WP 39



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# Human footprint in Antarctica and the long-term conservation of terrestrial microbial habitats

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# Human footprint in Antarctica and the long-term conservation and study of terrestrial microbial habitats

### Working Paper submitted by Belgium, South Africa, United Kingdom and SCAR

#### Summary

Recent advances in molecular biology techniques have shown the presence of diverse microbial communities and the existence of species endemic to Antarctica. This means that the scientific value of Antarctic terrestrial microbial habitats can be compromised to a greater extent than previously realised by human visitation. The ever-increasing human footprint in Antarctica make consideration of this topic more pressing, as the number of locations known to be pristine habitats, where increasingly sophisticated cutting-edge research techniques may be used to their full potential, declines. The purpose of this paper is to highlight potential threats both to the conservation of terrestrial microbial ecosystems in Antarctica, and to future scientific research requiring study of these ecosystems.

#### Background

Ice-free ground in Antarctica covers only 44,000 km<sup>2</sup> or 0.34% of the continent's area, but hosts the bulk of terrestrial Antarctic life<sup>1</sup>. Microorganisms, including bacteria, algae, fungi, and viruses, generally comprise the majority of the biomass and biodiversity in Antarctic terrestrial and freshwater ecosystems, particularly under more climatically extreme habitats where higher organisms cannot survive<sup>2</sup>. Terrestrial sites may contain microbial communities that have persisted in Antarctica for at least several glacial cycles<sup>3</sup>, or species that have colonised more recently by natural processes such as wind currents or transport by migrating birds<sup>4</sup>.

In recent years, many important insights into Antarctic microbial ecology have been achieved using molecular biological techniques. Many new discoveries would likely not have been possible using traditional culture-dependant or microscopy techniques. Use of molecular techniques has shown that:

- 1. microbial biodiversity differs at sites across Antarctica<sup>5</sup>;
- 2. high levels of endemism exist in some microbial groups, despite the widely assumed ease of natural colonisation processes<sup>6</sup>;
- 3. microorganisms can exist in complex assemblages and at higher levels of diversity than estimated by earlier studies based upon microscopic and culture-dependent techniques<sup>7</sup>; and

<sup>&</sup>lt;sup>1</sup> Laws, R. M (Ed). (1984). Antarctic Ecology. Academic Press. London.

<sup>&</sup>lt;sup>2</sup> Friedmann, E. I. (Ed.). 1993. Antarctic microbiology. John Wiley and Sons, Inc., New York. and Cowan, D. A., Khan, N., Heath, C., and Mutondo, M. (2010). Microbiology of Antarctic terrestrial soils and rocks. In: Bej, A. K, Aislabie, J., and Atlas, R. M. (Eds.) Polar microbiology – the ecology, biodiversity and bioremediation potential of microorganisms in extremely cold environments. CRC Press, Boca Raton, pp 1-29.
<sup>3</sup> Convey, P., Gibson, J. A. E., Hillenbrand, C.-D., Hodgeson, D. A., Pugh, P. J. A., Smellie, J. L., and Stevens, M. I. (2008). Antarctic terrestrial life

<sup>-</sup> challenging the history of the frozen continent? Biological Reviews 82: 103-117 and Vyverman, W., Verleyen, E., Wilmotte, A., Hodgson, D. A., Willem, A., Peeters, K., Van de Vijver, B., De Wever, A., Leliaert, F., and Sabbe, K. (2010). Evidence for widespread endemism among Antarctic micro-organisms. Polar Science 4: 103–113.

<sup>&</sup>lt;sup>4</sup> Schlichting, J. R., Speziale, B. J., and Zink, R. M. (1978). Dispersal of algae and protozoa by Antarctic flying birds. Antarctic Journal of the United States 13: 147-149 and Pearce, D.A., Bridge, P.D., Hughes, K., Sattler, B., Psenner, R., and Russell, N. J. (2009). Microorganisms in the atmosphere over Antarctica. FEMS Microbiology Ecology 69: 143-157.

<sup>&</sup>lt;sup>5</sup> Cowan, D. A., Khan, N., Heath, C., and Mutondo, M. (2010). Microbiology of Antarctic terrestrial soils and rocks. In: Bej, A. K, Aislabie, J., and Atlas, R. M. (Eds.) Polar microbiology – the ecology, biodiversity and bioremediation potential of microorganisms in extremely cold environments. CRC Press, Boca Raton, pp 1-29 and Chong, C. W., Pearce, D. A., Convey, P., Yew, W. C., and Tan, I. K. P. (2012). Patterns in the distribution of soil bacterial 16S rRNA gene sequences from different regions of Antarctica. Geoderma 181: 45-55.

<sup>&</sup>lt;sup>6</sup> Vincent, W. F. (2000). Evolutionary origins of Antarctic microbiota: Invasion, selection, and endemism. Antarctic Science 12: 374–385; Lawley, B., Ripley, S., Bridge, P., and Convey, P. (2004). Molecular analysis of geographic patterns of eukaryotic diversity in Antarctic soils. Applied and Environmental Microbiology 70: 5963-5972; De Wever, A., Leliaert, F., Verleyen, E., Vanormelingen, P., Van der Gucht, K., Hodgson, D.A., Sabbe, K., and Vyverman, W. (2009). Hidden levels of phylodiversity in Antarctic green algae: further evidence for the existence of glacial refugia. Proceedings of the Royal Society of London Series B 276: 3591-3599; Vyverman, W., Verleyen, E., Wilmotte, A., Hodgson, D. A., Willem, A., Peeters, K., Van de Vijver, B., De Wever, A., Leliaert, F., and Sabbe, K. (2010). Evidence for widespread endemism among Antarctic microorganisms. Polar Science 4: 103–113 and Peeters, K., Hodgson, D. A., Convey, P., and Willems, A. (2011) Culturable diversity of heterotrophic bacteria in Forlidas Pond (Pensacola Mountains) and Lundstrom Lake (Shackleton Range), Antarctica. Microbial Ecology 62: 399-413.

4. Antarctic microorganisms have specialized genes that enable them to survive and function in polar habitats<sup>8</sup>.

Molecular biological techniques have seen dramatic increases in sophistication and reductions in cost in recent years. As these trends continue, understanding of scientific issues of local and global relevance, such as the effects of climate change on microbial systems, are likely to advance rapidly<sup>9</sup>.

In recognition of the need to understand comprehensively the biodiversity of the Antarctic continent and its surrounding oceans, SCAR has recently approved two new Scientific Research Programmes: State of the Antarctic Ecosystem (AntEco) and Antarctic Thresholds – Ecosystem Resilience and Adaptation (AnT-ERA). These new research programmes will deepen current understanding of the biodiversity of Antarctic ecosystems, and its response to current and future impacts, including the effects of human activities.

### Anthropogenic contamination of microbial terrestrial habitats

In Antarctic terrestrial sites, impacts such as chemical contamination or the introduction of a non-native plant or animal can be highly visible with a clear impact. In contrast, microbial or genetic contamination is not visually obvious and may not be considered routinely by non-microbiologists, or by those undertaking Environmental Impact Assessments. However, long-term impacts can result from:

- 1. Non-native microorganisms. Non-native microorganisms introduced from areas outside Antarctica, or from other biologically-distinct areas of Antarctica, may possess ecological, biochemical or physiological capabilities that alter the structure and function of the indigenous community.
- 2. *Transfer of novel genetic material.* Non-native species may contain genes that encode properties not found normally in pristine Antarctic microbial communities (e.g. resistance to antimicrobial agents or other physiological or biochemical benefits). These genes may be transferred from the non-native species to indigenous microorganisms by lateral gene transfer. If the genes are expressed in indigenous microorganisms, they may alter the functional properties of the individual species and the community as a whole<sup>10</sup>.

## Sources of microbial and genetic contamination

Introduced non-native microorganisms and microbial genetic contamination have been identified in association with:

1. dust, equipment and fibres from  $clothing^{11}$ ;

<sup>&</sup>lt;sup>7</sup> Sawstrom, C., Lisle, J., Anesio, A. M., Priscu, J. C., and Laybourn-Parry, J. (2008). Bacteriophage in inland polar waters. Extremophiles 12: 167-175; López-Bueno, A., Tamames, J., Velázquez, D., Moya, A., Quesada, A., and Alcamí, A. (2009). High diversity of the viral community from an Antarctic lake. Science 326: 858-61; Pointing, S. B., Chan, Y., Lacap, D. C., Lau, M. C. Y., Jurgens, J. A., and Farrell, R. L. (2009). Highly specialized microbial diversity in hyper-arid polar desert. Proceedings of the National Academy of Science USA 106: 19964-19969 and Cary, S. C., McDonald, I. R., Barrett, J. E., and Cowan, D. A. (2010). On the rocks: the microbiology of Antarctic Dry Valley soils. Nature Reviews Microbiology 8: 129-138.

<sup>&</sup>lt;sup>8</sup> D'Amico S, Collins T, Marx JC, Feller G, Gerday C. (2006). Psychrophilic microorganisms: challenges for life. EMBO Reports 7: 385-389 and Casanueva A, Tuffin M, Cary C, Cowan DA (2010) Molecular adaptations to psychrophily: the impact of 'omic' technologies. Trends in Microbiology

<sup>18:374-81</sup> 

<sup>&</sup>lt;sup>9</sup> Wall, D. H. (2007). Implications of change: above- and belowground biotic interactions in a low diversity ecosystem. Philosophical Transactions of the Royal Society London, Series B. 362: 2291–2306 and Loman, N. J., Constantinidou, C., Chan, J. Z., Halachev, M., Sergeant, M., Penn, C. W., Robinson, E. R., and Pallen, M. J. (2012). High-throughput bacterial genome sequencing: an embarrassment of choice, a world of opportunity. Nature Reviews Microbiology 10: 599-606.

<sup>&</sup>lt;sup>10</sup> Baker, G., Ah Tow, L., and Cowan, D. A. (2003). PCR-based detection of non-indigenous microorganisms in 'pristine' environments. J Microbiol Methods, 53: 157–164; Storrie-Lombardi, M. C., and Williamson, S. J. (2010). Possible role of bacteriophage-mediated horizontal gene transfer on microbial adaptation to environmental stressors in polar ecosystems. In: Bej, A. K, Aislabie, J., and Atlas, R. M. (Eds.) Polar microbiology – the ecology, biodiversity and bioremediation potential of microorganisms in extremely cold environments, p 179-199 and Cowan, D. A. Chown, S. L. Convey, P., Tuffin, M., Hughes, K. A., Pointing, S., Vincent, W. F. (2011). Non-indigenous microorganisms in the Antarctic - assessing the risks. Trends in Microbiology 19: 540-548.

<sup>&</sup>lt;sup>11</sup> Broady, P., and Smith, R. (1994). A preliminary investigation of the diversity, survivability and dispersal of algae introduced into Antarctica by human activity. Proc NIPR Symp Polar Biol 7: 185–197; Cowan, D. A., and Ah Tow, L. (2004) Endangered Antarctic Environments. Annual Reviews in Microbiology 58: 649-690; Takashima, M., Shirai, F., Sageshima, M., Ikeda, N., Okamoto, Y., and Dohi, Y. (2004). Distinctive bacteriabinding property of cloth materials. American Journal of Infection Control 32: 27-30; Shukla, S., Gupta, R., and Kashyap, A. (2006). Algal colonization of Schirmacher Oasis, Antarctica. Fifteenth Indian Expedition to Antarctica, Scientific Report, 1999. Department of Ocean

- 2. human waste<sup>12</sup>; and
- 3. shed human skin cells (it is estimated that a person may shed c. 300 million dead skin cells per day of which c. 10% may carry commensal microorganisms)<sup>13</sup>.

Therefore, visitation of Antarctic terrestrial sites by small numbers of people, or even individuals, will inevitably result in the release of some microbial contaminants. Culture-dependent and molecular studies have already shown that microbial contamination resulting from human activities can be detected in Antarctic soils long after the activities have ceased<sup>14</sup>.

#### Do we know which locations are free of potential microbial contamination?

The human footprint in Antarctica continues to expand, but our records and understanding of which areas have been visited is not yet comprehensive. Furthermore, there may be limited temporal and spatial precision in locating past scientific activities<sup>15</sup>.

Therefore two issues arise:

- 1. It is increasingly difficult to identify pristine areas that are known categorically to be free of earlier human activity, and that could represent the most appropriate locations for microbiological research using sophisticated technologies<sup>16</sup>.
- 2. Undertaking research at a site may compromise, to some degree, the value of that location for future molecular biological research. Given the technological advances in molecular biological methods over the past decade, we may reasonably expect that future scientists will have access to technologies with vastly greater scope and sensitivity than those we currently use. Therefore, the different scientific disciplines should consider choosing their research sites to minimize this risk, for example, rather than visit a pristine location use a previously impacted location, if an appropriate alternative exists<sup>17</sup>.

#### Gaps in our knowledge

To help inform our future works, we have identified some gaps in our knowledge.

1. Currently we understand little about the scale and kinetics of transport processes for non-native microorganisms after the initial introduction event, i.e. the extent to which non-native microorganisms from a heavily contaminated site may be transported by wind or other process to more pristine sites<sup>18</sup>.

Development, Technical Publication No. 13, pp 109-116 and Teufel, L., Pipal, A., Schuster, K.C., Staudinger, T., Redl, B. (2009). Material dependent growth of human skin bacteria on textiles investigated using challenge tests and DNA genotyping. Journal of Applied Microbiology 108: 450-461.

Sjoling, S., and Cowan, D. A. (2000). Detecting human bacterial contamination in Antarctic soils. Polar Biol 23: 644-650

<sup>&</sup>lt;sup>13</sup> Wilson, J. (2001). Infection control in clinical practice. 2nd Edition. Balliere Tindall, Edinburgh and Cowan, D. A. Chown, S. L. Convey, P., Tuffin, M., Hughes, K. A., Pointing, S., Vincent, W. F. (2011). Non-indigenous microorganisms in the Antarctic - assessing the risks. Trends in Microbiology 19: 540-548.

<sup>&</sup>lt;sup>14</sup> Hirsch, P., Gallikowski, C., and Friedmann, E. (1985). Microorganisms in soil samples from Linnaeus Terrace, southern Victoria Land: Preliminary Observations. Antarctic Journal of the United States 20: 183-186; Nedwell, D., Russell, N., Cresswell-Maynard, T. (1994). Longterm survival of microorganisms in frozen material from early Antarctic base camps at McMurdo Sound. Antarctic Science 6: 67-68; Sjoling, S., and Cowan, D. A. (2000). Detecting human bacterial contamination in Antarctic soils. Polar Biol 23: 644-650; Baker, G., Ah Tow, L., and Cowan, D. A. (2003). PCRbased detection of non-indigenous microorganisms in 'pristine' environments. J Microbiol Methods, 53, 157-164; Hughes, K. A. and Nobbs, S. (2004). Long-term survival of human faecal microorganisms on the Antarctic Peninsula. Antarctic Science 16: 293-297 and Cowan, D. A. Chown, S. L. Convey, P., Tuffin, M., Hughes, K. A., Pointing, S., Vincent, W. F. (2011). Non-indigenous microorganisms in the Antarctic - assessing the risks. Trends in Microbiology 19: 540-548.

<sup>&</sup>lt;sup>5</sup> Germany. (2000). Inventory of location of past scientific activities of Germany in Antarctica - ongoing studies. SATCM XII, IP 4.

<sup>&</sup>lt;sup>16</sup> United Kingdom. (2010). Assessing cumulative environmental impacts: identifying the distribution and concentration of national operator activities in Antarctica. ATCM XXXIII, WP 23 and Hughes, K. A., Fretwell, P., Rae, J. Holmes, K., and Fleming, A. (2011). Untouched Antarctica: mapping a finite and diminishing environmental resource. Antarctic Science 23: 537-548. <sup>17</sup> United Kingdom. (2010). Assessing cumulative environmental impacts: identifying the distribution and concentration of national operator activities

in Antarctica. ATCM XXXIII, WP 23 and Hughes, K. A., Fretwell, P., Rae, J. Holmes, K., and Fleming, A. (2011). Untouched Antarctica: mapping a finite and diminishing environmental resource. Antarctic Science 23: 537-548; Terauds ?; ASOC (2011). Evolution of footprint: spatial and temporal dimensions of human activities. ATCM XXXIV, IP 86; ASOC (2012). Data sources for mapping the human footprint in Antarctica. ATCM XXXV, IP 52.

<sup>&</sup>lt;sup>18</sup> Miwa, T. (1976). Anaerobic bacteria of Antarctica – isolation of Clostridia from the soil around Syowa Station. National Institute of Polar Research Memoirs, Series E 32: 56-63; Toyoda, S., Enokido, M., Matsumae, A., and Aiso, M. (1985). Microbiological investigations of the human pollution at

- 2. Few data exist describing how microbial diversity and function are altered following human visitation<sup>19</sup>.
- 3. Useful guidelines have been produced to reduce the risk of non-native species introductions and minimise environmental impacts at Antarctic terrestrial locations<sup>20</sup>, which are likely to be effective for larger organisms and their propagules. However, the effectiveness of these guidelines for reducing the risk of microbial contamination is not fully known.
- 4. Some Antarctic Specially Protected Areas (ASPAs) protect microbiological values and their Management Plans contain permit conditions to reduce microbial contamination (e.g. use of protective over-clothing) (e.g. ASPAs 126 and 130). However, the effectiveness of protective overclothing in preventing the release of non-native microorganisms within Antarctic protected areas has not itself been characterised<sup>21</sup>.
- 5. Much effort has been devoted to developing cutting-edge technologies and guidelines to facilitate access to sub-glacial lakes, while at the same time conserving and protecting these pristine environments<sup>22</sup>. However, clean technologies to allow sampling of pristine Antarctic terrestrial environments to an equivalent standard have not been developed. Further research into this area may help safeguard the pristine nature of some terrestrial microbial habitats.
- 6. A better understanding of which areas remain free of all past human activity may help inform conservation planning for microbiological habitats. Such locations may include inland sites situated far from research stations and logistics hubs.

### Recommendations

Belgium, South Africa, United Kingdom and SCAR recommend that the Committee:

- 1. consider microbial contamination of pristine sites during the Environmental Impact Assessments for activities in locations that are likely never to have been visited;
- 2. consider using the Antarctic Protected Area System more actively to protect microbial habitats for future science and for their own intrinsic value. A possible tool is the inclusion in the series of ASPAs of areas kept inviolate from human interference, as described in Annex V (Article 3); and
- 3. encourage Parties to undertake further research on the processes and impacts of microbial contamination in Antarctic environments, to inform future discussions within CEP.

A draft resolution is included in Attachment A for Parties' consideration.

Syowa Station in Antarctica. Journal of Antibacterial and Antifungal Agents 13: 541-546; Abyzov, S., Rusanov, V., and Smagin, V. (1986). Preliminary results of microbiological investigations at Mirny. Sovetskaia Antarkticheskaia Ekspeditsiia 78: 133-136; Ah Tow, L., Cowan, D. A. (2005). Dissemination and survival of nonindigenous bacterial genomes in pristine Antarctic environments. Extremophiles 9: 385–389; Russian Federation. (2006). Monitoring of pathogenic micro-biota in the Antarctic. ATCM XXIX, IP 72; Russian Federation. (2009). Microbiological monitoring of the expedition infrastructure facilities in the Antarctic. ATCM XXII, IP 46 and Pearce, D. A., Hughes, K. A., Lachlan-Cope, T., Harangozo, S. A., and Jones, A. E. (2010). Biodiversity of air-borne microorganisms at Halley station, Antarctica. Extremophiles 14: 145-159.

<sup>&</sup>lt;sup>19</sup> Kerry, E. (1990). Microorganisms colonising plants and soil subjected to different degrees of human activity, including petroleum contamination, in the Vestfold Hills and MacRobertson Land, Antarctica. Polar Biology 10: 423-430.

<sup>&</sup>lt;sup>20</sup> Committee for Environmental Protection (CEP). (2011). Non-native species manual – 1st Edition. Manual prepared by Intersessional Contact Group of the CEP and adopted by the Antarctic Treaty Consultative Meeting through Resolution 6 (2011). Buenos Aires, Secretariat of the Antarctic Treaty, Scientific Committee on Antarctic Research. (2009). SCAR's environmental code of conduct for terrestrial scientific field research in Antarctica. ATCM XXXII, IP 04
<sup>21</sup> Lankester, B. J. A., Bartlett, G. E., Garneti, N., Blom, A. W., Bowker, K. E., and Bannister, G. C. (2002). Direct measurement of bacterial

<sup>&</sup>lt;sup>21</sup> Lankester, B. J. A., Bartlett, G. E., Garneti, N., Blom, A. W., Bowker, K. E., and Bannister, G. C. (2002). Direct measurement of bacterial penetration through surgical gowns: a new method. Journal of Hospital Infection 50: 281-285; Owers, K. L., James, E., Bannister, G. C. (2004). Source of bacterial shedding in laminar flow theatres. Journal of Hospital Infection 58: 230-232; Eisen, D. B. (2011). Surgeon's garb and infection control: What's the evidence? Journal of the American Academy of Dermatology 64: 960-973; Hickman-Davis, J. M., Nicolaus, M. L., Petty, J. M., Harrison, D. M., and Bergdall, V. K. (2012). Effectiveness of shoe covers for bioexclusion within an animal facility. Journal of the American Society for Laboratory Animal Sciences 51: 181-188.

<sup>&</sup>lt;sup>22</sup> SCAR (Scientific Committee on Antarctic Research) (2011). SCAR's code of conduct for the exploration and research of subglacial aquatic environments. ATCM XXXIV IP33.

## Attachment A

# **Draft Resolution XX**

# On the long-term conservation of terrestrial microbial habitats

The Representatives,

*Recalling* Article 3.2 (a) of Annex V to the Protocol on Environmental Protection to the Antarctic Treaty which provides for the designation in the series of Antarctic Specially Protected Areas and the inclusion therein of areas kept inviolate from human interference so that future comparisons may be possible with localities that have been affected by human activities;

*Recalling* further that Article 3.2 of Annex V states that Parties shall seek to identify such areas within a systematic environmental-geographic framework;

*Recalling* also that Resolution 3 (2008) recommended that the 'Environmental Domains Analysis of the Antarctic Continent' annexed to that Resolution be used consistently and in conjunction with other tools agreed within the Antarctic Treaty system as a dynamic model for the identification of areas that could be designated as Antarctic Specially Protected Areas within the systematic environmental-geographical framework referred to in Article 3(2) of Annex V of the Protocol;

*Recalling* Resolution 6 (2012) that the Antarctic Conservation Biogeographic Regions be used in conjunction with the Environmental Domains Analysis and other tools agreed within the Antarctic Treaty system to support activities relevant to the interests of the Parties, including as a dynamic model for the identification of areas that could be designated as Antarctic Specially Protected Areas within the systematic environmental-geographic framework referred to in Article 3.2 of Annex V to the Environmental Protocol;

*Recalling* Article 4 of Annex II to the Protocol on Environmental Protection to the Antarctic Treaty which states that Each Party shall require that precautions are taken to prevent the accidental introduction of micro-organisms (e.g., viruses, bacteria, yeasts, fungi) not present naturally in the Antarctic Treaty area;

#### **Recommend that**:

- microbial contamination of pristine sites will be considered by Parties during the Environmental Impact Assessments for activities in locations that are likely never to have been visited; and
- the Antarctic Protected Area System be used more actively to protect microbial habitats for future science and for their own intrinsic value, including through the designation of Areas kept inviolate from human interference, as described in Annex V (Article 3).