR & TUMSAT), Norio Kurosawa (Soka Univ.)
	14:15 - 14:30	Break	
Part 3: Terrestrial ecosystem Chair: Satoshi Imura (NIPR)			
OBo8	14:30 - 15:00	To be notified	*Melinda Waterman (Univ. of Wollongong),

OB09	15:00 - 15:30	Diversity and species composition of bacteria in the Sor Rondane Mountains	*Shohei Hayashi (Shimane Univ.), Taka Ishida (Shimane Univ.), Kensuke Tadome (Saitama Nature Study Center), Shinji Takamura (Japan Mountain Guides Association), Satoshi Imura (NIPR)	OB_Hayashi_00612_01.pdf
OB010	15:30 - 16:00	Uphill energy transfer mechanism for photosynthesis in the Antarctic alga	*Makiko Kosugi (Astrobiology Center), Masato Kawasaki (High Energy Accelerator Research Organization), Yutaka Shibata (Tohoku Univ.), Kojiro Hara (Akita Prefectural Univ.), Shinichi Takaichi (Tokyo Univ. of Agriculture), Toshio Moriya (High Energy Accelerator Research Organization), Naruhiko Adachi (High Energy Accelerator Research Organization), Yasuhiro Kamei (National Institutes for Basic Biology), Yasuhiro Kashino (Univ. of Hyogo), Sakae Kudoh (NIPR), Hiroyuki Koike (Chuo Univ.), Toshiya Senda (High Energy Accelerator Research Organization)	OB_Kosugi_00257_01.pdf

### Poster presentations (15 November - 18 November, core time: 17 November 16:15 -

Core time in 17 November				
OBp1	16:15 - 16:45	Inter-species comparison on vertical distribution and seasonal population structure of the five sympatric mesopelagic copepods belonging same family (Aetideidae) in the western Arctic Ocean	*Yunosuke Koguchi (Hokkaido Univ.), Koki Tokuhiko (Hokkaido Univ.), Carin J. Ashjian (Woods Hole Oceanographic Institution), Robert G. Campbell (Univ. of Rhode Island), Atsushi Yamaguchi (Hokkaido Univ.)	OB_Koguchi_00594_01.pdf
OBp2	16:15 - 16:45	Spatial-temporal changes in the macrozooplankton community in the eastern Indian sector of the Southern Ocean during austral summer; Comparison between 1996 and 2018/2019	*Ippei Urabe (Hokkaido Univ.), Kohei Matsuno (Hokkaido Univ.), Rikuto Sugioka (Hokkaido Univ.), Ryan Driscoll (Alfred Wegener Institute Helmholtz Centre for Polar and Marine Research), Fokje L. Schaafsma (Wageningen Marine Research), Atsushi Yamaguchi (Hokkaido Univ.), Ryuichi Matsukura (Japan Fisheries Research and Education Agency), Hiroko Sasaki (Japan Fisheries Research and Education Agency), Hiroto Murase (TUMSAT)	OB_Urabe_00538_01.pdf
OBp3	16:15 - 16:45	Features of the Antarctic wintering team's meals (menus) -Breakfast, Lunch, Dinner, Snacks, Special meals (Party food), etc-	*Ritsuna Noguchi (Teikyo Heisei Univ.), Kazuki Hayakawa (Teikyo Heisei Univ.), Momoka Matsuda (Teikyo Heisei Univ.), Keisuke Motoyanagi (Teikyo Heisei Univ.), Keita Yamaguchi (Teikyo Heisei Univ.), Toru Koyama (NIPR), Satoshi Imura (NIPR)	OB_Noguchi_00615_02.pdf
OBp4	16:15 - 16:45	Factors affecting gestation periods in elasmobranch fishes	*Soma Tokunaga (Nagasaki Univ., SOKENDAI), Yuuki Watanabe (NIPR), Mai Kawano (Kyushu Univ. & Tokyo Univ.), Yuuki Kawabata (Nagasaki Univ.)	OB_Tokunaga_00611_01.pdf
OBp5	16:15 - 16:45	Behavioural thermoregulation linked to foraging in blue sharks	*Yuuki Watanabe (NIPR), Itsumi Nakamura (Nagasaki Univ.), Wei-Chuan Chiang (Fisheries Research Institute, Taiwan)	OB_Watanabe_00038_01.pdf
OBp6	16:15 - 16:45	Population trends of Adelie penguins in Lutzow-Holm Bay, East Antarctica	*Akinori Takahashi (NIPR)	OB_Takahashi_00127_01.pdf
OBp7	16:15 - 16:45	Isolation and Identification of Mortierella spp. from a Sanionia Moss Colony	*Natsumi Fujii (Osaka Prefecture Univ.), Ruka Kawasumi (Osaka Prefecture Univ.), Maria-Luz Herrero (NIBIO), Masaki Uchida (NIPR), Motoaki Tojo (Osaka Prefecture Univ.)	OB_Tojo_00101_02.pdf
OBp8	16:15 - 16:45	The abundance of microscopic phototrophs and invertebrates in microbial mat of freshwater lakes and an ephemeral wetland of the Sôya Coast, East Antarctica	*Tomotake Wada (SOKENDAI), Sakae Kudoh (NIPR & SOKENDAI), Hiroshi Koyama (SOKENDAI), Josef Elster (National Institute of Botany, Academy of Sciences of the Czech Republic & Centre for Polar Ecology, Univ. of South Bohemia), Jana Kviderová (National Institute of Botany, Academy of Sciences of the Czech Republic & Centre for Polar Ecology, Univ. of South Bohemia), Satoshi Imura (NIPR & SOKENDAI)	OB_Wada_00129_01

OBp9	16:45 - 17:15	Microbial community structures of sinking particles collected by using gel sediment traps in the Indian sector of the Southern Ocean during austral summer	*Masayoshi Sano (NIPR), Ryosuke Makabe (NIPR), Norio Kurosawa (Soka Univ.), Masato Moteki (TUMSAT)	OB_Sano_00270_01.pdf
OBp10	16:45 - 17:15	Morphological development of larval and juvenile Antarctic paralepidid fish <i>Notolepis coatsi</i> in Indian Ocean sector of the Southern Ocean	*Chiho Mitsuda (TUMSAT), Yusuke Iwai (TUMSAT), Aiko Tachibana (TUMSAT), Masato Moteki (TUMSAT & NIPR)	OB_Mitsuda_00519_01.pdf
OBp11	16:45 - 17:15	Animal-borne video observations of fish and squid capture by northern elephant seals in the mesopelagic zone	*Atsuya Ogata (SOKENDAI), Akinori Takahashi (NIPR & SOKENDAI), Taiki Adachi (NIPR & Univ. of St. Andrews), Yasuhiko Naito (NIPR), Rachel Holser (Univ. of California Santa Cruz), Arina Favilla (Univ. of California Santa Cruz), Daniel Costa (Univ. of California Santa Cruz)	OB_Ogata_00308_01.pdf
OBp12	16:45 - 17:15	Evaluation of group behaviour of fish by 3-D positioning biotelemetry	*Junichi Takagi (NIPR), Ko Fujioka (Japan Fisheries Research and Education Agency), Hiromu Fukuda (Japan Fisheries Research and Education Agency), Yoshimi Itaya (GKyoto Univ.), Kotaro Ichikawa (Kyoto Univ.), Nobuaki Arai (National Fisheries Univ.), Hiromichi Mitamura (Kyoto Univ.)	OB_Takagi_00234_01.pdf
OBp13	16:45 - 17:15	Distribution patterns of three common species of flying seabird off Vincennes Bay (East Antarctica)	*Yukina Suzuki (TUMSAT), Aiko Tachibana (TUMSAT), Amakazu Kazuo (TUMSAT), Masato Moteki (TUMSAT, NIPR)	OB_Suzuki_00563_01.pdf
OBp14	16:45 - 17:15	Analysis of mycobionts and associated bacterial microorganisms of rock tripe lichens in a continental and a maritime Antarctic areas	*Zichen He (Hiroshima Univ.), Megumu Tsujimoto (Keio Univ.), Satoshi Imura (NIPR), Takeshi Naganuma (Hiroshima Univ.)	OB_He_00185_01.pdf
OBp15	16:45 - 17:15	CO <sub>2</sub> emission resulting from rain-on-snow events amounts to 10% of yearly soil CO <sub>2</sub> emission in Ny-Ålesund	*Seiichiro Yonemura (Prefectural Univ. of Hiroshima), Masaki Uchida (NIPR), Ayaka Kishimoto-Mo (NIAES), Noriko Oura (NIAES)	OB_Yonemura_00506_01.pdf
OBp16	16:45 - 17:15	Microfungi associated with Antarctic and Arctic moss profiles	*Takashi Osono (Doshisha Univ.), Satoru Hobara (Rakuno Gakuen Univ.), Dai Hirose (Nihon Univ.), Masaki Uchida (NIPR)	OB_Osono_00009_01.pdf
OBp17	17:15 - 17:45	Seasonal variability in vertical distribution of zooplankton in the Antarctic seasonal ice zone	*Ryosuke Makabe (NIPR), Masayoshi Sano (NIPR), Masato Ito (NIPR), Keigo Takahashi (SOKENDAI), Shintaro Takao (NIES), Norio Kurosawa (Soka Univ.), Masato Moteki (TUMSAT)	OB_Makabe_00172_01.pdf
OBp18	17:15 - 17:45	Device of marine educational workshop kit, 'Let's observe the Diversity of Marine Plankton'	*Hisae Sakurai (NIPR), Masayoshi Sano (NIPR), Sakae Kudoh (NIPR), Kunio T. Takahashi (NIPR)	OB_Sakurai_00028_01.pdf
OBp19	17:15 - 17:45	Distribution depth of some pelagic and mesopelagic fishes in the Southern Ocean inferred from their eye and retinal morphology	*Mio Shibata (Mie Univ.), Taeko Miyazaki (Mie Univ.), Tetsuo Iwami (Tokyo Kasei-Gakuin Univ.), Hiroto Murase (TUMSAT)	OB_Shibata_00495_01.pdf
OBp20	17:15 - 17:45	Food management for Antarctic wintering team How long vegetables stay fresh, How to Preserve Vegetables, Hydroponics for Vegetables	*Ritsuna Noguchi (Teikyo Heisei Univ.), Daiki Maehara (Teikyo Heisei Univ.), Chisato Niwaike (Teikyo Heisei Univ.), Satoshi Imura (NIPR), Toru Koyama (NIPR)	OB_Noguchi_00615_01.pdf
OBp21	17:15 - 17:45	Wintering habitats of two congeneric diving seabirds in the southeastern Bering Sea	*Nobuo Kokubun (NIPR), Takashi Yamamoto (Meiji Univ.), Alexander Kitaysky (Univ. of Alaska Fairbanks), Akinori Takahashi (NIPR)	OB_Kokubun_000150_01.pdf
OBp22	17:15 - 17:45	Protecting marine biodiversity beyond national jurisdiction: a penguins' perspective	*Jean-Baptiste Thiebot (NIPR), Magali Dreyfus (CNRS & CERAPS-Lille Univ.)	OB_Thiebot_00157_01.pdf
OBp23	17:15 - 17:45	Genetic structures of genes involved in aromatic hydrocarbons metabolism from Antarctic soil bacterium strain BS19	*Azham Zulkharnain (Shibaura Institute of Technology), Seiryu Take (Shibaura Institute of Technology), Siti Aqlima Ahmad (Universiti Putra Malaysia)	OB_Zulkharnain_00143_01.pdf
OBp24	17:15 - 17:45	Isolation of Polylactic acid Degradable Bacterium strain N-3 from Antarctic Soil	*Kenta Sato (Shibaura Institute of Technology), Siti Aqlima Ahmad (Universiti Putra Malaysia), Azham Zulkharnain (Shibaura Institute of Technology)	OB_Sato_00517_01.pdf

OBp25	17:15 - 17:45	Optimization of carbazole degradation by Antarctic cold-tolerant bacterium strain BS1	*Tatsuya Niwano (Shibaura Institute of Technology), Siti Aqlima Ahmad (Universiti Putra Malaysia), Azham Zulkharnain (Shibaura Institute of Technology)	OB_Niwano_0158_01.pdf
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## Diversity and species composition of bacteria in the Sør Rondane Mountains

Shohei Hayashi<sup>1</sup>, Taka Ishida<sup>1</sup>, Kensuke Tadome<sup>2</sup>, Shinji Takamura<sup>3</sup> and Satoshi Imura<sup>4,5</sup>

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In order to reveal the biological diversity in Antarctica, the terrestrial biological research team of the 61st Japanese Antarctic Research Expedition surveyed the terrestrial biology of bacteria, lichen, and moss from January to February 2020. We stayed at Princess Elizabeth Antarctica (Belgium) and collected soil and sand samples from the northwestern region of the Sør Rondane Mountains (Figure 1). The region is inland (about 200 km from the coast) and severe environment for organisms. We isolated bacterial strains mainly of Actinobacteria (Figure 2) from the samples in four sites (two lichen containing sites, one nesting site, one poor site) in Mt. Yukidori Toride. We identified the isolated strains by 16S rRNA sequencing. The bacterial diversity of the isolated strains from the lichen containing and nesting sites was higher than that from the poor site. The species composition of the isolated strains from the nesting site was different from that of the lichen sites. In two lichen sites, the species composition was different. Therefore, the diversity and the species composition of bacteria depend on the character of sampling sites. The results suggest that bacteria adapted to the environmental condition. The strains which belonged to the genus *Rhodococcus* were isolated only from the nesting site. It indicates that the strains related to *Rhodococcus* can utilize excretion products of snow petrel and/or were carried by snow petrel.

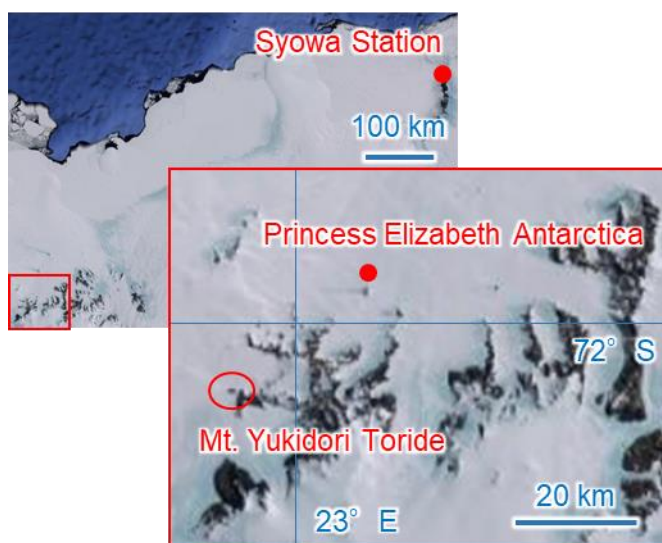


Figure 1. Research area.

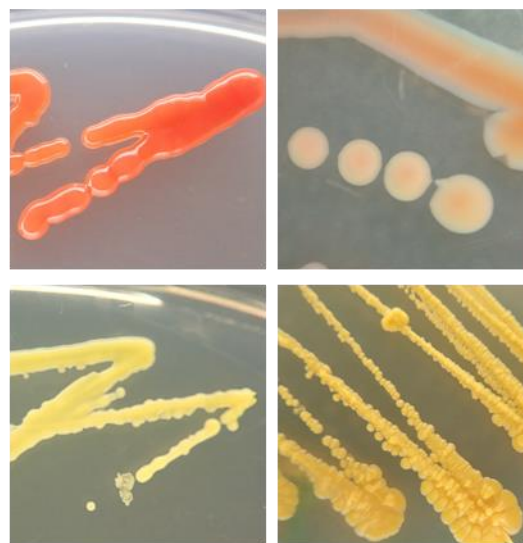


Figure 2. Isolated bacteria.

### References

Tadome, K., S. Hayashi, S. Takamura and S. Imura, Report of the terrestrial biological research activities in the Sør Rondane Mountains, 2019–2020 (JARE-61), Nankyoku Shiryo (Antarctic Record), 65, 45-69, 2021.

# Analysis of mycobionts and associated bacterial microorganisms of rock tripe lichens in a continental and a maritime Antarctic areas

Zichen He<sup>1</sup>, Megumu Tsujimoto<sup>2</sup>, Satoshi Imura<sup>3</sup>, Takeshi Naganuma<sup>1</sup>

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Rock tripe lichens that grow on fellfield rocks in a continental and a maritime Antarctic areas were sampled to analyze phylogenetic characteristics of mycobionts (fungi) and diversity of bacterial microorganisms in the lichen-symbionts by culture-independent phylogenetic analysis, id est, phylotype analysis. A total of 17 lichen samples, that is, 13 from the Syowa Station area and 4 from Signy Island, were collected.

The sequences of 18S rRNA gene were obtained by Sanger sequencing, and then the phylotypes were determined by BLAST search on the standard database of NCBI nucleotide collection (nr/nt). It was determined that lichen-forming fungi in all the samples were affiliated to the common lichen-forming genus of ascomycetes, *Umbilicaria*, and all the results showed the greatest similarity with the species *Umbilicaria aprina*. However, there were some slight differences between the samples collected in two different areas. The phylogenetic tree showed that the samples from Syowa Station and Signy Island are divided into two different clusters and that the relationship between Signy Island samples are more closely related.

V3-V4 region of bacterial 16S rRNA genes were obtained by Illumina MiSeq sequencing. A total of 761,633 reads, that is, 666,195 reads from Syowa Station and 95,438 reads from Signy Island, were analyzed. The phylotypes were determined with over 97% minimal similarity. The predominant phylum in all samples of Syowa Station were Bacteroidetes, and the proportion of every sample was greater than 50%, which was in absolute dominant position. In Signy Island, while in only one sample, the predominant phylum was Bacteroidetes, and the proportion was only 33.26% (only 4.26% higher than the subdominant phylum, Proteobacteria). In addition, the predominant phylum between the 4 samples of Signy Island showed variety, and the proportion of different phyla contained in each sample were close. By comparing overlap degree of the Venn diagram, the similarity of bacterial phylotypes contained in the samples of the two areas reached 65.38% on the phylum level, while only 18.48% on the species level. Through PCA (Principal Component Analysis), it was further confirmed that bacterial phylotypes contained in the samples from the two areas showed significant differences, while the bacterial phylotypes contained in the samples from same areas (especially Syowa Station) showed only small differences. On the other hand, the detailed results of phylum- and species-levels were showed respectively by the Chord diagram and the LefSe algorithm, which strongly support the above-mentioned conclusions. In summary, although the lichen-forming fungi were relatively similar in the two different areas of Antarctica, but associated bacterial microorganisms are quite different.

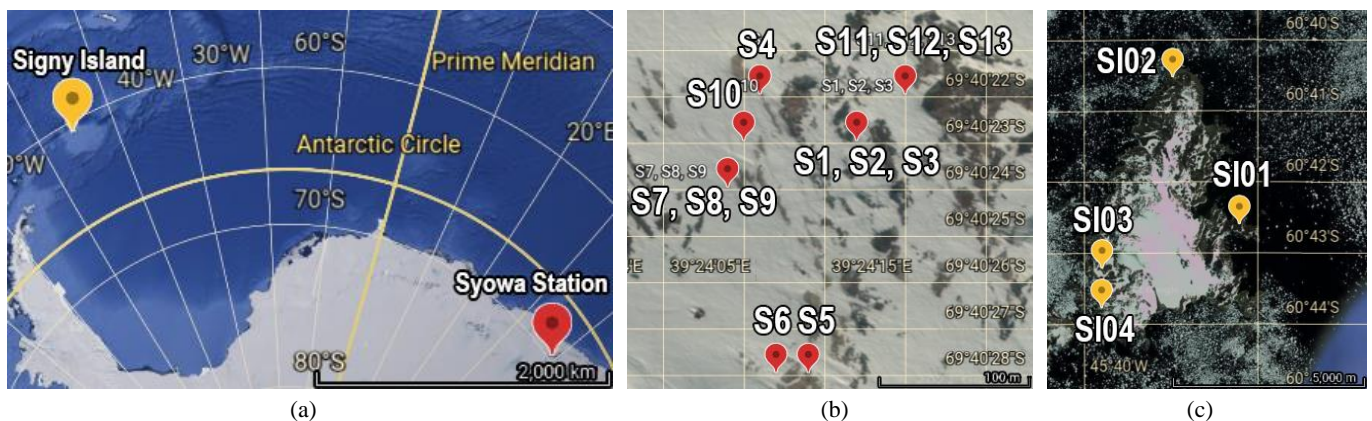


Figure 1. The sampling region and sites of lichen samples.

- (a) Sampling regions of Antarctica.
- (b) Sampling sites of Syowa Station.
- (c) Sampling sites of Signy Island.

## Inter- and intra-colony food competition in foraging Adélie penguins

Kentaro Ito<sup>1</sup>, Akinori Takahashi<sup>1,2</sup>, Hina Watanabe<sup>2</sup>, Junichi Takagi<sup>1</sup>, Nobuo Kokubun<sup>1,2</sup>, and Yuuki Y. Watanabe<sup>1,2</sup>

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Competition among conspecifics could be an essential factor affecting the foraging behavior of animals. Breeding seabirds may face intense competition for food around their colonies, especially when multiple colonies are closely located. Many seabird studies on competition avoidance have focused on either inter- or intra-colony competition, although these two types of competitions could interact with each other. For instance, previous studies on inter-colony foraging area segregation rarely considered the effect of intra-colony competition. The purpose of this study is to combine inter- and intra-colony competition and quantify the effect of inter-colony foraging area segregation on mitigating overall food competition. We tracked the foraging movements of Adélie penguins *Pygoscelis adeliae* from two neighboring (2 km apart) colonies by using bird-borne GPS-depth loggers in two summer seasons with contrasting sea ice conditions; extensive fast sea ice in 2018/2019 (“fast ice” year) and almost no sea ice in 2016/2017 (“open sea” year). The intensity of competition was quantified based on the sum of foraging area overlaps of all GPS deployed individuals to take into account both inter- and intra-colony competition (hereafter total overlap). The intensity of competition was compared with the simulated values from the null-model that assumes no avoidance behavior between colonies. GPS results showed that foraging areas of two colonies segregated clearly in the fast ice year, and overlapped extensively in the open sea year. Nevertheless, the total overlap was much larger in the fast ice year than in the open sea year, because clear segregation contributing to smaller inter-colony overlap also involved much larger intra-colony overlap. In the fast ice year, individuals tended to avoid foraging close to the other colony in comparison with the simulated foraging places, which reduced inter-colony overlap but increased intra-colony overlap: overall, most individuals had smaller total overlaps than the simulated values. Our results demonstrate that both inter- and intra-colony competition, not either one, should be considered to evaluate the effect of inter-colony foraging area segregation on competition avoidance. Furthermore, we suggest that Adélie penguins in fast sea ice may change inter- and intra-colony overlap by avoiding near other colonies and consequently mitigate overall competition.



# Environmental DNA analysis for revealing fish community structure in the Arctic Ocean

Tatsuya Kawakami<sup>1</sup>, Aya Yamazaki<sup>2</sup>, Hiromichi Ueno<sup>1</sup>, Susumu Hyodo<sup>3</sup> and Akihide Kasai<sup>1</sup>

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<sup>2</sup>Connectivity of Hills, Humans and Oceans Unit, Kyoto University

<sup>3</sup>Atmosphere and Ocean Research Institute, The University of Tokyo

Here, we introduce our challenges to reveal fish community structure in the Arctic Ocean using environmental DNA (eDNA) analysis, which is conducted under the Arctic Challenge for Sustainability II (ArCS II) Project, an Arctic region research project initiated in Japan.

Climate change in the Arctic such as increasing water temperature and rapid reduction in the extent of sea ice would induce drastic change in the Arctic biodiversity and ecosystem. Although recent studies have extended our knowledge about the response of the Arctic ecosystem to environmental changes (reviewed in Hirawake et al., 2021), the paucity of reliable baseline data still hinders the evaluation of how climate change alters the Arctic ecosystem (Wassmann et al., 2012). However, comprehensive research in the Arctic Ocean has often been difficult by conventional methods (e.g., bottom trawling, acoustic census, and statistical analysis based on catchment data) because those are labor-intensive and have limitations in operating in the distant area where is difficult to access and seasonally covered by thick ice.

A novel approach, analyzing eDNA, which is DNA extracted from environmental samples (e.g., water, sediment, soil, and air), would have a great potential to facilitate the study of the Arctic ecosystem. eDNA shed from aquatic organisms in various forms (e.g., cells, feces, gametes) can be retrieved from seawater by filtration and used in quantitative PCR assay or metabarcoding to detect organisms present in the study area. Recent studies employing eDNA as a trace of aquatic macro-organisms including fish successfully demonstrated that eDNA is a powerful tool to collect their ecological information in the diverse aquatic environment including the Arctic region (Lacoursière - Roussel et al., 2018). eDNA analysis requires a small amount of water, so that it is less laborious and cost-effective and can be performed where the conventional method is hardly conducted.

The primary aim of this study is to describe the present status of fish community structure in the Arctic Ocean using eDNA analysis. Previous studies have indicated that fish can be very susceptible to global warming (Wassmann et al., 2012). Elevating temperature, sea-ice cover reduction, and changes in phenology of primary production would induce poleward expansion of their distributions and anomalous fluctuations in the abundances. Fishes are also important for the Arctic culture and economy, so their ecological changes will also influence human activity. Therefore, eDNA analysis of fishes would provide valuable information to understand the impacts of climate change.

eDNA samples were collected from surface seawater in the Arctic Ocean (the Chukchi Sea and the Canada Basin), the Bering Sea, and the North Pacific Ocean, during the cruise of R/V Mirai (JAMSTEC) held from September to November 2020 (Fig.1). Preliminary metabarcoding analysis successfully detected fish eDNA from all sites (44 sites), and they were assigned to 170 taxa. In the Arctic Ocean, a total of 21 taxa was detected with *Ammodytes hexapterus* as the most frequently found species. The number of detected taxa was almost equivalent to that of the Bering Sea (30 taxa), but considerably lower than in the North Pacific (95 taxa). Cluster analysis based on dissimilarity index among regions indicated that the fish community in the Arctic Ocean was discernible from those of the Bering Sea and the North Pacific (Fig. 1). It is also found that the fish community around the Bering Strait was composed of communities belonging to both the Arctic and the Bering clusters. These results suggest that the frontal areas between the large-scale water masses may work as boundaries to delimit the fish distribution.

This study demonstrated that eDNA is a promising tool to study the ecological impact of climate change on Arctic fishes.

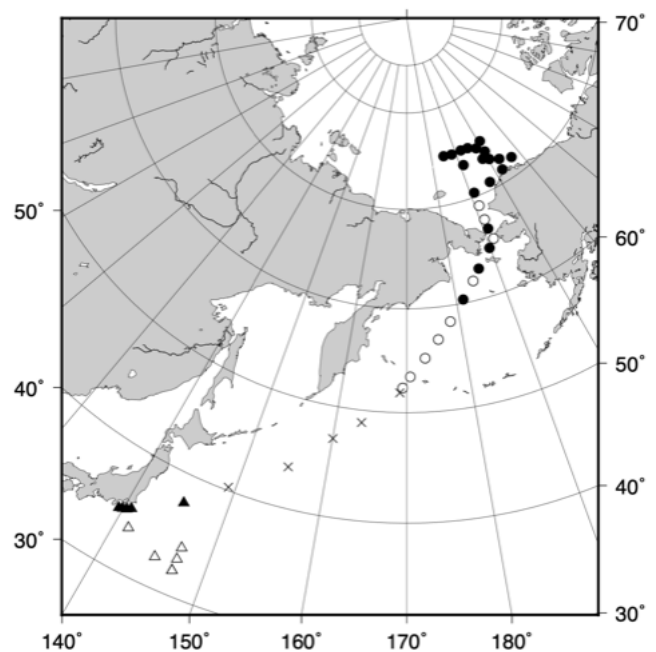


Figure 1 Sampling stations for environmental DNA. Different symbols indicate different fish communities.

However, many technical issues remain to be solved, e.g., insufficient reference sequence database, possible false-negative errors, optimization of sampling effort. These studies will expand the potential use of eDNA and encourage future studies to uncover the climate-induced changes in the Arctic ecosystem.

### **References**

Hirawake, T., M. Uchida, H. Abe, et al., Response of Arctic biodiversity and ecosystem to environmental changes: Findings from the ArCS project, *Polar Sci.*, 27:100533, 2021.

Lacoursière - Roussel, A., K. Howland, E. Normandeau, et al, eDNA metabarcoding as a new surveillance approach for coastal Arctic biodiversity, *Ecol. Evol.*, 8, 7763–7777, 2018.

Wassmann, P., C.M. Duarte, S. Agustí and M.K. Sejr, Footprints of climate change in the Arctic marine ecosystem, *Global Change Biol.*, 17, 1235–1249, 2011.

## Inter-species comparison on vertical distribution and seasonal population structure of the five sympatric mesopelagic copepods belonging same family (Aetideidae) in the western Arctic Ocean

Yunosuke Koguchi<sup>1</sup>, Koki Tokuhira<sup>1</sup>, Carin J. Ashjian<sup>2</sup>, Robert G. Campbell<sup>3</sup>, Atsushi Yamaguchi<sup>1</sup>

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In the marine ecosystem of the Arctic Ocean, zooplankton has an important role in the energy transfer for the higher trophic level organisms and mediator of the vertical material transportation (biological pump). While copepods dominate the zooplankton community in the Arctic Ocean, most of our knowledge of their ecology is derived from summer (midnight sun), and comparable information during the winter (polar night) and year-round observation is extremely scarce. Moreover, the ecological information of the Arctic planktonic copepods is mainly available for the species of the surface-dwelling small-sized copepods (e.g. *Pseudocalanus* spp.) or interzonal particle feeding copepods having diapause phase at deeper depth (*Calanus* spp.). Thus, little ecological information is available for the middle-sized mesopelagic copepods in the Arctic Ocean. As mesopelagic middle-sized copepods, Aetideidae is the most numerous and is reported to have an important role in both species diversity and vertical mass transport. However, because of the difficulty of species identification of the early copepodite stages, our knowledge of their vertical distribution and seasonal population structure is scarce. In this study, we identified the five sympatric aetideid copepods including their early copepodite stages, and evaluated vertical distribution and seasonal population structure throughout the year based on the year-round time-series samples collected at ice drifting station (SHEBA) in the western Arctic Ocean.

The SHEBA ice station drifted from the Canadian Basin to the Mendeleyev Basin during October 1997 to September 1998. Vertical stratification sampling was conducted by 1 m<sup>2</sup> mouth area closing net equipped 150 µm or 53 µm mesh from 2–7 depth layers of 0–3500 m depth at 10–14 days interval. Samples were preserved by 4% buffered formalin seawater. Five dominant aetideid copepods (*Chiridius obtusifrons*, *Gaetanus tenuispinus*, *G. brevispinus*, *Aetideopsis multiserrata*, *A. rostrata*) were identified and enumerated with each copepodite stage under a stereomicroscope. The mean copepodite stage (MCS) was calculated based on the abundance data for each sampling date. The vertical distribution center ( $D_{50\%}$ ) was calculated for each copepodite stage. The difference of  $D_{50\%}$  between the polar night and the midnight sun within a species was tested by Mann-Whitney *U*-test, and the difference of  $D_{50\%}$  between species at the same period was tested by one-way ANOVA and post hoc test.

At SHEBA station, polar night and midnight sun were at from early November-early February and from late April-late August, respectively. For both periods, *A. rostrata* occurred at the deepest depths ( $D_{50\%}$ : 1032–1065 m). The other four species distributed similar depths ( $D_{50\%}$ : 609–709 m) during the midnight sun. For the polar night, *C. obtusifrons* and *G. tenuispinus* were distributed at shallower depths ( $D_{50\%}$ : 298–381 m). Seasonal changes in vertical distribution and population structure indicate that there were two patterns in the vertical distribution and reproduction timing of the four aetideid copepods. Thus, *C. obtusifrons* and *G. tenuispinus* ascended to shallow depths during the polar night and had reproduction around the end of the polar night to the beginning of the daylight increasing season. On the other hand, *G. brevispinus* and *A. multiserrata* remained at the same depths both polar night and midnight sun, and their reproductions occurred for the earlier timing (beginning of the polar night). Such differences in vertical distribution and reproduction timing within the five mesopelagic middle-sized copepods belonging to the same family (Aetideidae) may have a function to reduce species competition at the mesopelagic layer of the Arctic Ocean where food resources are scarce.

## Wintering habitats of two congeneric diving seabirds in the southeastern Bering Sea

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Spatial segregation between closely related species is a key to understand the mechanisms by that they coexist in an ecosystem, and their possible different responses to environmental change. Common (*Uria aalge*; hereafter COMUs) and thick-billed (*U. lomvia*; hereafter TBMUs) murres are one of the most abundant and closely related diving seabirds in the southeastern Bering Sea. Although their inter-specific differences in diving behavior and food habits were reported in detail during breeding season (Kokubun et al. 2016), those during non-breeding seasons are not well understood (but see McFarlane et al. 2015; Takahashi et al. 2021). In this study we aimed to reveal wintering locations and diving behavior of the two species. We deployed 5 and 15 geolocators on leg of COMU and TBMU in 2013, and 11 and 16 geolocators on COMU and TBMU in 2014, on Saint George Island, southeastern Bering Sea. Among them, we obtained sufficient datasets of wintering locations and diving from 5 COMUs and 7 TBMUs deployed in 2013, and 5 COMUs and 7 TBMU deployed in 2014. We classified the wintering locations into four categories by geographic features and bathymetry: Bering Sea Shelf (bottom depth  $\leq$  200m, north of Aleutian Islands), Bering Sea Basin (bottom depth  $>$  200m, north of Aleutian Islands), Aleutian Islands (within 50km from the Aleutian Islands chain), and North Pacific (south of Aleutian Islands). We investigated diving depths and compared those with the estimated bottom depth at the location. If the bird dived to more than 80% of the water column, we assumed that the dive as a benthic dive. We found that both COMUs and TBMUs stayed in the Bering Sea Shelf areas during autumn (September to November). After that, COMUs stayed longer in the Bering Sea Shelf until next spring (April to May) whereas TBMU moved towards south or deeper areas (Bering Sea Basin, Aleutian Islands, or North Pacific). Maximum dive depth showed a peak during winter (January to March) for both species, but it was deeper for TBMUs (100 to 120m) than COMUs (80 to 100m). COMUs performed benthic dives more frequently (up to 35% of total dive) in the Bering Sea Shelf areas compared with TBMUs (less than 15%). These inter-specific differences in habitat use were consistent between the two study years. These results suggest that 1) the two species segregate their foraging habitat during winter in neighboring areas as follows: more benthic layer on the Bering Sea Shelf for COMUs and more pelagic/epipelagic layer in the oceanic areas for TBMUs, and that 2) their foraging habitat overlap most during autumn on the Bering Sea Shelf. The segregation in the wintering habitat between the two species may partly explain their different responses to the circumpolar marine environmental change (Irons et al. 2008). Nonetheless, habitat overlap between the species during autumn highlights the ecological importance of shelf domain of the southeastern Bering Sea (Alabia et al. 2021).

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# Uphill energy transfer mechanism for photosynthesis in the Antarctic alga

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*Prasiola crispa*, an aerial green alga, forms large layered colonies under the severe terrestrial conditions of Antarctica, which include severe cold, drought, and strong sunlight in summer season. As a result of these conditions, the surface cells of a colony faces high risk of photodamage. On the other hand, cells of deeper layer can escape from photodamage at the sacrifice of photosynthetically active radiation except far-red light (>700 nm) (Figure 1). Our present analysis showed efficient photosynthesis activation with far-red light was achieved in the cells of *P. crispa*, whereas chlorophyll *a*-based photosystems need excitation energy of visible light (~680 nm) to split water molecules (Kosugi et al. 2020). The result suggested that *P. crispa* achieves effective photosynthesis by low energy far-red light for photosystem II excitation by “uphill” excitation energy transfer. Here, we identified a novel far-red light-harvesting complex of photosystem II in *P. crispa*, Pc-frLHC (*Prasiola crispa* far-red light-harvesting Chl-binding protein complex), and proposed a molecular mechanism of uphill excitation energy transfer based on its cryogenic electron-microscopy structure. While Pc-frLHC is associated with photosystem II, it is evolutionarily related to the light-harvesting complex of photosystem I. Pc-frLHC forms a ring-shaped homo-undecamer in which all chlorophyll *a* molecules are energetically connected and contains chlorophyll *a* trimers. It seems that the trimers are long-wavelength-absorbing chlorophylls for far-red light at 708 nm, and further absorbance extension is accomplished by Davydov-splitting in dimeric chlorophylls. The chlorophyll network should enable a highly efficient entropy-driven uphill excitation energy transfer using far-red light up to 725 nm.

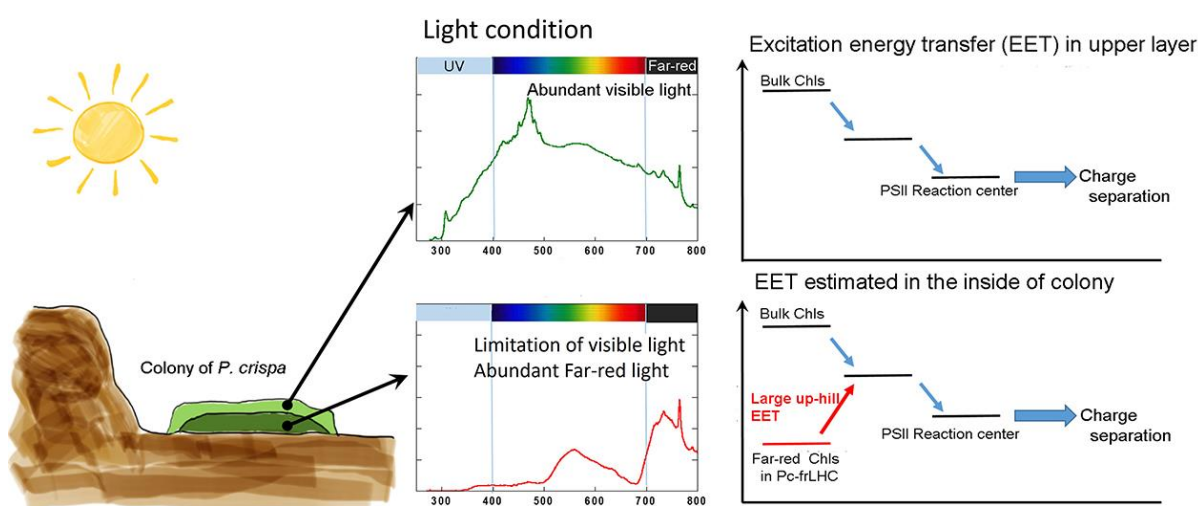


Figure 1. Light conditions and estimated excitation energy transfer pathways in upper layer and inside of colony of *P. crispa*.

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# Inter-specific and intra-specific competition of two sympatrically breeding seabirds, Chinstrap and Gentoo penguins, at two neighboring colonies

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Theory predicts that sympatric predators compete for food under conditions of limited resources. Competition would occur even within the same species, between neighboring populations, because of overlapping foraging habits. Thus, neighboring populations of the same species are hypothesized to face strong competition. To test the hypothesis that intraspecific competition is more intense than inter-specific competition owing to a lack of niche partitioning, we estimated the foraging area and diving depths of two colonial seabird species at two neighboring colonies. Using GPS and time-depth recorders, we tracked foraging space use of sympatric breeding Chinstrap and Gentoo penguins at Ardley Island (AI) and Narębski Point (NP) at King George Island, Antarctica. GPS tracks showed that there was a larger overlap in the foraging areas between the two species than within each species. In dive parameters, Gentoo penguins performed deeper and longer dives than Chinstrap penguins at the same colonies. At the colony level, Gentoo penguins from NP undertook deeper and longer dives than those at AI, whereas Chinstrap penguins did not show such intraspecific differences in dives. Stable isotope analysis of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  isotopes in blood demonstrated both inter- and intra-specific differences. Both species of penguin at AI exhibited higher  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values than those at NP, and in both locations, Gentoo penguins had higher  $\delta^{13}\text{C}$  and lower  $\delta^{15}\text{N}$  values than Chinstrap penguins. Isotopic niches showed that there were lower inter-specific overlaps than intra-specific overlaps. This suggests that, despite the low intra-specific spatial overlap, diets of conspecifics from different colonies remained more similar, resulting in the higher isotopic niche overlaps. Collectively, our results support the hypothesis that intra-specific competition is higher than inter-specific competition, leading to spatial segregation of the neighboring populations of the same species.

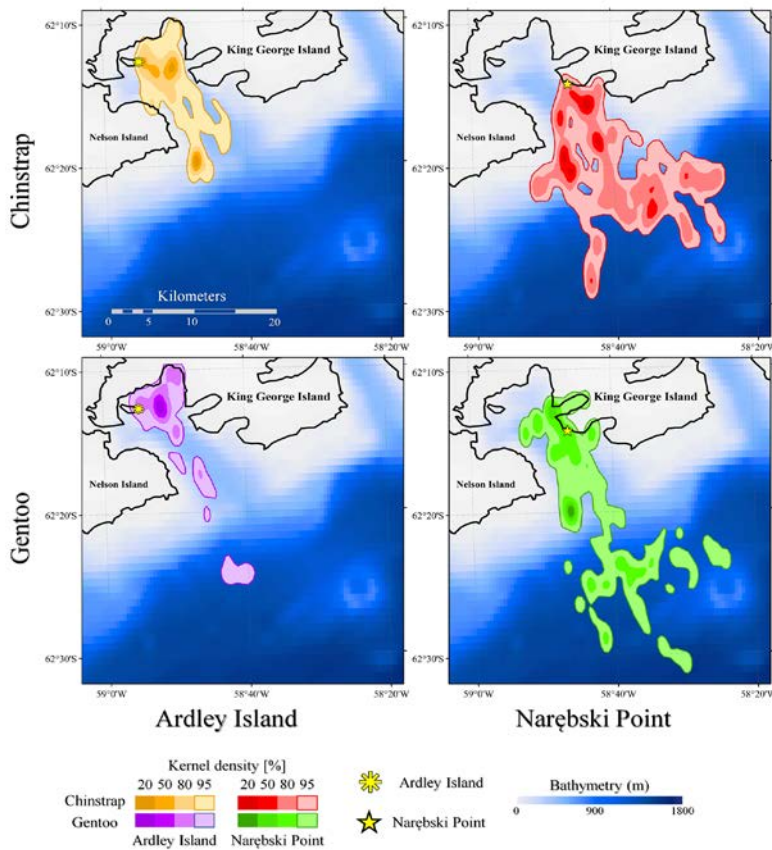


Figure 1. Foraging areas compared between species (Chinstrap vs. Gentoo penguins) and between colonies (Ardley Island vs. Narębski Point)

# Seasonal variability in vertical distribution of zooplankton in the Antarctic seasonal ice zone

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Zooplankton plays important roles as secondary producers in marine food web and are drivers of biological carbon pump in the Southern Ocean ecosystem. They undertake a daily/seasonal vertical migration (DVM/SVM), enhancing an export of organic materials from surface productive layer to deeper layers. These functions of zooplankton have been studied by ship-based snap-shot observations by net tows and ship-mounted echosounder or time-series observation using a mooring system equipped with an echosounder. Our knowledge on time-series variabilities in their vertical distribution has been limited so far due to difficulties to ensure higher temporal and spatial resolutions especially in the surface layers.

We then conducted two drifter observations with various sensors and an upward-looking acoustic doppler current profiler (ADCP) in the seasonal ice zone during austral summer. The first observation was conducted in around the center of Vincennes eddy (63.5°S, 110°E) off Wilkes Land (East Antarctica) in the training vessel *Umitaka-maru* cruise from 14-19 January, 2019. In this site, a subsurface chlorophyll maximum (SCM) was developed with a higher chlorophyll *a* concentration ( $>1.5 \mu\text{g L}^{-1}$  at 30-40 m depth). The second observation was conducted from 9 December 2019 to 16 February 2020 during ice braker *Shirase* cruise. A drifter was deployed at 64.26°S, 115.98°E, where sea ice concentration was higher than 90%, and retrieved at 64.27°S, 104.83°E. In both observations, the ADCP was deployed at around 70 m depth; raw data obtained (20 vertical bins) were converted to volume backscattering strength (SV). We used ten minutes mean SV for analyzing temporal changes in vertical distribution patterns of relative zooplankton biomass.

The first observation found two scattering layers. The shallower one located around the SCM and the deeper one was found only midnight (Fig. 1a), suggesting that there were two different types of zooplankton in migration pattern. Species compositions of the two communities will be elucidated by further analysis of net samples during the drifting period. During the second observation, the highest SV was recorded at subsurface layer through the observation period (Fig. 1b). SV near the surface layer of 10-20 m depth was relatively higher when sea ice concentration rapidly decreased (from end of December to early January). These observations demonstrated that zooplankton distribution was becoming deeper after sea ice melting as well as their diel vertical migration pattern was being distinctive.

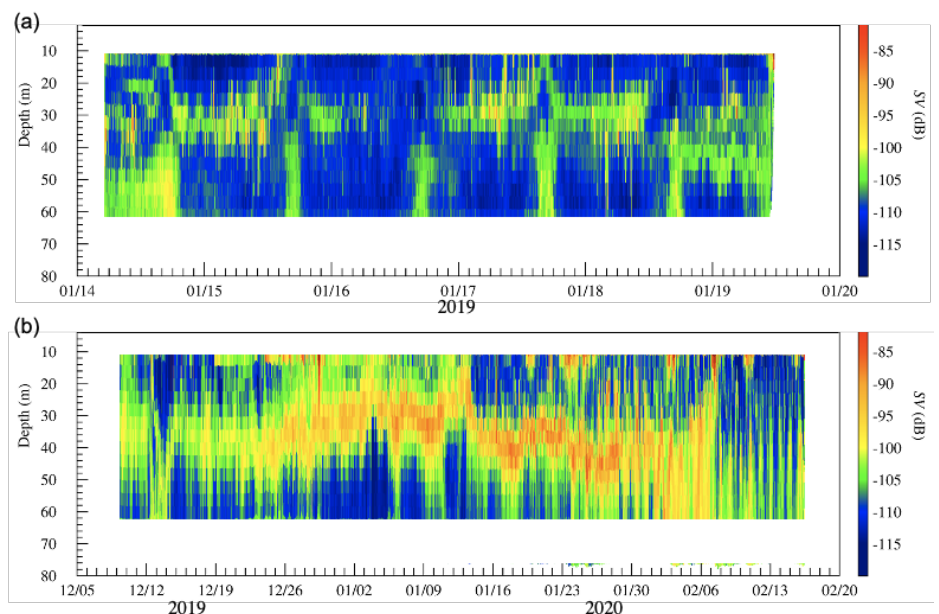


Fig. 1. Temporal changes in vertical profile of volume backscattering strength (SV) during drifter observations in January 2019 (upper) and from December 2019 to February 2020 (bottom) off Wilkes Land, East Antarctica.

# Identification and phylogenetic analysis of fecal pellet-like dinoflagellates in the seasonal ice zone of the Southern Ocean

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## Introduction

Ice edge phytoplankton blooms have important impacts on the lower trophic level of the food web and on carbon transfer in the seasonal ice zone of the Southern Ocean (Deppeler and Davidson, 2017). Sea ice melting from spring to summer releases iron and phytoplankton, which are incorporated into sea ice, causing occasional phytoplankton blooms in the surface layer. Ice melting decreases salinity, leading to the formation of a shallow surface mixed layer. As a result, vertical material cycling between the surface and deeper layers is suppressed. Large amounts of phytoplankton are grazed by zooplankton or consumed by bacteria. The remaining zooplankton sinks to the deeper layer as particle organic carbon (POC), which includes zooplankton faecal pellets, marine snow, and phytodetritus (Turner, 2015). This vertical carbon transport through biological processes is referred to as a biological pump. We sampled sinking particles using a sediment tarp off Wilkes Land, East Antarctica, during austral summer in 2016 to determine the composition of sinking particles, which is assumed to vary with the timing of sea ice melt. In this preliminary study, large numbers of faecal pellet (FP)-like dinoflagellates (FLD) containing phytoplankton cells were found in samples collected at a depth of 50 m. FLD-derived carbon accounted for 40% of POC fluxes within the total FP, suggesting that FLD is a key organism of the biological pump and is essential for understanding the lower trophic levels of food webs in the seasonal ice zone of the Southern Ocean. To examine the species composition of FLD, we examined their taxonomic and phylogenetic status using 18S rRNA gene sequences.

## Material and Methods

We deployed sediment traps at depths of 60 and 150 m (64°16'S, 116°58'E) from 10 December, 2019, to 20 January, 2020. Particle samples were fixed in neutral Lugol's solution at a final concentration of 5–20% and stored at 4°C until analysis. A total of 66 FLD cells (60 m: 53 cells; 150 m: 13 cells) were randomly collected based on morphology and used for size measurements and DNA analyses. Total DNA extracted from each cell was subjected to polymerase chain reaction (PCR) amplification of the partial 18S rRNA gene using a *Gyrodinium*-specific primer set. The PCR products were sequenced and similarities of the 18S rRNA sequences of FLDs to those of validly published species were calculated using the BLAST program. The phylogenetic tree was reconstructed using published sequences of *Gyrodinium* species.

## Results and Discussion

We identified 50 of 53 cells sampled at a depth of 60 m as *Gyrodinium rubrum*, with 99.7–99.9% sequence similarity using the 18S rRNA gene sequence, and 11 of 13 cells sampled at a depth of 150 m as *Gyrodinium heterogrammum*, with 99.7–99.9% similarity. We detected clear differences in the cell size distribution of FLDs between the two species (Fig. 1). Phylogenetic analysis showed that the species were clustered with *G. rubrum* and *G. heterogrammum*, respectively (Fig. 2). Previous studies detected *G. rubrum* in the water column and as sinking particles in Ryder Bay, Kerguelen Plateau and in the Scotia Sea in the Southern Ocean using high-throughput sequencing of environmental DNA (Piquet et al., 2011; Georges et al., 2014; Duret et al., 2019). *Gyrodinium heterogrammum* or closely related species were also detected in the Amundsen Sea, Polynya (Swalethorp et al., 2019). *Gyrodinium* species have been reported to have a cell length of ~24 to ~140 µm (Takano and Horigoshi, 2004; Larsen, 1994). However, the FLD cells observed in this study were significantly larger than previously reported; therefore, we infer that these cells were likely to have been hypertrophied through stuffing with food items. Such cells presumably sank earlier than those of normal size, suggesting that *G. rubrum* and *G. heterogrammum* contribute to the biological pump in the seasonal ice zone of the Southern Ocean.



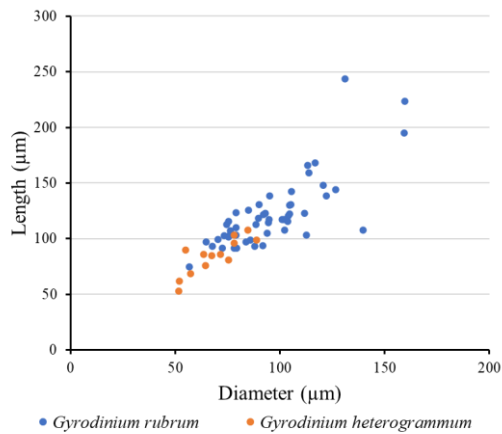


Fig. 1. Scatter plot of diameter and length of faecal pellet-like dinoflagellates (FLD). Blue and orange dots indicate *Gyrodinium*

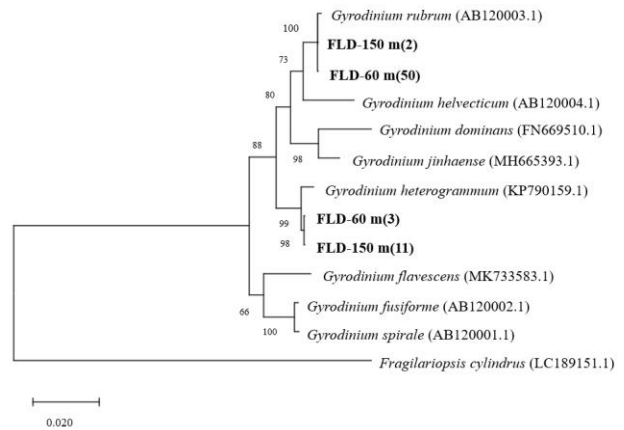


Fig. 2. Neighbour-joining phylogenetic tree based on 18S rRNA gene sequences. GenBank accession numbers are provided in parentheses. Bar indicates 0.02 substitutions per nucleotide position. Numbers at nodes indicate bootstrap probabilities based on 1,000 resamples.

## Acknowledgment

We would like to thank the deceased Professor Tsuneo Odate for his scientific and financial contributions.

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# Morphological development of larval and juvenile Antarctic paralepidid fish *Notolepis coatsi* in Indian Ocean sector of the Southern Ocean

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Mesopelagic fishes, which have an enormous biomass in the world oceans, have a role to link lower trophic levels to higher animals. In the high Antarctic zone, *Notolepis coatsi* is a common mesopelagic species. Although information on geographical and vertical distributions of the species is accumulating, morphological development of early stages that is essential to understand early life history is scarcely studied. We described early morphological development of feeding- and swimming-related characteristics of *N. coatsi*.

Fish were sampled by a ring net and open-close frame trawl net (MOHT, Matsuda-Oozeki-Hu Trawl) in January 2019 off Vincennes Bay, East Antarctica. Seventy-four larvae and juveniles were measured for six body parts. Forty-two specimens were cleared and stained to observe osteological development. In order to verify behavioral development with the morphological development, gut content were analyzed for thirty-two larvae.

Apparent changes were observed in following developmental characteristics between 30- and 35-mm body length (BL). During this period, flexion points were observed in allometric growth of preanal length, eye diameter and snout length. With respect to the feeding-related characteristics, upper and lower jaw teeth, upper pharyngeal teeth and palatine teeth increased rapidly by 35 mm BL. Fin rays occurred and their number increase in 30-35 mm BL. Vertebrae were first observed and osteological elements of pectoral and caudal fins were completed in number at ca. 35 mm BL. Although the most of gut content were impossible to be identified to species/genus level due to being fragmented to small pieces, copepods (frequency 100%), ostracods (7.1%) and crustacean pieces (100%) were found in guts. Crustacean individual number, which were estimated from the number of mandibles in crustacean pieces, increased gradually by ca.30 mm BL, and increase rapidly thereafter.

Morphological observations revealed that feeding- and swimming-related characteristics of *N. coatsi* advance the developmental stage since 30-35 mm BL. However, vertebral number did not complete by 35 mm BL, although all fin-elements occurred. These results indicate that it is hardly to distinguish between larval and juvenile stages using definitions applied generally. Rapid increase of gut content was observed during the same period. In addition, a previous study demonstrates that this species undertakes ontogenetic vertical changes from epipelagic to mesopelagic layers during this period. These facts suggests that this species almost reaches juvenile stages functionally and changes their ecology by ca.35 mm BL.

## Seabird-fishery interactions revealed by bird-borne GPS and camera loggers: Interspecific comparisons of two albatross species

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Albatrosses and petrels are often attracted to fishing vessels to feed on fishery discards and bait. Large numbers of seabirds are killed by drowning in longline fisheries after caught on baited hooks and in trawl fisheries after entangled in nets. However, little is known about the spatial-temporal overlap between individual birds and vessels, and behaviors in relation to vessels. To examine these, we attached both GPS- and camera- loggers on two North Pacific albatross species; 15 black-footed albatrosses (*Phoebastria nigripes*) breeding on Torishima, Japan and 20 Laysan albatrosses (*P. immutabilis*) breeding on Oahu, Hawaii, during the early chick-rearing periods. Black-footed albatrosses concentrated mainly along the Izu chaine and some birds used off Fukushima 600 km away from the colony. Two birds encountered pole and line fishing vessels at the north of Torishima, but did not follow them. Five Laysan albatrosses from Hawaii encountered fishing vessels in subtropical waters close to their colony although they travelled over the North Pacific Transition Zones up to 800 km north of the colony. With clear images of these fishing vessels, we identified as Hawaiian tuna/swordfish longliners. These Laysan albatrosses detected the longliners at an average distance of 5.4 km and followed them up to 5.3 hours. Our study indicates the usefulness of combining bird-borne GPS- and camera-loggers for studying fine-scale seabird-fishery interactions.

# **Optimization of carbazole degradation by Antarctic cold-tolerant bacterium strain BS1**

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Carbazole (CAR) is a heterocyclic compound that is increasing demand as industrial raw material for pharmaceuticals and plastics industries. CAR has been detected in petroleum-contaminated soil and seawater, along with their decomposition products. In recent years, it has also been detected in Antarctica due to increasing human activities. CAR is persistent in nature and has been confirmed to be carcinogenic and mutagenic, hence it is necessary to remove CAR from the environment. However physical methods are costly and impractical. Bioremediation approaches should be used to remove CAR pollution as it is low cost to implement while being low impact on the environment. A number of studies have reported on the degradation of CAR using bacteria to be effective at relatively warm temperatures, but information on CAR degrading bacteria from cold environments is limited, hence there is a need to study and understand CAR degrading bacteria in such environments. Furthermore, the Antarctic Treaty restricts the introduction of organisms from the outside world, bioremediation for Antarctic region can only be conducted using native microbes. Cold-tolerant bacterial strain BS1 has been isolated from Antarctic soil and its various properties have been studied. In this study, we aim to optimize the degradation efficiency of the Antarctic cold-tolerant bacterium BS1 strain for practical use. Multiple culture parameters for optimization assessed were temperature, pH, salt concentration, iron concentration, and nitrogen concentration for CAR degradation efficiency. As a result of the experiment, the optimum culture conditions were pH 6~8, temperature 20~25°C, salt concentration 0.5 g/100 ml, and while nitrogen concentration had little effect. Future studies of strains BS1 will enhance our understanding and increase our knowledge on developing bioremediation measures for Antarctica and other cold climate environments.

# Food management for Antarctic wintering team

## How long vegetables stay fresh, How to Preserve Vegetables, Hydroponics for Vegetables

Ritsuna Noguchi<sup>1</sup>, Daiki Maehara<sup>1</sup>, Chisato Niwaike<sup>1</sup>, Toru Koyama<sup>2</sup>, Satoshi Imura<sup>2</sup>

<sup>1</sup>*Department of Health and Dietetics, Faculty of Health and Medical Science, Teikyo Heisei University,*

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【背景】南極での食事は以下のような特殊性を持つ。①南極到着後食糧の補給はなく、一度持ち込んだ食糧のみで食生活を維持する。②保存性・貯蔵性に乏しい食品(野菜・果物など)は、食せない時期(越冬後半)がある。③メニューは調理隊員が決定し、個人による選択の余地はない。④ゴミ減量化、排水制限等の制約がある。⑤南極生活での数少ない楽しみの1つであり、隊員同士の紐帯の源である。こうした特殊性は、災害時の食事と共通点が多い。南極調理隊員による食糧の選択と配分、食べられない食品の代替メニュー等は、災害用備蓄や災害時の食事に活用できると思われる。さらに、隔離・閉鎖された空間である南極での食事が、隊員にとってどのような存在なのか、調理隊員は何を心がけているのかを知ることは、災害時の食事を単なる栄養補給ではなく、被災者に寄り添う食として捉える上で重要であると考えられる。

【目的】本研究の目的は、南極越冬隊の食事の特徴を明らかにし、災害食への応用を検討することである。本発表では、南極での食材管理(野菜の保存期間・保存方法)と野菜栽培について報告する。

【方法】第1次隊(1956-58年)から第60次隊(2018-20年)までの日本南極地域観測隊報告書を対象とし、野菜の保存期間と方法、野菜栽培に関する記載を記述的に分析した。

【結果】LL牛乳は1年、卵は8ヶ月(生食4ヶ月)使用していた。果物は生食、冷凍、コンポートで食していた。野菜は、定期的に外皮をむいて保存していた(白菜・キャベツオペレーション)。国産野菜と途中で積載するオーストラリア野菜を比較すると、国産野菜の方が長持ちしていた。また、越冬後半の野菜不足を乗り切るため、もやし・胡瓜・貝割れ・トマトなどを水耕栽培していた。

## **Features of the Antarctic wintering team's meals (menus) -Breakfast, Lunch, Dinner, Snacks, Special meals (Party food), etc-**

Ritsuna Noguchi<sup>1</sup>, Kazuki Hayakawa<sup>1</sup>, Momoka Matsuda<sup>1</sup>, Keisuke Motoyanagi<sup>1</sup>, Keita Yamaguchi<sup>1</sup>, Toru Koyama<sup>2</sup>,  
Satoshi Imura<sup>2</sup>

<sup>1</sup>*Department of Health and Dietetics, Faculty of Health and Medical Science, Teikyo Heisei University,*

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**【目的】**本研究の目的は、南極越冬隊の食事の特徴を明らかにし、災害食への応用を検討することである。本発表では、南極での献立の特徴について報告する。

**【方法】**第1次隊(1956-58年)から第60次隊(2018-20年)までの日本南極地域観測隊報告書を対象とし、献立に関する記載を記述的に分析した。

**【結果】**朝食はバイキング、昼食は短時間で食べられる麺類か丼もの、夕食は定食スタイルであった。お菓子は、持参した分が最後までなくならないよう、調理隊員が管理して配分していた。BARが定期的に開かれ、お酒が自由に飲めるようになっていた。さらに、曜日感覚を維持するために毎週金曜日はカレーとする、季節感感覚を維持するために日本の季節に合わせた特別食を実施する(7月に流しそうめん等)などの工夫がされていた。

# Animal-borne video observations of fish and squid capture by northern elephant seals in the mesopelagic zone

Atsuya Ogata<sup>1</sup>, Akinori Takahashi<sup>1,2</sup>, Taiki Adachi<sup>2,3</sup>, Yasuhiko Naito<sup>2</sup>, Rachel Holser<sup>4</sup>, Arina Favilla<sup>4</sup>, Daniel Costa<sup>4</sup>

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<sup>4</sup>*University of California Santa Cruz, USA*

The mesopelagic zone (200-1000 m) in the ocean holds large fish biomass and is an important foraging habitat for deep-diving marine mammals. Examining how environmental parameters (e.g., dissolved oxygen concentration) affect predators' foraging success is required to better understand the ecological effects of ongoing environmental changes in the mesopelagic zone. It has been hypothesized that deep-diving northern elephant seals may feed efficiently on sluggish fish in mesopelagic oxygen minimum zones (Naito et al., 2017 *Ecol Evol*). However, evidence of easier prey captures in low-oxygenated waters is lacking. In this study, we investigated the 3D spatial distribution of prey capture events and the prey's motion during captures in relation to environmental parameters that directly affect ectothermic prey (dissolved oxygen concentration and seawater temperature). We analyzed video footage obtained from head- and jaw-mounted video cameras attached to 15 female northern elephant seals (Yoshino et al. 2020 *J Exp Biol*) during their post-breeding foraging trips. The videos recorded 697 successful prey capture events between depths of 500-1000 m in the northeastern Pacific. For each prey capture location and depth, we extracted dissolved oxygen concentrations (DO) and seawater temperature (T) from the Global Ocean Biogeochemistry Hindcast dataset (provided by Copernicus Marine Service). From the 70 fish and 20 cephalopods that were clearly visible from the video footage, we identified 6 and 5 taxonomic groups, respectively. Gadidae and Merlucciidae (n = 29 events) were preyed upon mainly in the California Current region at depths of 400-600 m (DO: 10-40 mmol m<sup>-3</sup>; T: 5-7 °C). Myctophidae and Microstomatidae (n = 28 events) were preyed upon offshore in the Eastern North Pacific at 400-900m depths (DO: 30-140 mmol m<sup>-3</sup>; T: 3-7°C). Macrouridae and Scorpaenidae (n = 13 events) were preyed upon near the seafloor in the Gulf of Alaska at 450-700 m depths (DO: 50-80 mmol m<sup>-3</sup>; T: 4-5°C). Unlike the fish taxonomic groups, there were no clear patterns among cephalopod taxonomic groups in the spatial and depth ranges of prey capture events as well as environmental parameters ((DO: 20-173 mmol m<sup>-3</sup>; T: 3-6.2°C). The motion of fish during captures appeared to differ among taxonomic groups: Myctophidae and Microstomatidae tended to move actively before being captured, but Gadidae and Merlucciidae tended to be motionless until the capture actions of the seals. However, the presence/absence of escape motion by fish did not vary across the ranges of environmental dissolved oxygen concentrations and seawater temperatures. These results suggest that 1) female northern elephant seals feed on different types of fish depending on biogeographic regions, and 2) dissolved oxygen environments appear not to affect the escape motion of fish prey within each taxonomic group.

## Microfungi associated with Antarctic and Arctic moss profiles

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Mosses are major components of terrestrial ecosystems of the Arctic and Antarctica. The accumulated live, senescent, and dead moss tissues harbor a variety of fungi and other organisms that drive carbon and nutrient dynamics. Thus, examining vertical patterns of fungal assemblages within moss profiles consisting of live, senescent, and dead tissues in different stages of decomposition sheds light on the functional roles of fungi in the decomposition of moss tissues. The purposes of the present study were to examine the abundance, diversity, and succession of fungi and their relationship with chemical changes in moss tissues for two moss species (*Racomitrium lanuginosum* and *Hylocomium splendens*) in the Canadian Arctic (80°50' N) and one moss species (*Bryum pseudotriquetrum*) in Lutzow-Holm Bay area, east Antarctica (69°00' S) (Osono et al. 2019). Moss blocks were collected and divided into 4 to 6 layers according to the color and texture to analyze fungal assemblages, fungal succession, and hyphal length in relation to chemical changes in decomposing moss tissues. A total of 19, 18, and 14 fungal species were isolated from moss tissues of *R. lanuginosum*, *H. splendens*, and *B. pseudotriquetrum*, respectively, with *Pseudogymnoascus pannorum* being found in both the Arctic and Antarctica and was considered to be a cosmopolitan. Turnover of fungal species was found within the two arctic moss profiles, indicating the successional changes in fungal assemblages during decomposition of moss tissues, whereas in *B. pseudotriquetrum*, the frequencies of occurrence of two major fungi *Phoma herbarum* and *P. pannorum* increased downward without turnover of the major fungal species. Mean values of total hyphal lengths measured by direct observation using an agar film method were 1,164 m/g in *R. lanuginosum*, 4,446 m/g in *H. splendens*, and 3,224 m/g in *B. pseudotriquetrum*. Total hyphal length increased from the uppermost green layers toward the lower layers within both of the arctic moss profiles, whereas no significant changes were found for total hyphal length among vertical layers of Antarctic moss. The amount of holocellulose relative to recalcitrant compounds classified as acid unhydrolyzable residues (AUR) decreased downward from the upper to the lower layers of two arctic moss profiles, whereas in the profile of Antarctic moss no significant change was found for the relative amount of holocellulose and AUR during decomposition. The carbon to nitrogen (C/N) ratio of moss tissues decreased during decomposition of all three moss profiles, leading to relative accumulation of nitrogen at the lower layers. The vertical patterns of abundance and richness of fungi and chemical changes in the Arctic and Antarctic moss profiles exhibited interesting similarities and contrasts; that is, the hyphal length and the fungal richness were generally similar, while marked differences were found for fungal succession and decomposition of organic chemical components between these moss profiles.

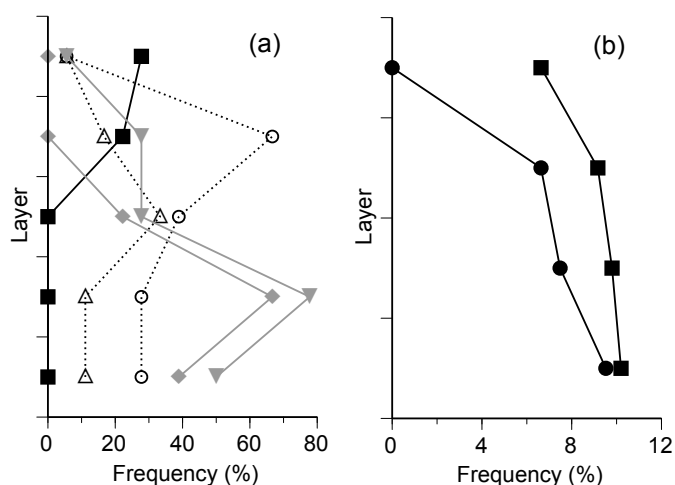


Figure 1. Patterns of fungal succession in Arctic and Antarctic moss profiles. The profiles were divided into five and four layers in the Arctic and Antarctica, respectively, from the upper surface layer to the lower bottom layer. (a) *Racomitrium lanuginosum* in the Arctic. Black squares and solid line, *Cladosporium herbarum*; open squares and dotted line, *Penicillium* sp.1; open triangles and dotted line, *Umbelopsis ramanniana*; gray triangles and gray line, *Pseudogymnoascus pannorum*; gray diamonds and gray line, *Absidia cylindrospora*. (b) *Bryum pseudotriquetrum* in Antarctica. Black squares and solid line, *Phoma herbarum*; black circles and solid line, *Pseudogymnoascus pannorum*.

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## Device of marine educational workshop kit, “Let’s observe the Diversity of Marine Plankton World”

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**Abstract:** In recent years, a learning style with observation and experiment has been recommended for active learning at schools. The partnership programs between schools and museums are becoming more important. National Institute of Polar Research is promoting the improvement and increase of production of resin embedded marine plankton specimens sets for marine education. At 2020, we devised a "learning material set" that includes resin-embedded specimens, images and video data that capture the colors and movements of living organisms in the actual field, and introduce for school teachers and hearing their needs. In this year, we device of marine educational workshop kit, “Let’s observe the Diversity of Marine Plankton World”, consist of resin embedded specimens, observation tools, booklet, worksheets, movies for teacher and participant and workshop programs (in Japanese) (Fig. 1). The kit is designed mainly for junior high school students and can be used for remote workshops to meet the needs of COVID-19 pandemic. In order to make more useful programs, we are asking for school teachers to try this kit and find how to use. We will start lending the kit at Nov. 2021. We hope to use the kit by all teachers not have chances of marine education until now.

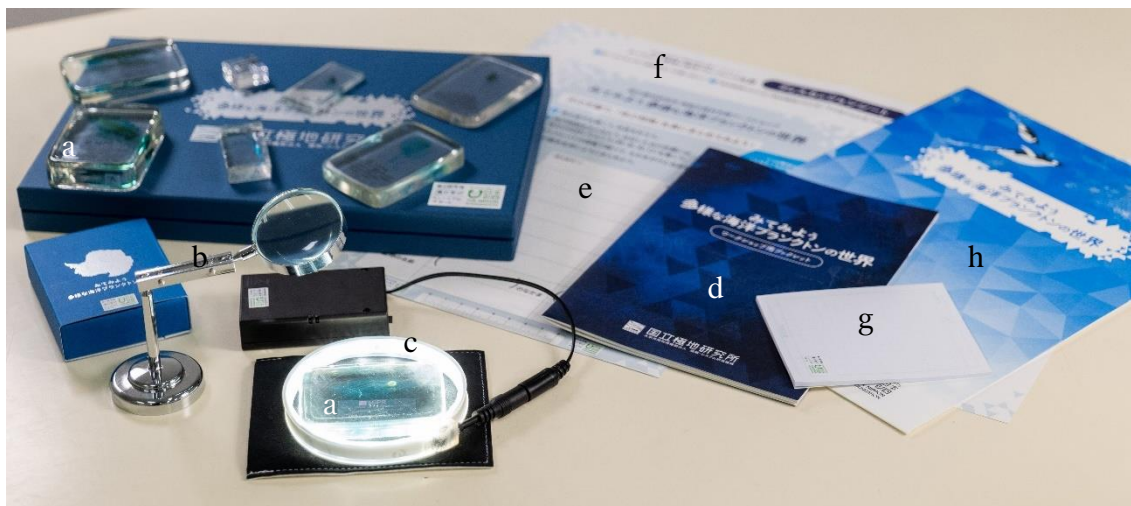


Fig.1 Marine educational workshop kit, “Let’s observe the Diversity of Marine Plankton World”.

(a) resin embedded specimens, (b) stand loupe, (c) LED ring light, (d) booklet, (e) worksheet, (f) discussion sheet, (g) sticky notes for sketch, (h) paper folder.

# Microbial community structures of sinking particles collected by using gel sediment traps in the Indian sector of the Southern Ocean during austral summer

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<sup>3</sup>The Graduate University for Advanced Studies

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Sinking particles, an important contributor to biological carbon pump, consist of various types of particles. Export efficiencies and sinking speeds are different among types of sinking particles, for example, phytodetritus and fecal pellets. In the Southern Ocean, the contribution of fecal pellets to carbon flux is expected to be high because of the high abundance of the large zooplankton, such as salps, krills and pteropods which excrete fast-sinking large fecal pellets. However, it is difficult to interpret their sinking process by microscopic analysis, because those particles are morphologically similar. Knowledge on microbial community structures of sinking particles is still limited, but some studies reported that microbial community structures of sinking particles changed with depths. Thus, microbial community structures are considered to reflect the extent of degradation of sinking particles. However, details of microbial community structures of individual sinking particles have not yet been known. This study analyzed the microbial community structures of individual sinking particles collected by gel sediment traps in the Indian sector of the Southern Ocean and examined relations between each type of sinking particles and depth.

Sinking particle were sampled from a station (63.5°S, 110°E) during the Southern Ocean cruise of the training vessel *Umitaka Maru* of Tokyo University of Marine Science and Technology. The drifter array consisted of gel-filled sediment traps deployed at 50, 200 and 500 m depths. Traps were left to drift for 24 h (17 January 2019 to 18 January 2019). After recovery, each gel was preserved at <-60°C until analysis. Sinking particles were classified into six types, large-size cylindrical fecal pellet (LCFP), medium-size cylindrical fecal pellet (MCFP), small-size cylindrical fecal pellet (SCFP), oval fecal pellet (OFP), fecal aggregate (FA) and aggregate (AG). To assess the microbial community structure of the sinking particles, the 16S rRNA gene V3-V4 region was amplified and the amplicons were sequenced by a high throughput sequencer. Bioinformatics analysis of the sequence data were performed using the Qiime2 software package.

Microbial community structures of the sinking particles mainly consisted of Gammaproteobacteria and Bacteroidia. Particularly, those of LCFP mainly consisted of Bacteroidia. Principal coordinate analysis (PCoA) using weighted UniFrac distances based on the composition and phylogenetic distances of microbial communities resulted in clustering by depth and/or types of sinking particles. The results of the PCoA using weighted UniFrac distances showed that LCFP were not significantly different between 50 to 500 m depth samples (PERMANOVA, FDR  $q < 0.05$ ). On the other hand, MCFP, SCFP and OFP were significantly different among some depths (PERMANOVA, FDR  $q < 0.05$ ). These results suggested that while MCFP, SCFP and OFP were consumed and repackaged during sinking process, LCFP, sinking faster than those particles, of 500m depth sank to this depth directly without degradation and fragmentation. Microbial community structures are considered to useful for interpretation of export process of sinking particles.

## Isolation of Polylactic acid Degradable Bacterium strain N-3 from Antarctic Soil

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Plastic is widely used in our daily life. However, it is difficult to be degraded in the natural environment and remains persistent for extended period of time. In this environmental problem, ecologically friendly bioplastics have been attracting a lot of attention as these categories of plastics can be degraded by microorganisms. Polylactic acid (PLA) is a type of bioplastic, which is synthesized from biological resources. In recent years, several strains of psychrophilic bacteria had been reported to possess the ability to degrade different types of plastics<sup>1</sup>. The objective of this study is to isolate bacterial strains capable of degrading PLA from Antarctic soil and to determine its properties and its ability to degrade PLA films.

Antarctic soil from British base on Signy Island was added to a medium containing inorganic salts and PLA and incubated by shaking. As a result of enrichment culture, Gram-negative rod-shaped bacterium with catalase activity was successfully isolated. This isolate was designed as strain N-3. From the 16S rRNA sequence analysis, this isolate showed high similarity to *Massilia (Naxibacter) alkalitolerans*. To estimate the biodegradation rate of PLA, isolates was added to a medium containing only inorganic salts and PLA film and incubated for 4 weeks. The number of bacteria increased after the incubation, as shown by the CFU counting method and the measurement of protein concentration. In addition, the weight of the film decreased by 6.4% compared to that before incubation. These results indicated that this isolates in the Antarctic soil have the ability to degrade PLA.

### References

1) Atanasova, N.; Stoitsova, S.; Paunova-Krasteva, T.; Kambourova, M. Plastic Degradation by Extremophilic Bacteria. *Int. J. Mol. Sci.* **2021**, *22*, 5610. <https://doi.org/10.3390/ijms22115610>

# Distribution depth of some pelagic and mesopelagic fishes in the Southern Ocean inferred from their eye and retinal morphology

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## Background

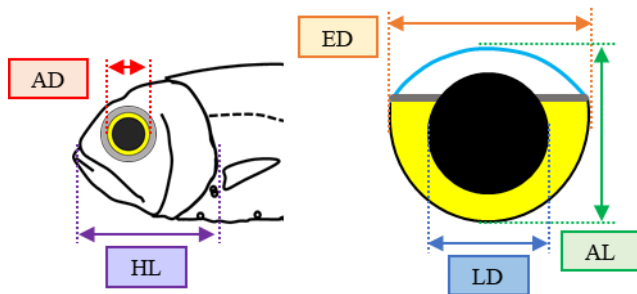
Distribution depth of fish species can be inferred from towing depth of net. However, some species are occasionally caught shallower depths than known distribution depth which could related to their vertically diurnal migration. Hence, the inference has some limitations. Morphological and histological features in eye and retina of fish are known reflecting distribution depth and feeding ecology (e.g., direction of visual interest, detection of prey organism and difference in diurnal and nocturnal behavior). Because these features change as they grow, they could be used to infer ontogenetic shift in distribution depth. In this study, we conducted morphological and histological investigation on the eyes and retinas of eight fish species sampled by Rectangular Mid-Water Trawl (RMT) in the eastern Indian sector of the Southern Ocean. Their visual sensitivity are compared based on the results.

## Materials and Methods

Eight fish species (*Bathylagus sp.*, Argentiniiformes; *Notolepis coatsi*, Aulopiformes; *Electrona antarctica*, *Gymnoscopelus braueri*, *Krefflichthys anderssoni*, *Protomyctophum bolini*, Myctophiformes; *Chaenodraco wilsoni*, *Pleuragramma antarcticum*, Perciformes) sampled mainly shallower than 200 m water depth by the RMT were used in this study. The survey was conducted by R/V *Kaiyo-Maru* (Fisheries Agency, Japan) in the eastern Indian sector of the Southern Ocean in austral summer of 2018/19. Immediately after the hauls, their eyes were flash-photographed with a digital camera to confirm presence of tapetum lucidum. The specimens were then fixed in 10% formalin solution or Bouin's fixative solution. The head length (HL, mm), aperture diameter of eye (AD, mm), eyeball diameter (ED, mm), axial length (AL, mm), and lens diameter (LD, mm) were measured under a stereomicroscope at the laboratory (Fig 1). After we dissected the eyes, flattened retina was cut in small pieces, embedded in resin, and semi-thin radial sections were cut at 1  $\mu$ m thickness. Further observations were carried out under a light microscope, and development of the pigment epithelial layer and the type of photoreceptor cells were examined. Using the measured values of the eye morphology, the ratios characterizing development of the visual sensitivity were calculated.

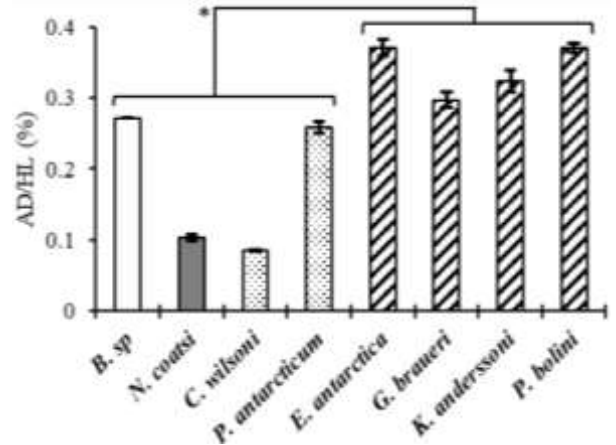
## Results and Discussion

There was a significant difference ( $p < 0.05$ , *U*-test) between myctophids and other species in AD/HL (%) (Fig 2). It could be associated with difference in distribution depth. No significant difference was observed for other ratios. Retinal pigment epithelium of the myctophids were undeveloped, and no cone photoreceptors were observed in their retina (Fig 3). Their eyes could be specialized for dim-light environments deeper than 200 m. The thickness of the rod photoreceptor layer to the entire retina was thicker in *P. bolini* and thinner in *G. braueri*. Among four myctophids, tapetum lucidum was absence only in *G. braueri*, suggesting that this species could be inferior in scotopic vision to other species. Three species belonging to other families have cone cells, being capable of photopic vision at the depths shallower than 200 m where sufficient sun light is available. Because *N. coatsi* has tapetum, this species could adopt photopic vision by reflecting small amount of photon in the eyeball even in dim-light conditions. Distribution depth for the eight species inferred from these results are as shown in Fig 4. In the future, determination of the maximum absorbance spectrum of the visual pigment using rhodopsin gene isolated the retina would be useful to evaluate their wavelength sensitivity.



▲ Fig 1. Measurements of eye morphology. Black part, lens; yellow part, gap between the lens and the pupil margin; gray part, iris.

► Fig 2. Comparison of ratio of aperture diameter of eye /head length (%) among 8 fish species. Values are the average, vertical error var indicates standard deviation. \* indicates significant difference ( $p < 0.05$ , U-test).



	<i>B. sp.</i>	<i>N. coatsi</i>	<i>C. wilsoni</i>	<i>P. antarcticum</i>	<i>E. antarctica</i>	<i>G. braueri</i>	<i>K. anderssoni</i>	<i>P. bolini</i>
AD/HL (%)	27.2	10.3	8.5	25.9	37.1	29.7	32.4	37.0
tapetum								
retina							no data	
	photopic vision				scotopic vision			

Fig 3. Eye and retinal morphologies. AD/HL, average of ratios (%). Tapetum, column for species having tapetum lucidum is shown in yellow. Retina, cross section is shown for *C. wilsoni*, no data for *K. anderssoni*. Scale bar is 30  $\mu$ m. PE, pigment epithelium. RL, rod layer.

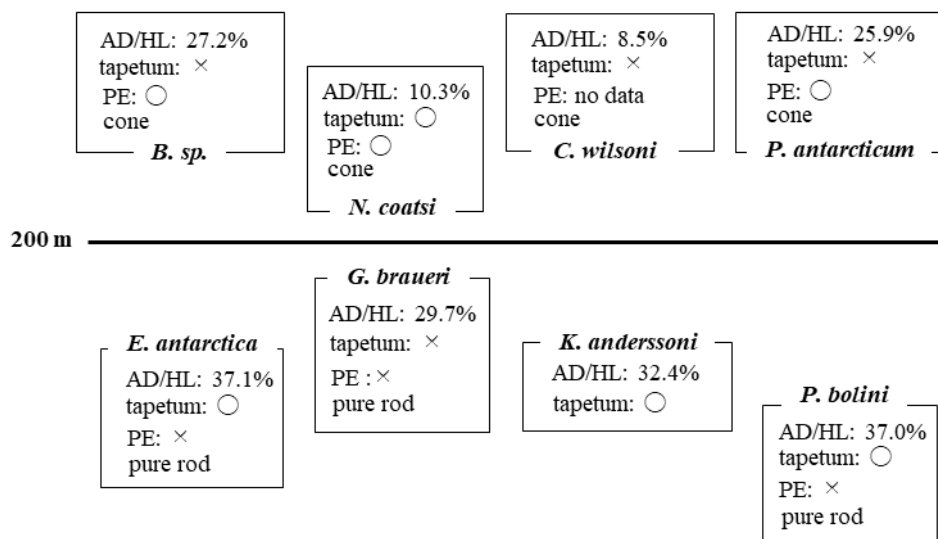


Fig 4. Ranking of water depth among eight species inferred from eye and retinal morphologies. The ranking was made with the boundary of 200 m, which is the definition of the deep sea.

## Distribution patterns of three common species of flying seabird off Vincennes Bay (East Antarctica)

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In the Southern Ocean, distribution patterns of seabirds in relation to oceanic front systems have been well studied. Frontal zones generally possess large biomasses; therefore, seabird distributions are likely to be influenced by the position of the front. This study aimed to clarify the influence of zooplankton abundance on seabird distribution of the short-tailed shearwater *Puffinus tenuirostris* (STSH), Antarctic petrel *Thalassoica antarctica* (ANPE) and Cape petrel *Daption capense* (CAPE) off Vincennes Bay in the Indian Ocean sector.

A seabird visual census, hydrography, and echosounder observations were conducted between 19-30 January, 2014. The visual census was conducted in a 100 m-bin off one side of the ship for 15 min every hour from sunrise to sunset. Seabird abundance (ind. / km) was calculated based on a total of eighty-nine census events. A scientific echosounder (frequency 70 kHz) was used to obtain the area's backscattering strength (SA) as an indicator of zooplankton abundance.

Based on water temperature and salinity profiles, the survey area was divided into three zones. STSH were mainly distributed in warm waters  $> 1.5^{\circ}\text{C}$  at the surface in the northern survey area as well as cold waters of  $< 0^{\circ}\text{C}$  at a depth of 200 m in the slope area. They were also found in intermediate waters between the cold and warm waters. CAPE were distributed in cold and intermediate waters. ANPE were observed abundantly in intermediate waters as well as cold waters. Higher SAs were observed in cold and intermediate waters. Correlations between seabird and SA (zooplankton abundance) were examined for each area of analysis (distance): 5 km, 10 km, 20 km, and 30 km. For STSH, no significant correlation was observed for any analysed area. Although no correlation was found between CAPE and SA over 5 km, a higher correlation was recorded over larger areas of analysis. For ANPE, correlations over 5 km and 10 km were higher than those over 20 and 30 km.

Previous studies have demonstrated that oceanic front systems or the geographical distributions of water masses influence sea bird distributions. However, in the present study, only hydrographical structure had a minor effect on seabird distribution. As well as food organism distribution, the timing of sea ice melting or distance to nesting sites are likely to be factors determining the distribution patterns of seabirds. Although CAPE and ANPE feed mainly on krill, the distributions of the two species were slightly different. Differences in the correlations of sea bird abundance to SA between the two species over various analysis scales imply differences in food preferences and feeding behaviour (flight distance). Among the three species, STSH were distributed most widely. This species feeds on lantern fish (myctophid) as well as on Antarctic krill. Antarctic krill are concentrated along the slope zone in our survey area, suggesting that bird populations in our study were feeding on different food items in cold or warm waters. For STSH, the lack of any significant correlation between seabird distribution and SA elucidates differences in the species' feeding habits compared to the above two species.

# Environmental DNA reveals spatial distributions and trophic linkages of an Antarctic marine community

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Monitoring of biological community structure and diversity is essential to understanding the impacts of global climate change on the Antarctic marine ecosystem. DNA metabarcoding based on environmental samples (environmental DNA (eDNA) metabarcoding) analysis allows simultaneous examination of organisms across multiple trophic levels from a single water sample. To elucidate the biodiversity and community structure of the marine ecosystem in the Southern Ocean, we conducted monitoring via multilocus amplicon sequencing for eDNA metabarcoding. We conducted eDNA sampling in January 2019 from 60°S to 65°S along 110°E off Wilkes Land, East Antarctica (Fig. 1A), using the training vessel Umitaka-maru of Tokyo University of Marine Science and Technology (Tokyo, Japan). Multi-marker analysis (nuclear 18S rRNA and mitochondrial cytochrome oxidase subunit 1, COI) of eDNA provided broad taxonomic coverage across the eukaryotic domain, across trophic levels from low (phytoplankton) to high (fish and squid). Mitochondria COI indicated that the major zooplankton species in the Southern Ocean and their distribution were consistent with previous findings, but more detailed spatial variations were obtained using eDNA. We identified four groups through cluster analysis of surface-layer samples and detected indicator species within each cluster group, representing samples from a given depth range (Fig. 1B). The indicator species in each cluster group included multiple trophic levels including autotrophic, mixotrophic and heterotrophic, demonstrating that community structure shifted vertically in relation to trophic linkages. Partial least squares analysis demonstrated that several indicator species (far left and right clades in Fig. 1C) in cluster groups were strongly correlated with environmental variables (Fig. 1C). Upper epipelagic communities (grey and blue) were associated with higher concentrations of nitrate and the large-sized fraction of chlorophyll-a (Chl a<sub>10</sub> μm). In contrast, lower epipelagic communities (yellow and orange) were associated with higher levels of phosphate and the small-sized fraction of chlorophyll-a (Chl a<sub>2</sub> μm and Chl a<sub>GF/F</sub>). Our results suggest that vertical changes in the epipelagic community are linked with environmental variables, which also influence trophic linkages in the Antarctic Ocean ecosystem. Our study demonstrates that eDNA analysis provides detailed and fine-scale information about complex biological interactions linked to environmental changes.

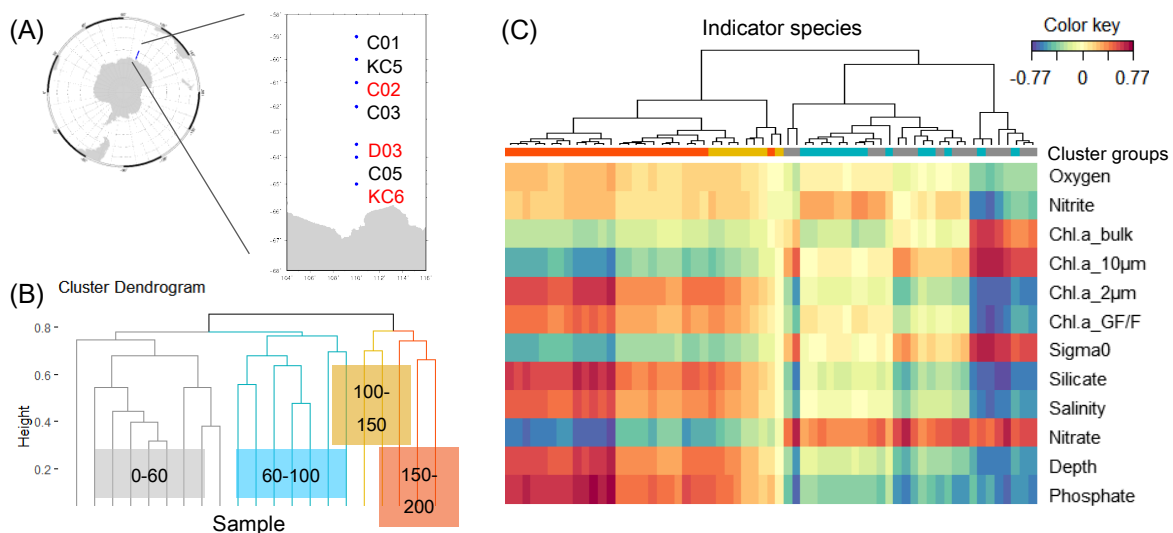


Figure 1. **A:** Map showing sampling stations off Vincennes Bay, East Antarctica. Samples used for the analyses presented in panels **B** and **C** were collected at stations C02, D03 and KC6 (red labels). **B:** Dendrogram based on clustering using the eDNA (normalised read abundance) index of each epipelagic sample (0–200 m) from stations C02, D03 and KC6. Colours indicate clustered groups. **C:** Heatmap showing the correlations of indicator species (columns), clustered based on epipelagic samples, with environmental variables (rows). Clustering of each indicator species is shown at the top of the heatmap using the same colours as panel **B**. The heatmap is coloured based on the value of the correlation coefficient.

## Evaluation of group behaviour of fish by 3-D positioning biotelemetry

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It has been a difficult task to continuously monitor group movement of animals living underwater over a long period of time in a natural environment. We aimed to investigate group behaviour based on continuous observation of fine-scale 3-D positions of individual fish within a confined space in wild using a biotelemetry method that we have developed. Field experiments were carried out in a square fish pen (12 m × 12 m × 6 m) off Kaminokae, Kochi, Japan in August 2020. A receiver (AQRM-2000; AquaSound Inc., Kobe, Japan) was deployed at each of four corners of the fish pen at a depth of 2 m. Eight oriental butterflyfish *Chaetodon auripes* (total length: 16.3 ± 0.4 cm) were captured near the fish pen using fish basket traps. They were attached with an ultrasonic transmitter having pressure sensor (AQPX-0625P; AquaSound Inc., Kobe, Japan) of which a transmitting interval was set to ~3 seconds. They were released into the fish pen and were monitored for 89 hours after release. All eight individuals survived during the experiment, however, unfortunately, two of eight transmitters did not emit a signal due to malfunction. 3-D positions of six fish were calculated by the hyperbolic positioning method. The calculated 3-D positions were formed into a time series of 3 second intervals by a linear interpolation, and then group behaviour was investigated based on those time series data. The fish swam along the net in the fish pen and formed a loose group with repeatedly dissipating and gathering. Sometimes, six individuals swam around the entire fish pen with forming a synchronized collective group. We evaluated those group movement by calculating nearest neighbour distance (NND) and separation swimming index (SSI). We showed that our biotelemetry method of fine-scale 3-D positioning can adequately describe group behaviour of fish in a confined space.



## **Population trends of Adélie penguins in Lützow-Holm Bay, East Antarctica**

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Adélie penguins are considered an excellent bio-indicator of Antarctic coastal marine ecosystems, as they consume a large amount of krill and fish, the key components to the ecosystems. Their breeding colonies have relatively easy access from nearby research stations in summer, which facilitates the monitoring of breeding performance and population changes. The number of breeding Adélie penguins has been monitored in Lützow-Holm Bay by the Japanese Antarctic Research Expedition (JARE) as early as 1961 (JARE 5). Annual population censuses have been conducted for 5-10 penguin colonies near Syowa station since 1982 (JARE 23). These population census data are among the most extended time series of any biological parameters in Antarctica and thus provide a valuable source of information to examine long-term changes in the Antarctic marine ecosystems. This presentation gives updates on the recent population trends of Adélie penguins in Lützow-Holm Bay. Overall, most penguin colonies increased over the past 40-60 years, with large fluctuations in the last two decades. The number of breeding pairs was highest in 2002-2004, gradually decreased until 2014-2016, and increased after 2016. Annual fast sea-ice cover in Lützow-Holm Bay also fluctuated, with extensive fast sea-ice remained throughout the summer for 2007-2012. I discuss how fluctuations in penguin breeding success, mediated through variability in fast sea-ice cover, would affect the population trends of Adélie penguins in Lützow-Holm Bay.

## **Protecting marine biodiversity beyond national jurisdiction: a penguins' perspective**

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The expansion of human activities in offshore maritime regions has outpaced the development of scientific knowledge and cooperative governance across these areas beyond national jurisdiction (ABNJ). In this context, current negotiations by the United Nations aim for an international legally-binding instrument to improve governance and sustainable use of biodiversity in ABNJ. Penguins are among the most threatened seabird groups today, notably at sea from fisheries and oiling. Here, we examine the available information on penguins' movements and evaluate their use of ABNJ across species and life-cycle stages. We highlight that in most of the 18 extant penguin species, the birds may undertake spectacular migrations, seasonally or throughout life-cycle stages. Long-range movements were reported in 16 species, with trans-jurisdictional distribution in 14 species, including 13 in ABNJ. Species richness in ABNJ varied extensively according to oceanic region, and also varied according to season. Transboundary movements, notably to ABNJ, are thus remarkably prevalent among penguin species. Consequently, provisions of the treaty under negotiation may crucially benefit penguin conservation, notably by creating a uniform legal framework to designate protected areas in ABNJ, and by promoting the precautionary approach to extraction activities. However, the seasonally-dynamic utilisation of ABNJ by penguins supports the pertinence of adding innovative protection tools, such as mobile Marine Protected Areas, to the negotiations for a more adequate protection of marine biodiversity. Underlining penguins' remarkable connectivity to the high seas can thus further strengthen this instrument aiming to protect a global ocean commons.

## Isolation and Identification of *Mortierella* spp. from a *Sanionia* Moss Colony in Ny-Ålesund, Spitsbergen Is., Norway

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The genus *Mortierella* is known as soil inhabiting saprobic fungi distribute worldwide (Wagner et al. 2013). Many species of the genus can produce polyunsaturated fatty acids focusing them on the potentially biotransforming organisms. Some of the species can be plant pathogens. *Mortierella* spp. have also been found in Arctic and Antarctic regions (Melo et al. 2014; Tsuji et al. 2016). However, species composition of this genus in the polar regions remains unclear. In the survey of Oomycete organs from a *Sanionia* moss colony in Ny-Ålesund, Spitsbergen Is., Norway in (Tojo et al. 2021), *Mortierella* spp. were also frequently isolated (12.5%, 79 of the isolates) from the moss. The isolates obtained were identified based on sequences of the rDNA-ITS region and cultural characteristics, and were identified into seven species including five new species candidates (Figure 1). The result suggested that *Mortierella* inhabiting *Sanionia* moss in Ny-Ålesund has a unique composition of species.

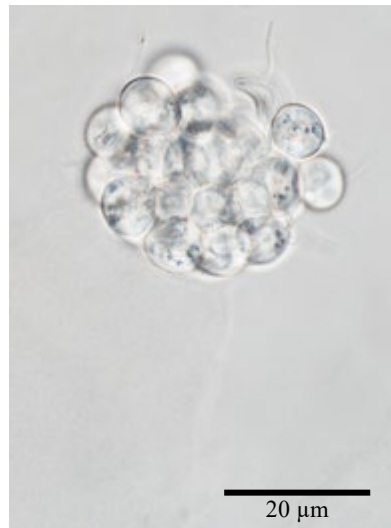


Figure 1. Part of sporangiophores with sporangia bearing sporangiospore of *Mortierella* sp. isolated from a *Sanionia* moss in Ny-Ålesund, Spitsbergen Is., Norway.

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## Factors affecting gestation periods in elasmobranch fishes

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The gestation periods of elasmobranchs vary among species from a few months to years, even possibly to >10 years. However, underlying factors remain unclear. Given that embryo growth rates affect gestation periods, body size and body temperature (i.e., major determinants of metabolic rates) are likely important factors explaining gestation periods. Although the effects of body temperature have been demonstrated for captive individuals, those of body size have never been examined. Whether metabolic rates of mothers or those of embryos affect gestation periods also remains unclear. Because metabolic scaling exponent ( $\beta$ ) is 0.8-0.9 in fishes and biological times generally scale with  $\text{mass}^{-1-\beta}$ , we hypothesized that elasmobranch gestation periods would scale with  $\text{mass}^{0.1-0.2}$ . We also hypothesized that regionally endothermic sharks (e.g., white sharks) with elevated metabolic rates would have shorter gestation periods than their ectothermic counterparts, assuming that the metabolic rates of mothers are an important factor. To test these predictions, we compiled data on gestation periods for 36 elasmobranch species, including four regionally endothermic sharks, from the literature. Phylogenetically-informed statistics showed that gestation periods scaled with  $\text{mass}^{0.1-0.2}$ . However, regional endothermy did not affect gestation periods. Our results suggest that metabolic rate affects elasmobranch gestation periods. Metabolic rates of embryos, which remain nearly motionless and are likely ectothermic in all species, may play more important role than those of mothers. Finally, to demonstrate the value of our approach, we estimated the gestation periods of two iconic species (frilled shark and Greenland shark), for which reproductive biology is poorly understood.

Table 1. Fitting of phylogenetically-informed multiple regression models

Inputted variable	Model	R <sup>2</sup>	AIC	ΔAIC
<b><i>M, m, E</i></b>	<b>1. <math>t \sim m</math></b>	<b>0.25</b>	<b>-17.3</b>	<b>0</b>
	2. $t \sim m + E$	0.26	-15.4	1.93

Models with  $\Delta\text{AIC} < 2$  were listed. The best models were represented in bold.  $t$ , gestation period (month);  $M$ , adult female mass (kg);  $m$ , birth mass (kg);  $E$ , whether the fish has regional endothermy. All continuous variables (i.e., gestation period, adult female mass, and birth mass) were log<sub>10</sub> transformed.

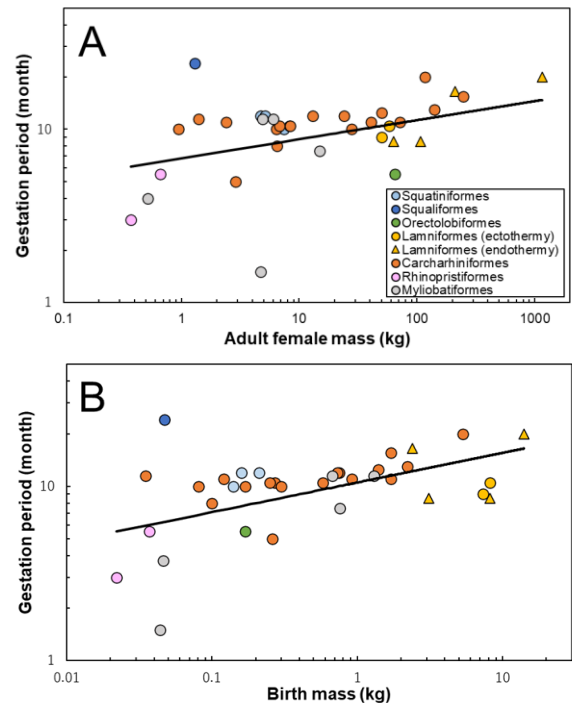


Figure 1. Gestation period  $t$  plotted against (A) adult female mass  $M$  and (B) birth mass  $m$ . Solid lines represent the PGLS regression lines [(A)  $t = 6.8 \times M^{0.11}$ ; (B)  $t = 10.5 \times m^{0.17}$ ]. Plots were colored by order to visualize phylogenetic relationships.

# Spatial-temporal changes in the macrozooplankton community in the eastern Indian sector of the Southern Ocean during austral summer; Comparison between 1996 and 2018/2019

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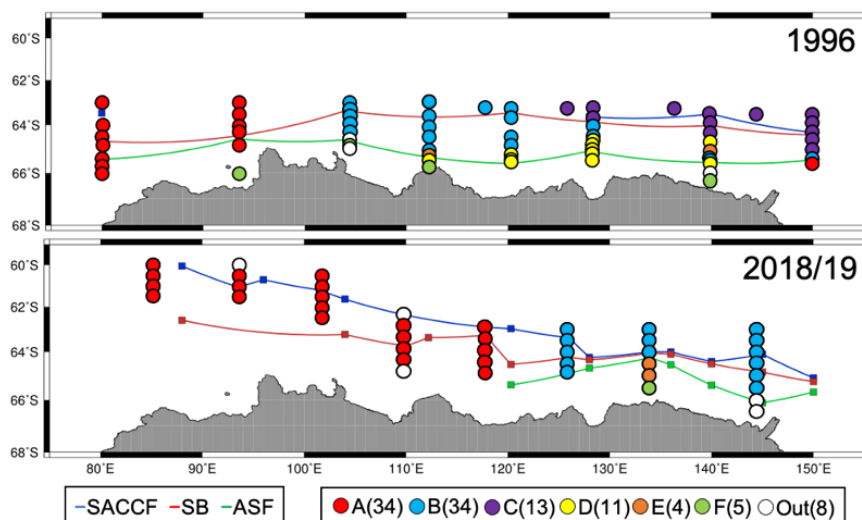
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The Indian sector of the Southern Ocean is featured by oceanic fronts developed by strong ocean currents such as Antarctic Circumpolar Current (ACC). As a result, the zooplankton community differs from north to south, with salps, small krills, amphipods, copepods and chaetognaths dominating in the north, while large krills and fish larvae dominating in the south. Although the importance of various zooplankton species has been suggested, most studies of macrozooplankton in the Southern Ocean have focused on *Euphausia superba*, with few reports of other members of the macrozooplankton community. Hosie (2000) reported the spatial-temporal variability of the macrozooplankton community in the eastern Indian sector of the Southern Ocean during the austral summer of 1996 under the Baseline Research on Oceanography, Krill and the Environment (BROKE) project, however, no large-scale surveys have been conducted in this area since then. In this study, we examined the horizontal distribution of the macrozooplankton community in the eastern Indian sector of the Southern Ocean during austral summer in 2018/19. In addition, we compared the macrozooplankton communities of 2018/19 and 1996 (Hosie 2000) to evaluate the relationship between the spatial-temporal variability of the macrozooplankton community and the environmental properties.

Surveys by the *R/V Aurora Australis* (Australian Antarctic Division) from the 30<sup>th</sup> of January to the 4<sup>th</sup> of April, 1996 and by the *R/V Kaiyo Maru* (Fisheries Agency of Japan) from the 15<sup>th</sup> of December, 2018 to the 23<sup>rd</sup> of February, 2019 were conducted in the range 80.00–150.00°E, 60.00–66.48°S. Zooplankton were collected by 0–200 m oblique tows with RMT8 (mouth area: 8 m<sup>2</sup>, mesh size: 4.5 mm) at 66 (in 1996) and 43 (in 2018/19) stations, respectively. Based on a cluster analysis, the macrozooplankton community was divided with six groups, and their spatial distribution were explicitly different between offshore and inshore regions (Figure 1). In the inshore region, *Euphausia crystallorophias* was abundant throughout the sampling period in both years. While in the offshore region, *Thysanoessa macrura*, *Clione limacina antarctica*, *Limacina helicina rangii*, and *Themisto gaudichaudii* were dominant in abundance. *Salpa thompsoni* was much more abundant in the area east of 120°E in 1996, but relatively low abundances were observed in 2018/19. The results of PERMANOVA analysis showed that the water masses commonly affected the zooplankton community classification in both years. On the other hand, the results of BIOENV analysis showed that the environmental factors affecting the abundance of macrozooplankton community differed between the two years, with salinity being the driver in 1996 and water temperature being the driver in 2018/19.



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Figure 1. Results of cluster analysis based on zooplankton abundance by Bray-Curtis similarity connected with UPGMA. SACCF: Southern Antarctic Circumpolar Current Front, SB: Southern Boundary of Antarctic Circumpolar Current, ASF: Antarctic Slope Front.

# The abundance of microscopic phototrophs and invertebrates in microbial mat of freshwater lakes and an ephemeral wetland of the Sôya Coast, East Antarctica

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The Microbial mats are known to harbour numerous microscopic phototrophs and invertebrates, which is comparable to that found in terrestrial mosses and lichens in Antarctica (Vincent & Quesada 2012; Verleyen et al. 2012). In recent decades, comprehensive diversity studies of microbial mats conducted using 16S rRNA and 18S rRNA gene analysis as well (Nakai et al. 2012; Hirose et al. 2020), however quantitative biological contribution of microorganisms in microbial mats remain unknown. In the present study, we surveyed the microscopic phototrophs and invertebrates from microbial mats of five lakes and an ephemeral wetland on the Sôya Coast, East Antarctica. We found seven taxonomic groups of phototrophs and three taxonomic groups of invertebrates in samples. Of the seven taxonomic groups, five group (Chlorococcales, Nostocales, Oscillatoriales, Chroococcales and Bacillariophyceae) and three invertebrates were found at all sites. Biovolume of invertebrates and phototrophs in the wetland was 7 to 30 times higher than in the five lakes. This suggests that the ephemeral wetland, which exist only for a very short period in Antarctica, are one of the most abundant habitats for microscopic organisms in the Antarctic terrestrial environment.

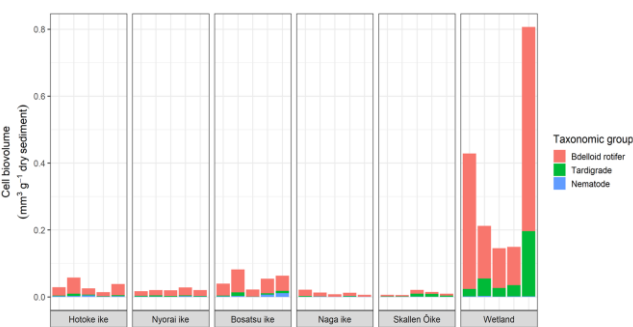
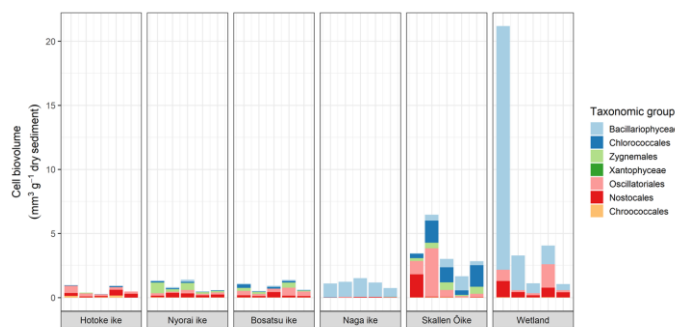


Figure 1. The abundance of seven taxonomic phototrophs at six sites.

Figure 2. The abundance of three invertebrates at six sites.

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## **Behavioural thermoregulation linked to foraging in blue sharks**

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Large pelagic fishes often dive and surface repeatedly as if they were airbreathers, raising a question about the functions of these movements. Some species (e.g., bigeye tuna, ocean sunfish) apparently alternate foraging in deep cold waters and rewarming in shallow warm waters. However, it is unclear how prevalent this pattern is among species. Blue sharks are the widest-ranging pelagic shark with expanded vertical niches, providing a model for studying foraging-thermoregulation associations. We used electronic tags, including video cameras, to record the diving behaviour, muscle temperature, and foraging events of two blue sharks. During repeated deep dives (max. 422 m), muscle temperature changed more slowly than ambient water temperature. Sharks shifted between descents and ascents before muscle temperature reached ambient temperature, leading to a narrower range (8 °C) of muscle temperature than ambient temperature (20 °C). 2.5-h video footage showed a shark catching a squid, during which a burst swimming event was recorded. Similar swimming events, detected from the entire tag data (20–22 h), occurred over a wide depth range (5–293 m). We conclude that, instead of alternating foraging and rewarming, blue sharks at our study site forage and thermoregulate continuously in the water column. Furthermore, our comparative analyses showed that the heat exchange rates of blue sharks during the warming and cooling process were not exceptional among fishes for their body size. Thus, behavioural thermoregulation linked to foraging, rather than enhanced abilities to control heat exchange rates, is likely key to the expanded thermal niches of this ectothermic species.

### 3-D dive paths of foraging Adélie penguins under Antarctic fast sea-ice

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Linking marine predator movements with prey distribution may contribute to our understanding of predator-prey interactions in 3-D marine environment. Recent developments in bio-logging have enabled us to simultaneously record 3-D dive paths and feeding activities of marine predators. In this study, we monitored the underwater 3-D diving behavior of Adélie penguins (*Pygoscelis adeliae*), to examine how they adjust their fine-scale foraging behaviors to prey distribution. Fieldwork was conducted at Hukuro Cove colony (69.21°S, 39.63°E) in Lützow-Holm Bay, East Antarctica, from late December 2018 to early January 2019. Fast sea-ice covered the sea around the breeding colony in our study area. We attached three types of data loggers to 13 penguins: 1) a GPS logger to examine movements on sea ice during foraging trips, 2) a multi-channel data logger to reconstruct their 3-D dive paths, and 3) a head-mounted accelerometer to examine their feeding events. During the foraging trips, penguins walked on fast sea-ice after leaving the breeding colony, and they conducted repeated dives (dive bouts) to feed on Antarctic krill (*Euphausia superba*) in small open waters along tidal cracks and leads. According to reconstructed 3-D dive paths, penguins swam horizontally for 60-100 m during a dive, fed on prey around the maximum horizontal distances they reached and mostly returned toward the starting point of dives after feeding. The horizontal travel distances in a dive tended to increase during a dive bout. This suggests that penguins gradually expanded the range of prey search during a dive bout in response to changes in the distribution (depletion or dispersion) of their prey (Antarctic krill). These results contribute to a better understanding of penguin-krill interactions under Antarctic fast sea-ice.



# CO<sub>2</sub> emission resulting from rain-on-snow events amounts to 10% of yearly soil CO<sub>2</sub> emission in Ny-Ålesund

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## Introduction

Global warming is the most critical issue for all the organisms in this earth including humankind. Polar soil is critically subject to global warming much more than soils at mid-latitudes or tropics and its transformation and degradation resulting from the global warming is one of critical issues for humankind. Permafrost soil stocks large amount of organic carbon and the loss rates of the organic carbon in permafrost soil resulting from the temperature increases are further important to estimate positive feedback of global warming (Biskaborn et al. 2019).

In Ny-Alesund, rain-on-snow events has been frequently observed because of recent increasing trends in winter temperature. However, the effect of rain-on-snow events on the degradation of organic carbon was not estimated so far. We aim at the estimation of contribution of CO<sub>2</sub> emission during winter seasons with/without rain-on-snow events to yearly soil CO<sub>2</sub> emission in Ny-Ålesund.

## Materials and Methods

From the previous results presented in the previous symposium, we made clear that the temperature dependence of negative temperature is much higher than that of negative temperature also for the soils at site 3 ecosystem monitoring site in Ny-Ålesund; temperature dependence among different places; the temperature dependence was smaller at shallower soils, because of supply of labile organic carbon from new plant litter. Furthermore, modeling the temperature dependence of soil CO<sub>2</sub> emission, connection of the emission across the positive and negative temperature is very important and we hypothesized that the temperature dependence expressed by the Arrhenius equation, we obtained the cross temperature. In connection with the purpose of our study, the soils at site3 are more following findings were revealed at the site3. Yoshitake et al. (2011) showed that microbial respiration is limited by carbon and nitrogen.

We used soil temperatures at site 3. Soil temperatures have been monitored hourly at 0cm, 2cm, 5cm, 10cm, 20cm, and 50cm by CR1000 datalogger (Campbell. Scientific, Utah, USA). Using the soil temperature data and the incubated soil respiration data, 0-50cm column of soil respiration was estimated during period from August 2016 to July 2017.

## Results and Discussion

The estimated yearly-integrated soil respiration using daily temperature data was the same within 0.5% of the estimated yearly integrated soil respiration using hourly data. So, the daily temperatures were used for the estimation of the integrated soil respiration (Figure 1).

The integrated soil respiration of 0-50cm of soil at Site3 from 2016 August to July 2017 amounted 88.6 molC m<sup>-2</sup> (1062 gC m<sup>-2</sup>). Soil

respiration of the 0-10 cm layer explained about 64.4% of the soil respiration integrated over 0-50 cm. Soil respiration rates from January to February during periods of two rain snow events were found to enhance 10% of the yearly-integrated soil respiration. Soil respiration

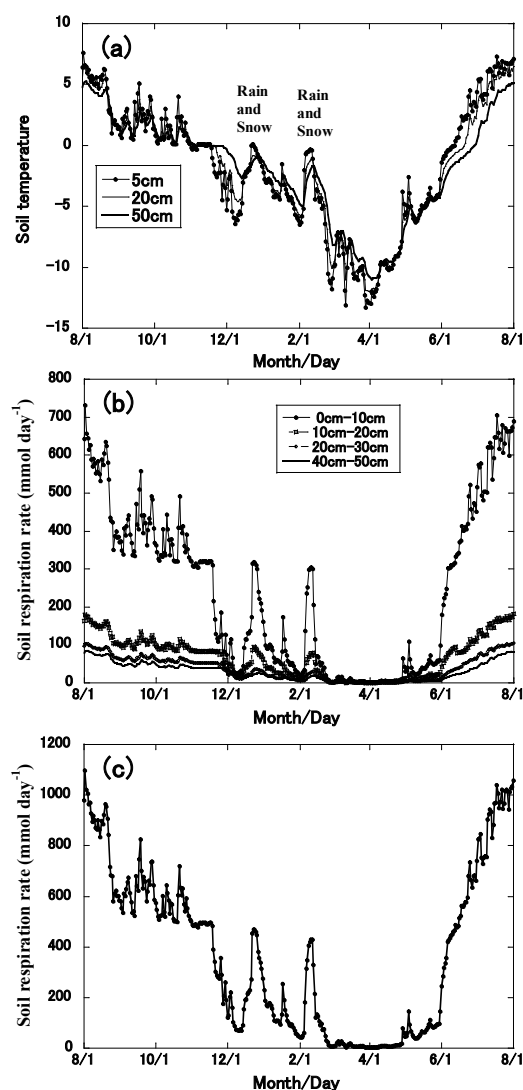


Figure 1. Soil temperatures (a), estimated soil respiration rates for 10cm-soil layers (b), and vertically-integrated soil respiration rates from soil surface (c) at Site3 from August 2006 to July 2007.

of 0-15 cm from February to March without rain on snow events explained only 0.5% of yearly soil respiration. The finding indicates the importance of the increased frequency of rain snow events which more happen with the progression of the global warming.

More frequency of rain on snow can drastically increase yearly-integrated soil respiration rates, because rain snow alters soil temperature from negative and positive, accelerating soil respiration rates. Then, the more frequent rain snows cause accelerated decomposition of soil organic matters. As pointed by Morgner et al. (2010) and Cooper (2014), warming during winter seasons are critically important in soil carbon dynamics. Monitoring of rain snow events and simultaneous studies of carbon cycles and soil dynamics in the polar ecosystems are important as the global warming is progressing. At the same time, it should be investigated whether the increased frequency of rain snow events can accelerate primary production by plants in polar ecosystems.

Furthermore, effects of rain-on-snow on yearly decomposition of soil carbon can be more if we do more accurate estimation of soil respiration using the relation with soil moisture. Soil respiration positively depend on soil moisture unless the soil moisture level is very high as to lead soil to anaerobic conditions. In summer at Ny-Ålesund, surface soil can be drier conditions when sunny days continue (data of soil moisture, now shown), leading to less contribution of soil respiration during summertime.

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## Genetic structures of genes involved in aromatic hydrocarbons metabolism from Antarctic soil bacterium strain BS19

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Oil pollution in soil and seawater bring devastating impact to the environment for extended periods of time. Increasing anthropogenic activities such as research programs and tourism add more demand for oil in the Antarctic region further increasing the likelihood for oil spillage accidents, while response to oil spills in Antarctica is hindered by several factors such as extreme weather and unreachable remote location. The retention of oil in the Antarctic environment is longer as the extreme low temperatures minimize effects from natural attenuation. Hydrocarbon-degrading bacteria especially *Pseudomonas*, *Parvibaculum* and *Rhodococcus* play important role in removing oil contaminants from the environment. Typically, attention is given to aliphatic hydrocarbons for monitoring the progress of any bioremediation effort. However, aromatic hydrocarbons are equally important as they could persist longer than their aliphatic counterparts.

The objective of this study is to identify genetic structures involved in aromatic hydrocarbons metabolism from an Antarctic soil bacterium *Sphingobium* sp. strain BS19. This strain was isolated as a heterocyclic aromatic hydrocarbon degrading bacterium from soil samples obtained at King George Island, Antarctica in 2019. The genome of this strain was sequenced using DNBSEQ system with an incomplete genome of 4.7 Mb sequenced, *de novo* assembled to 96 contigs. Using publicly available Rapid Annotation using Subsystem Technology (RAST) server (<https://rast.nmpdr.org/>), genes coding for terminal monooxygenase and dioxygenase possibly involved in aromatic hydrocarbon and heterocyclic compounds metabolisms were identified. Their genetic structures were also analyzed in comparison to homologous gene clusters reported in strains from warmer environments.