



HEALTH SCIENCES

Fiocruz in Antarctica – health and environmental surveillance facing the challenges of the 21st century

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Abstract: FioAntar, FIOCRUZ's research project in Antarctica, is based on the *One Health* approach. FioAntar aims to generate relevant information that will help reduce the risk of future pandemics and improve the search for chemical compounds and new biological molecules. After four expeditions to Antarctica under the scope of PROANTAR, Fiocruz has identified Influenza H11N2 virus in environmental fecal samples, as well as *Histoplasma capsulatum* and *Bacillus cereus* in soil samples. In addition, in a prospective virome analysis from different lakes in the South Shetland Islands, six viral orders were described, supporting future research related to the biodiversity and viral ecology in this extreme ecosystem. Our findings of environmental pathogens of public health importance are a warning about the urgency of establishing a surveillance agenda on zoonoses in Antarctica due to the imminent risks that ongoing environmental and climate changes impose on human health across the planet. FioAntar strives to establish a comprehensive surveillance program across Antarctica, monitoring circulation of pathogens with the potential to transcend continent boundaries, thereby mitigating potential spread. For Fiocruz, Antarctica signifies a new frontier, teeming with opportunities to explore novel techniques, refine established methodologies, and cultivate invaluable knowledge.

Key words: Microorganisms, One health, Pathogens, Risk analysis, Surveillance

INTRODUCTION

In recent years, epidemics such as Ebola, Zika, Chikungunya, and most recently Covid-19, have drawn attention to the risks of human intervention in the environment and the consequent loss of biodiversity that affects various biomes of the planet. Several studies indicate the relationship between climate and environmental changes and the emergence of new pathogens and infectious diseases (El-Sayed & Kamel 2020, Heffernan 2018). The

Intergovernmental Panel on Climate Change report (IPCC 2021) asserts that human-induced climate change is causing dangerous and widespread disruptions in nature and affecting the lives of billions of people worldwide, and these disruptions are expected to intensify over increasingly shorter periods of time. This understanding has been brought by the World Health Organization (WHO) to the last two United Nations Climate Conferences - COP26 in Glasgow (WHO 2021) and COP27 in Egypt (WHO 2022).

Recently, it has been demonstrated in the Arctic, Himalayas and Antarctica, that both ice and permafrost, can harbor many microorganisms, known and unknown. These microorganisms can be harmful to human and animal health and this subject is of extreme concern, since ice in those regions is already melting (Lemieux et al. 2022, Alempic et al. 2023, Silva et al. 2022).

For a considerable time, the connection between animals, humans, and the environment appeared to have limited significance for public health. However, in the present era, the socio-economic model, food production methods, and rapid global transportation have unveiled compelling evidence and robust interconnections, emphasizing the profound linkages between global human health and animal health. Those evidence are supported in many case studies across the world (Carlson et al. 2018, Rabinowitz & Conti 2013, Blehert 2012). Climate change was also evidenced as an important issue that can promote environmental changes that affects biology and distribution of vectors and hosts (Abid & Abid 2023, Ellwanger et al. 2020, Fouque & Reeder 2019, Franklinos et al. 2019).

The Antarctic region, and its unique surrounding ocean, is one of the regions heavily affected by climate change, with consequences for human, animal, and planetary health. It is a region that has been little studied from a public health perspective, particularly regarding the risks and opportunities that microorganisms present in the region may pose to human health.

Wild birds are an example of reservoirs for a wide variety of infectious agents that threaten humans and animals, and are considered the natural origin of many viruses, especially Influenza A (Webster et al. 1992). Such reservoirs can contribute to the establishment of new endemic foci far from where an infection was acquired. The Antarctic region is home

of populations of seabirds that, during the summer, form colonies with thousands of individuals and migrate to distant places during the winter, reaching Brazil and further the northern hemisphere. At least 34 southern migrant seabird species from eight different families reach Brazil during migration (CEMAVE/ICMBio 2022).

The constant growth of tourism, along with other human activities in the Antarctic region, increases contact between humans and wildlife, increasing the risk for transmission of a pathogen to wildlife and vice versa. In the Antarctic environment, the close coexistence of different animal species and humans creates a favorable environment for the emergence of new viruses due to possible co-infections between viruses found in different species of birds and other animals (Smeele et al. 2018). In addition, the presence of migratory species that use highly anthropized sites in their migration routes could favor the spread of such viruses. Thus, the identification of pathogenic microorganisms and the monitoring of potential hosts of these pathogens have become important tools for surveillance and prevention of new diseases.

In addition to the need for surveillance and monitoring, there is also the importance and urgency of biomolecule research. The need to bioprospect previously unknown species for their potential for biosynthesis is highlighted by the growing demand for innovative products. Because next-generation sequencing technologies can give insight into an organism's entire biosynthetic landscape without requiring cultivation, the range of organisms that can be investigated for their biosynthetic content has highly increased. Extreme and little-known regions, such as Antarctica, present extreme survival circumstances that give rise to intricate ecosystems necessary for the life of microorganisms. Given this, it is predicted

that Antarctic microorganisms have distinct biosynthetic pathways and, consequently, new bioactive secondary metabolites that arise from their evolution and adaptation to constantly changing polyextreme environments.

FioAntar: the Fiocruz research project in Antarctica

The Fiocruz in Antarctica project (FioAntar) was conceived in 2018. It evolved with the approval of the research project submitted to the CNPq/MCTIC/CAPES/FNDCT Call No. 21/2018, within the scope of the Brazilian Antarctic Program (PROANTAR), having also been approved in an intramural grant program named Inova.

FioAntar is a multidisciplinary project with the objective of investigating the diversity and dispersal of potentially pathogenic microorganisms present in the Antarctic continent, applying this new knowledge to mitigate the risks of the emergence of new pathogens, antimicrobial resistance genes and to promote the bioprospection of molecules, genes or pathways that could benefit the Brazilian Unified Health System (SUS).

The identification of agents such as fungi, bacteria or viruses in soil, water, carcasses, and animal waste collected in Antarctica can reveal pathogenic organisms that represent risks to human health and to the health of Antarctic fauna. The possibility of finding different microorganisms in the same samples and places will allow studies of the relationships between them and their maintenance in the environment, helping to understand the local transmission processes that may favor dispersion to other species and territories. Lichens (symbiotic association between fungi and algae/cyanobacteria) are also targets of analysis, since they are notorious for the production of secondary metabolites with potential for the development of new drugs and biotechnological

products, such as antimicrobials, antitumor and photoprotector agents, amongst others. In this sense, FioAntar researchers have been collecting lichens for subsequent sequencing and analysis of their genomes, aiming to identify which organisms have the potential for the development of new technologies and products in health and industries, such as medicines and supplies.

FioLab: a piece of Fiocruz on the Antarctic continent

In January 2020, Fiocruz inaugurated the FioLab, a biosafety level 2 laboratory located at the Comandante Ferraz Antarctic Station (EACF) in Admiralty Bay, King George Island. This milestone was achieved through a Cooperation Agreement signed between the President of Fiocruz, Nísia Trindade, and Vice-Admiral Sergio Gago Guida, Secretary of the Secretariat of the Interministerial Commission for Marine Resources (SECIRM).

FioLab was established to effectively address the requirements of epidemiological and sanitary surveillance, while fostering integrated research that encompasses the realms of health and the environment in Antarctica. Adopting the One Health approach, the laboratory's activities are integrated with Fiocruz reference laboratories across Brazil. This collaborative integration enables comprehensive research to identify microorganisms present on the continent, revealing their biotechnological potential in various fields, including healthcare (novel drugs), environment (bioremediation), and industry (innovative enzymes). Furthermore, the FioLab plays a crucial role in the training and development of future specialists in health research pertaining to Antarctic studies.

The establishment of the FioLab represents Fiocruz's commitment to advancing scientific knowledge and contributing to the preservation

of Antarctica's unique ecosystem. As a signatory of the Antarctic Treaty, Brazil holds the responsibility to actively participate in decision-making processes concerning the continent's future. By spearheading the FioAntar project and integrating it into the PROANTAR under the coordination of the Interministerial Commission for Marine Resources (CIRM), Fiocruz demonstrates its dedication to strategically address present and future health emergencies through the prism of One Health, which recognizes the interconnectedness of human, animal, and environmental well-being.

MATERIALS AND METHODS

Five obligatory sampling points were established on different islands - King George, Nelson, Ardley, Penguin, and Deception - located in the South Shetlands. Afterwards, with a better understanding of the entire area, additional points were identified and were reached according to the operational feasibility in each expedition. The key criteria for selecting these points included the presence of diverse fauna, mammals and birds, the existence of tourism activities, areas with minimal human interference (ASPAS), and locations featuring lakes.

The initial FioAntar expeditions to Antarctica were focused on exploring the region, gaining insight into different areas, and identifying points of interest with significant findings. This initial exploration laid the groundwork for the establishment of a health surveillance plan encompassing areas affected and unaffected by human activity. The purpose of this plan was to monitor these points from a baseline perspective, enabling us to assess any changes over time.

To ensure a comprehensive understanding of the intricate relationship between health

and the environment, our surveillance program relies on a range of indicators. These indicators provide an integrated view, allowing us to grasp the multifaceted nature of this dynamic relationship (Traore et al. 2023, Leslie et al. 2007).

Drawing upon the INFORM Risk Index (2017), a risk management framework proposed by the European Commission, we are studying and structuring the most suitable indicators available. These indicators span three key dimensions: hazard and exposure, vulnerability, and mitigation capacity. The ongoing risk analysis is an integral component of FioAntar's mission, aiming to systematically organize and produce an annual reference index for health policies. This index will serve as a vital resource, offering valuable insights and guidance for future health-related decision-making.

At each designated sampling point, we meticulously collected a diverse range of samples to capture the ecological dynamics of the Antarctic environment. These samples included feces/excreta of various mammals and bird species, along with soil and lichen samples. Moreover, we collected water samples from six distinct lakes, namely the North and South lakes of Keller Peninsula at King George Island, Ardley North and South Lakes, Crater Lake at Penguin Island, and Kroner Lake at Deception Island. Regarding freshwater ecosystems, we intended to observe the potential microbial diversity present in these environments, with an initial focus on the virome. This comprehensive sampling strategy ensured that we obtained a broad range of data, allowing for a comprehensive understanding of the ecological dynamics in these areas.

Our team consists of specialized researchers in the fields of bacteria, fungi, helminths, viruses, genomics, bioinformatics, and experts interested in studying biomolecules present in lichens. This diverse expertise allows us to comprehensively

explore the microbial landscape of the region. To ensure an efficient sampling process, we have structured our approach based on the specific characteristics of each sample. This includes considering the pathogens of interest for each laboratory, tailoring the collection methods and appropriate packaging for each pathogen.

The first consideration for fecal samples is whether it comes from an individual or a colony. This differentiation is crucial, as it helps us target specific pathogens that may develop under the prevailing environmental conditions. Figure 1 provides an overview of the key characteristics of fecal samples, while Table I correlates these sample characteristics to the specific organisms being investigated. Additionally, the tables describe the preservation methods for the samples.

This careful organization ensures that our sample collection process is optimized for studying the pathogens of interest, facilitating a comprehensive approach on researching the diverse range of microorganisms present in Antarctica.

Given the multitude of samples to be collected and the unique challenges posed by fieldwork, we have implemented a systematic approach to ensure that no sample is overlooked. To achieve this, we have organized our field materials into kits, specifically tailored to each type of sample, as illustrated in Figure 2. Water samples were collected in 2L bottles and stored at 4°C.

In the laboratory on the ship, each kit is opened, and each tube or bag is labeled and packaged in the appropriate manner within the structure available on the Polar Ship Almirante Maximiano, which serves as the designated vessel for the FioAntar Project Staff.

Within the ship’s laboratory, our team ensures that every container is accurately labeled, clearly indicating its contents and any relevant information. Furthermore, we take great care to package the samples in a manner that ensures their integrity and maintains the appropriate conditions for transportation, storage, and analysis.

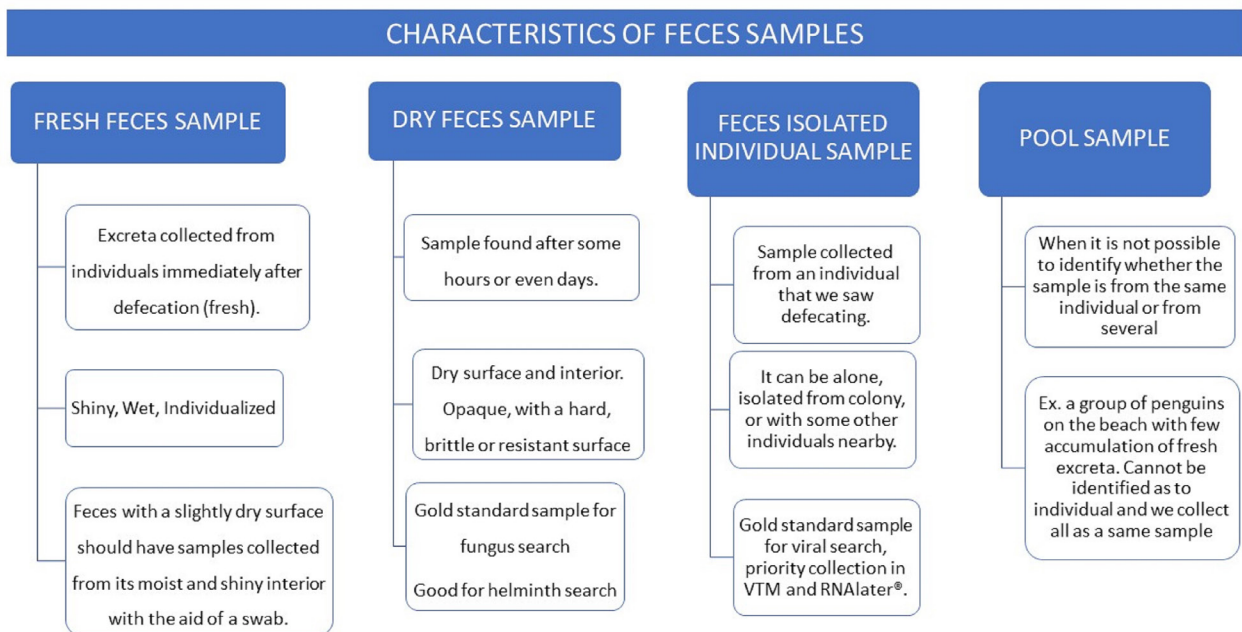


Figure 1. Key characteristics of fecal samples.

By following the established protocols onboard, we uphold the highest standards of quality control and meticulous data management. This systematic approach guarantees the traceability and reliability of our samples, from collection in the field to processing in the laboratory.

Upon arrival in Brazil, a fraction of each collected sample, in its respective conservation form, is allocated to the FioAntar laboratories within the institution. Flowcharts of the destiny of each sample collected are represented in Figure 3. A part of these samples will be preserved for future studies, when new questions and technologies might become available. This ensures that researchers from various interested groups have access to these samples, which were collected in a challenging and remote environment with extreme conditions.

The identified pathogens are deposited in 13 distinct collections (Figure 4), all housed at Fiocruz, providing a valuable resource for further research. Additionally, an aliquot of the natural sample is deposited in a *Fiocruz Biodiversity and Health Biobank (BBS-Fiocruz)*, enabling long-term storage and accessibility for future investigations.

Antarctic lichens were collected with a chisel and transported to Brazil, in dried and pressed form, or stored at -20°C . All samples were accompanied by detailed information about the collection site and the specimen collected. The dried lichen samples will be preserved in a herbarium, in the Rio de Janeiro Botanical Garden collection, facilitating their study and comparison over time. This ensures that as new questions and technologies arise, researchers can revisit these samples and obtain new answers, allowing comparisons between different time periods and environmental factors.

Lichen samples preserved at -20°C will be used in metagenome sequencing analyses. Briefly, they will have their DNA extracted with the DNeasy Plant Pro Kit (Qiagen, Hilden, Germany), according to the manufacturer's instructions for extracting plant DNA, and using the TissueLyser II equipment (Qiagen, Hilden, Germany), for mechanical lysis of cell. Subsequently, from the extracted DNA, the genomes will be sequenced on the MiSeq platform (Illumina) and the data obtained will be analyzed using bioinformatics tools to identify species and search for genes of biotechnological interest.

Each laboratory within the FioAntar project receives an aliquot of the collected samples and employs their respective protocols for pathogen analysis. This ensures that each laboratory can focus on the pathogens of interest and conduct their research in a specialized manner. Starting from Operantar XLI, metagenomes of collected samples will be sequenced in an attempt to better allocate each sample to one of FioAntar's specialized laboratories and for bioprospection.

While the comprehensive results of these laboratory analyses are extensive, we will provide an overview of the general findings. It is important to note that some of these results have already been published, while others are currently undergoing further studies or awaiting publication.

The published findings from our research efforts have contributed to the scientific community's understanding of the pathogens circulating in the Antarctic region. These publications have shed light on the diversity of microorganisms, including bacteria, fungi, and viruses that are present in this unique ecosystem. The data obtained through our studies have provided valuable insights into the potential risks and impacts of these pathogens on the local wildlife, as well as their implications for public health.

Table I. Sample characteristics to the specific organisms investigated and samples preservation methods description.

Sample characteristics	Conservation method	Stored Temperature	Pathogens seached
FECES			
Fresh or Dry Feces Sample	Railliet Henry Solution	ambient temperature	immature forms of helminths
Fresh or Dry Feces Sample	Natural, no conservation solution	4°C	Black fungus genera with pathogenic species - subcutaneous mycoses (cultivation); <i>Histoplasma capsulatum</i> ; <i>Paracoccidioides brasiliensis</i> ; <i>Coccidioides</i> spp.
Fresh Feces Sample	Viral Transport Medium (VTM) and RNA Latter	-80°C	Influenza A, Adenovirus and Avian Coronavirus
Fresh or Dry Feces Sample	Natural, no conservation solution	-80°C	Metagenomics analysis
SOIL			
We look for different kinds of soil. It must be collected soil samples close to the sea; on slopes; on glacier melt areas; permafrost; ornitogenic soil and under animal bed and carcasses	Railliet Henry Solution	ambient temperature	immature forms of helminths
	Natural, no conservation solution	4°C	Black fungus genera with pathogenic species - subcutaneous mycoses (cultivation); <i>Histoplasma capsulatum</i> ; <i>Paracoccidioides brasiliensis</i> ; <i>Coccidioides</i> spp.
	Natural, no conservation solution	-80°C	Metagenomics analysis
LIQUENS			
We look for liquens biodiversity	Natural, no conservation solution	ambient temperature	Deposit in the Rio de Janeiro Botanical Garden collection
	Etanol 70%	ambient temperature	Metagenomics analysis for molecules bioprospection
	Natural, no conservation solution	-80°C	
WATER			
Melt lake	natural water	4°C	Virome analysis
brackish water lake			

RESULTS

FioAntar participated, in a total, of three Antarctic operations: OPERANTAR 38 during 2019/ 2020 Antarctic summer, and Operantar 40 and 41. The expedition 39 was cancelled due to the Covid pandemic.

We have collected samples in 26 points around South Shetlands (Table II) of which 10 are ASPAs (Antarctic Specially Protected Area) and 14 are areas with some anthropization. The aim was to compare these two characteristics but due to the low number of positive results by point, this analysis will be performed in future.

We collected 122 water samples, 858 feces, 384 soil and 162 lichens (Figures 5 and

6). All FioAntar laboratories received samples (Figure 3) from the two first operations and characterization and distribution from the last operation (Op. XLI) are being prepared.

Following the specific protocols to study the virome from water, it was possible to observe that DNA viruses (99.4 %) prevailed over RNA viruses (0.6 %) in the lake samples. Six viral orders were identified in the metagenomic libraries: *Caudovirales* (dsDNA), which was prevalent in most lakes; *Picornavirales* (ssRNA+); *Sobelivirales* (ssRNA+); *Tolivirales* (ssRNA+); *Petitvirales* (ssDNA) and *Baphyvirales* (ssDNA), including eight viral families (*Herelleviridae*, *Siphoviridae*, *Myoviridae*, *Microviridae*,

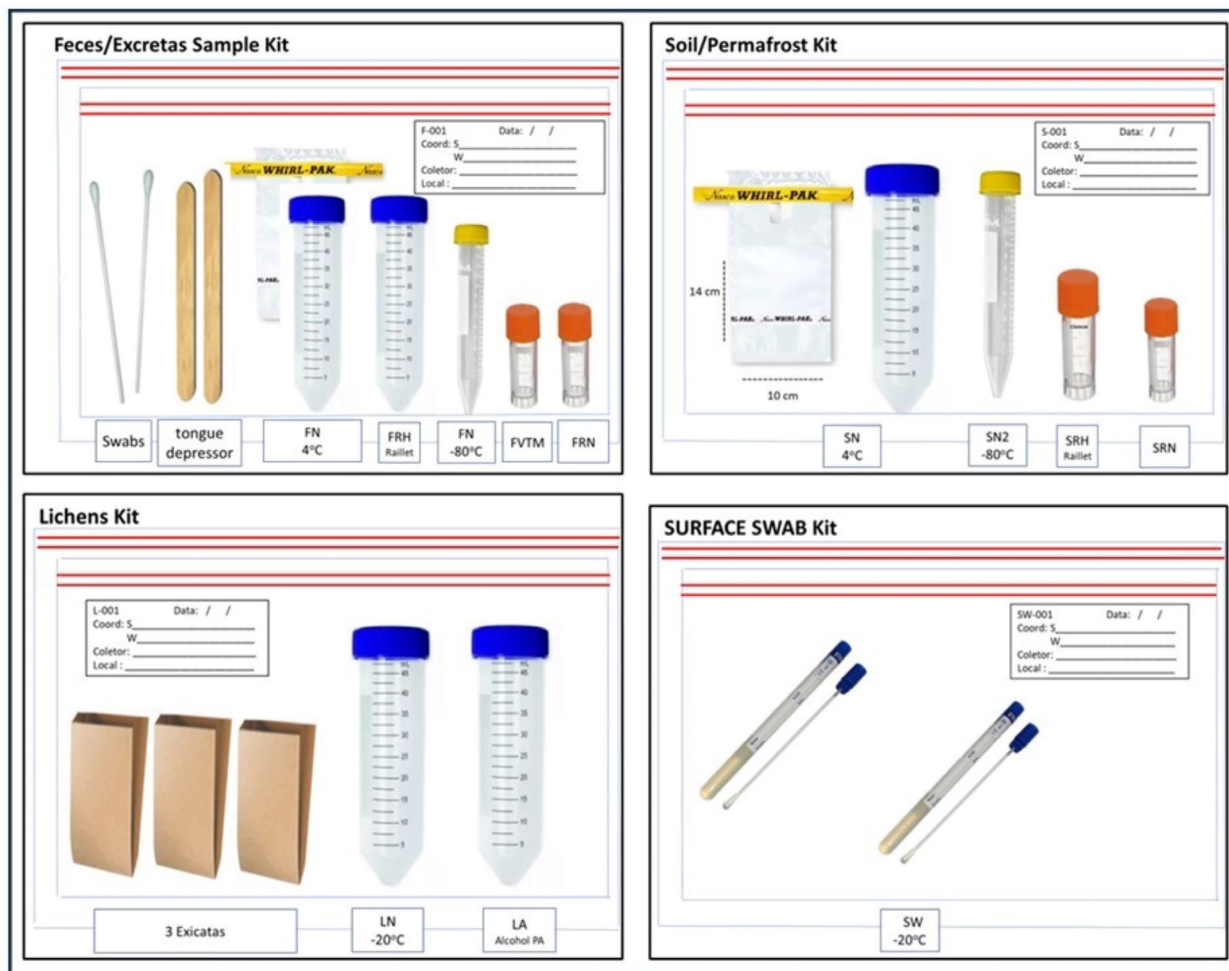


Figure 2. Sample Collection Kits, designed to organize samples in the field and ensure that no sample is overlooked.

Marnaviridae, *Bacilladnaviridae*, *Barnaviridae* and *Tombusviridae*) and several other, mainly non-classified ssRNA(+) viruses in the lakes of Ardley Island (Table III) (Prado et al. 2022).

Results of processed feces and soil samples are related to specific pathogen in Table IV. We point out that not all samples received from each laboratory are processed at the same time. Laboratories have their own interests and capabilities, and the results depend on multiple factors including qualified personnel, workforce and inputs.

We detected *Histoplasma capsulatum* (Moreira et al. 2022) in soil and penguin excreta in the Antarctic Peninsula by sequencing after

performing species-specific PCR, confirming previous observations that this pathogen occurs more broadly than suspected. Also, we detect influenza A (H11N2) virus in fecal samples from Adélie penguins (*Pygoscelis adeliae*) and from a colony of chinstrap penguins (*Pygoscelis antarcticus*) (Orgzewalska et al. 2022).

Bacillus cereus sensu stricto and related genera were found in feces and soil. *B. cereus* is a bacterium capable of causing various systemic and localized infections in immunocompromised and immunocompetent individuals. *B. cereus* related genera found (*Bacillus atrophaeus*, *Bacillus jeotgali*, *Bacillus licheniformis*, *Bacillus pumilus*, *Neobacillus ginsengisoli*,

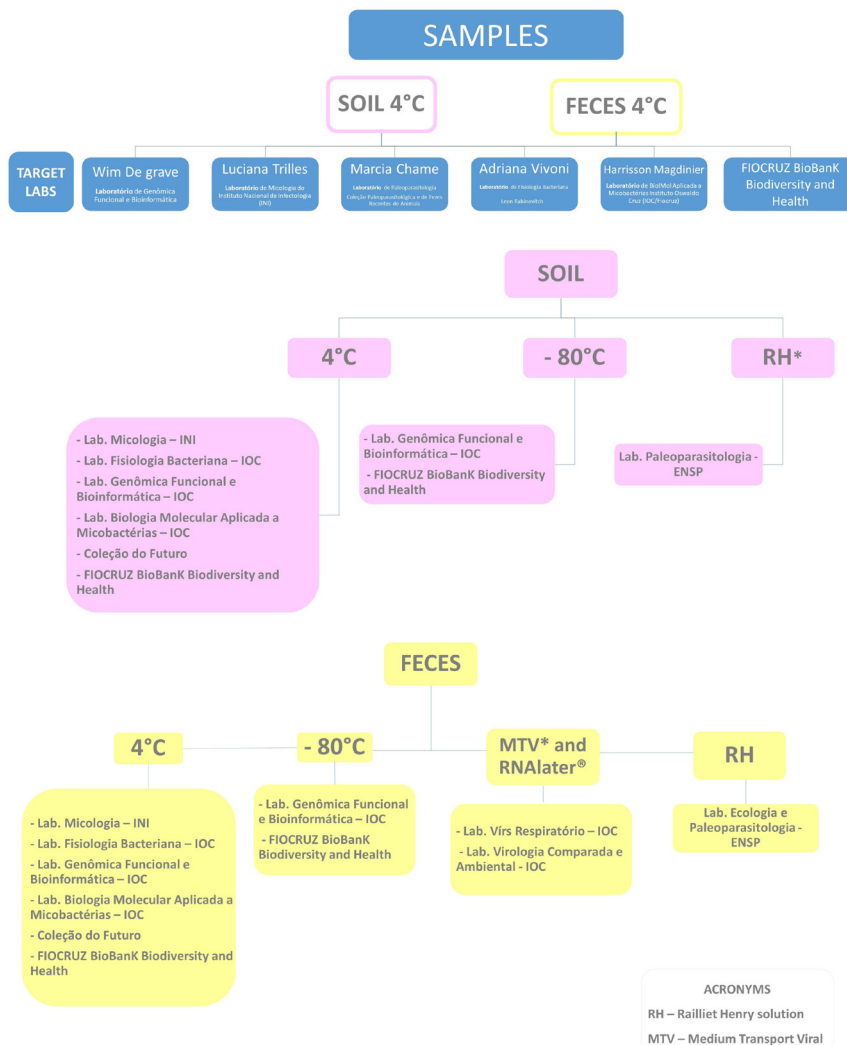


Figure 3. Flowchart of the destination of each subsample distributed among the laboratories that integrate FioAntar.

Paenibacillus dendritiformis, *Paenibacillus lautus*, *Paenibacillus macquariensis*, *Sporosarcina* sp.) are not pathogenic but are of interest in biotechnology.

Figure 7 presents the result of our first questions: “Are we carrying any pathogen to Antarctica during the expeditions? Are we bringing anything back”?

DISCUSSION

Beyond the findings already published, our ongoing efforts extend into comprehensive studies and analyses, delving even further into the intricacies of the collected samples. These investigations have a twofold purpose: firstly, to add insights into the identified pathogens, unraveling facets of their biology and other defining characteristics. Secondly, we aspire that our findings can guide us towards understanding the potential interactions that promote their circulation in the Antarctic environment.

The wealth of knowledge emerging from these endeavors is poised to deepen our

understanding of the dynamic relationship between pathogens and the Antarctic ecosystem. As we unlock more layers of information, we contribute to the broader scientific comprehension of these complex interactions, paving the way for informed insights and potential advancements in our approach to safeguarding this unique environment.

By sharing these findings and answers with transdisciplinary working groups, we aim to contribute to the broader scientific community and foster the importance of understanding the intricate relationships between pathogens, the environment, and public health in Antarctica.

In the present work, we report the isolation of the bacteria *Bacillus cereus* sensu stricto and related genera, in soil and feces samples in Antarctica. Brenner et al. (2013) described the isolation of *B. cereus* in Siberian permafrost soil samples 3 million years old, collected on Mammoth Mountain, Central Yakutia. This strain, which is estimated to be the same age as the soil from which it was isolated, shows great genetic similarity with modern strains of *B. cereus*,

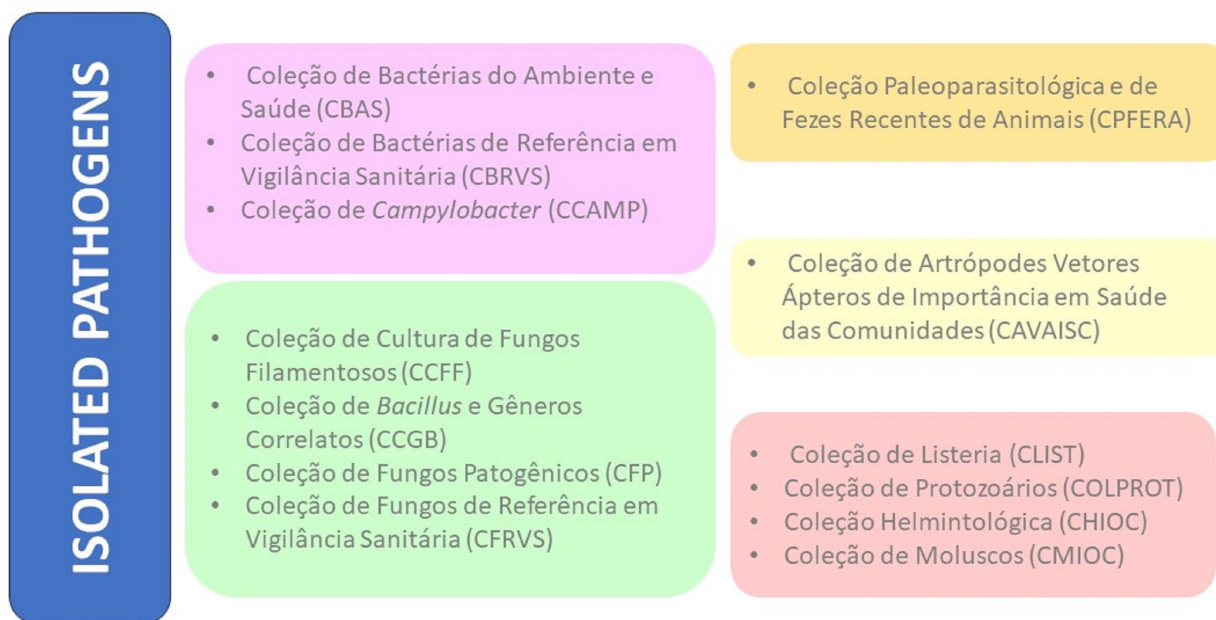


Figure 4. Fiocruz collections where the pathogens isolated by each group are being deposited.

and its spores were able to survive extreme conditions of temperature and deprivation of energy sources.

Bacteriophages (dsDNA) (*Herelleviridae* family) infecting the phylum *Firmicutes* and *Siphoviridae* were predominant in most lakes evaluated. Our working group has isolated bacteria of the soil samples around the lakes of King George Islands and preliminary results have demonstrated an abundance of *Bacillus*

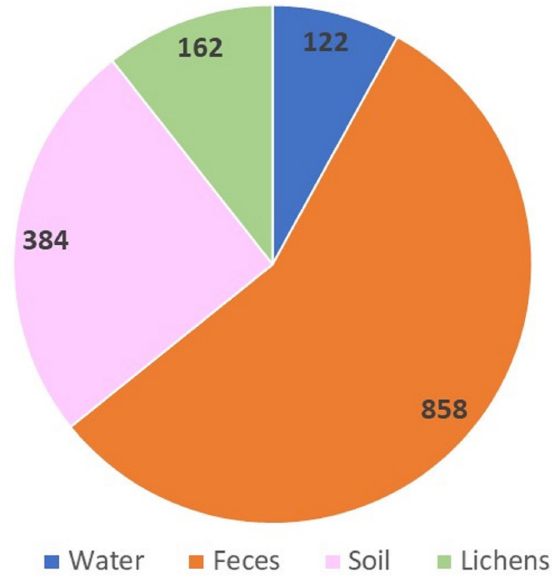
subtilis group bacteria, indicating that these microorganisms are part of the soil microbiome (Table III).

In the lakes of Ardley Island, a greater abundance of viruses with RNA genome was observed, most of them previously identified in Antarctic freshwater, and in the lake of Deception Island, double-strand DNA viruses that infect diatoms were detected. Functional analysis demonstrated that more than 60% of

Table II. Collection points executed by FioAntar from the NPo. Alm. Maximiano and EACF during three Operantar (XXXVIII, XL and XLI).

	Island	Collect Point	Operantar	Lat.	Long.	ASPA
1	Ardley	Ardley	XXXVIII; XL; XLI	-62.2162	-58.9230	Yes
2	Deception	Deception, Kroner Lake	XXXVIII; XL; XLI	-62.9806	-60.6551	Yes
3	Greenwich	Estação de Maldonado	XLI	-62.4482	-59.7434	No
4	King George	ASPA 128, Rakusa Point		-62.1615	-58.4661	Yes
5	King George	ASPA 128, Copacabana	XXXVIII; XL; XLI	-62.1735	-58.4681	Yes
6	King George	EACF – Refúgio 1	XXXVIII; XL; XLI	-62.0861	-58.3914	No
7	King George	Ipanema	XXXVIII; XL; XLI	-62.0847	-58.3923	No
8	King George	Lion's Rump	XXXVIII; XL; XLI	-62.1328	-58.1340	Yes
9	King George	Martins Head	XXXVIII; XL; XLI	-62.1625	-58.1749	No
10	King George	Morro da Cruz	XXXVIII; XL; XLI	-62.0815	-58.4040	No
11	King George	Península Keller, Pico Norte	XXXVIII; XL; XLI	-62.0661	-58.4038	No
12	King George	Península Keller, Refúgio 2	XXXVIII; XL; XLI	-62.0791	-58.4157	No
13	King George	Punta Hennequim	XXXVIII; XL; XLI	-62.1224	-58.3951	No
14	King George	Punta Ullmann	XXXVIII; XL; XLI	-62.0811	-58.3544	No
15	King George	Punta Plaza	XXXVIII; XL; XLI	-62.0523	-58.2400	No
16	King George	Potter Península	XXXVIII; XL; XLI	-62.2510	-58.9259	Yes
17	King George	Three Sister's	XL; XLI	-62.0775	-57.9138	No
18	King George	Turret Point	XL; XLI	-62.0843	-57.9492	Yes
19	King George	Wanda Glaciar	XXXVIII; XL; XLI	-62.1057	-58.3551	No
20	Livingston	Devil's Point	XL; XLI	-62.6662	-61.1478	Yes
21	Livingston	Hanna Point	XL; XLI	-62.6541	-60.6148	No
22	Nelson	Harmony Point	XL; XLI	-62.3035	-59.1929	Yes
23	Penguin	Penguim	XXXVIII; XL; XLI	-62.0964	-57.9255	No
24	Robert	Coppermine	XL; XLI	-62.3791	-59.7009	Yes

Total Samples Collected by FioAntar Operations XXXVIII, XL and XLI



Figures 5. Total Samples Collected by FioAntar on Operations XXXVIII, XL and XLI.

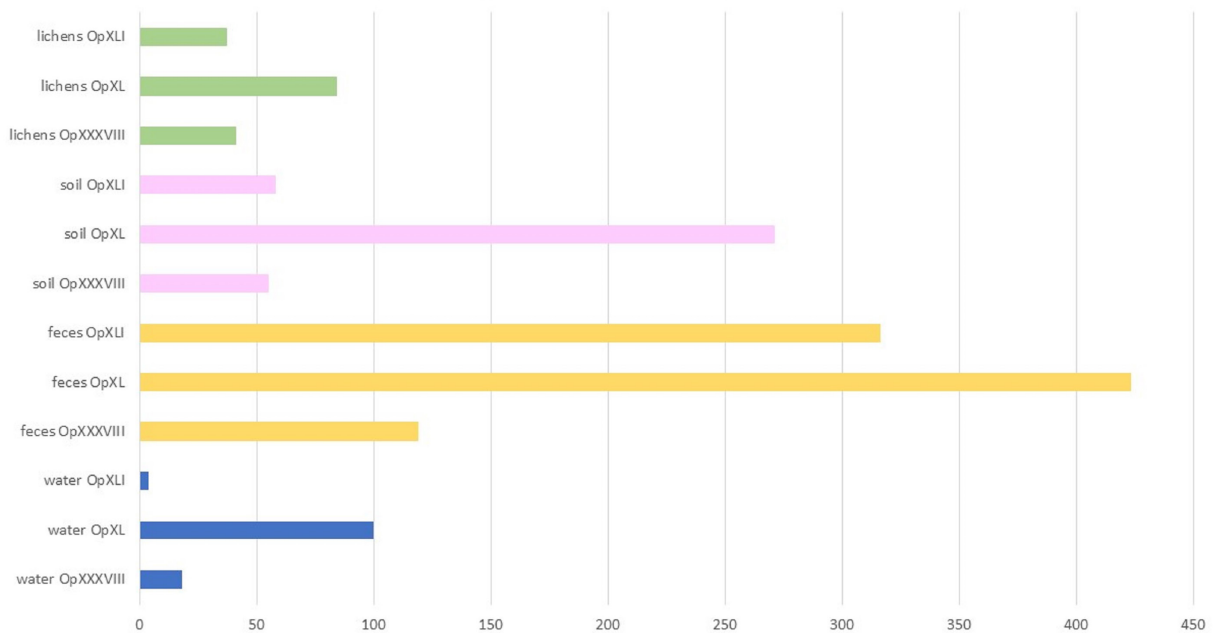
the genes have no predicted function, revealing the immense amount of information that still

needs to be elucidated in these polar freshwater ecosystems (Prado et al. 2022).

Although the role of wild birds in the ecology and transmission of avian influenza viruses is well established, there are still many doubts regarding the global distribution of these viruses, in relation to their prevalence, geographic distribution and/or importance in polar environments. In this study, Influenza A virus (H11N2) was found in penguin fecal samples and, recently, the literature describes the arrival of a highly lethal form of bird flu (H5N1) in Antarctica for the first time (British Antarctic Survey - BAS - communications, October 23, 2023). Therefore, it is critical to incorporate host and virus ecology into long-term surveillance studies to improve our understanding of the intricate relationship that avian influenza viruses have with their hosts in these environments.

The discoveries of *B. cereus*, *H. capsulatum*, and H11N2 underscore the imperative of sustained fauna monitoring to unravel the

Samples Type Collected by Operation



Figures 6. Total Samples by Type Collected on Operations XXXVIII, XL and XLI.

Table III. Viral genus and families identified in lakes from South Shetland Islands – Antarctica (2019/2020).

Lakes	Virus identified	Family	Predicted hosts
Crater Lake – Penguin Island	<i>Bacillus</i> phage SIOphi <i>CRESS</i> virus sp. <i>Picornavirales</i> sp. <i>Picornavirales</i> sp. <i>Jeonjuvirus</i>	<i>Herelleviridae</i> unclassified unclassified <i>Marnaviridae</i> <i>Herelleviridae</i>	<i>Bacillus subtilis</i> Invertebrate Invertebrate freshwater <i>Bacillus</i> sp.
King George Island – South Lake	<i>Bacillus</i> phage SIOphi (<i>Siophivirus</i>) <i>Bacillus</i> phage (<i>Pemunavirus</i>) <i>Bacillus</i> virus SPG24 (<i>Nitunavirus</i>) <i>Bacillus</i> phage phiNIT1 (<i>Nitunavirus</i>)	<i>Herelleviridae</i> <i>Myoviridae</i> <i>Herelleviridae</i> <i>Herelleviridae</i>	<i>Bacillus subtilis</i> <i>Bacillus</i> sp. <i>Bacillus</i> sp. <i>Bacillus</i> sp.
King George Island – North Lake	<i>Colwellia</i> virus 9A <i>Bacillus</i> phage SIOphi (<i>Siophivirus</i>) <i>Bacillus</i> phage (<i>Pemunavirus</i>)	<i>Siphoviridae</i> <i>Herelleviridae</i> <i>Myoviridae</i>	<i>Colwellia psychrerythaea</i> strain 34H <i>Bacillus subtilis</i> <i>Bacillus</i> sp.
Ardley Island - North	Shahe picorna-like virus 1 Antarctic picorna-like virus 3 Antarctic picorna-like virus 2 Tioga picorna-like virus 1 Changjiang tombus-like virus 8 <i>Bacillus</i> phage SIOphi (<i>Siophivirus</i>) <i>Marnaviridae</i> sp. <i>CRESS</i> virus sp. Beihai picorna-like virus 47 Beihai picorna-like virus 49 <i>Barnaviridae</i> sp. Sanxia picorna-like virus 5 Shahe picorna-like virus 2	unclassified unclassified unclassified unclassified unclassified <i>Herelleviridae</i> <i>Marnaviridae</i> unclassified unclassified <i>Marnaviridae</i> <i>Barnaviridae</i> <i>Marnaviridae</i> <i>Marnaviridae</i>	freshwater freshwater_ /Antarctica freshwater_ /Antarctica unknown Invertebrate <i>Bacillus subtilis</i> freshwater freshwater freshwater freshwater freshwater freshwater
Ardley Island - South	Antarctic picorna-like virus 3 Changjiang tombus-like virus 8 Wenzhou tombus-like virus 11 Gokushovirinae Fen7875_21 Antarctic picorna-like virus 2 Antarctic picorna-like virus 4	unclassified unclassified <i>Tombusviridae</i> <i>Microviridae</i> unclassified unclassified	Freshwater /Antarctica unknown environment environment freshwater /Antarctica freshwater /Antarctica
Kroner Lake – Deception Island	<i>Bacillus</i> phage (<i>Siophivirus</i>) <i>CRESS</i> virus sp. <i>Chaetoceros</i> protobacilladnavirus 1 <i>Bacillus</i> phage PM1 (<i>Pemunavirus</i>) <i>Microviridae</i> Fen7918_21 <i>Chaetoceros tenuissimus</i> DNA virus type-II <i>Chaetoceros tenuissimus</i> DNA virus type-II Wenzhou picorna-like virus 5 Beihai picorna-like virus 72 Marine RNA virus PAL_E4	<i>Herelleviridae</i> unclassified <i>Bacilladnaviridae</i> <i>Myoviridae</i> <i>Microviridae</i> unclassified unclassified <i>Marnaviridae</i> unclassified unclassified	<i>Bacillus subtilis</i> environment diatoms <i>Bacillus</i> sp. invertebrate (protochordate) <i>Chaetoceros tenuissimus</i> - diatom <i>Chaetoceros tenuissimus</i> strain - diatom protists / eucaryoto invertebrate environment

Data previously published in Prado et al. 2022.

Table IV. Results of processed feces and soil samples.

Searched Pathogens		N samples processed		Positive Results	
		feces	soil	feces	soil
Fungus	Histoplasma Capsulatum	12	9	3	2
Virus	PCR Influenza A	116		5	
	PCR Avian Coronavirus	116		0	
	PCR Avian Adenovirus	116		0	
Bacillus and related genera	Bacillus cereus	44	54	0	2

intricate pathways of these pathogens in the region. Questions loom: will they persist at their current locations, or will their prevalence surge? What nuances can we uncover about the pathogenicity and mutation tendencies of these species over time? These queries, among others, remain on our investigative horizon, marking the initial steps in a prolonged journey of research and knowledge. As we navigate this scientific terrain, our pursuit is anchored in uncovering answers that contribute to a deeper understanding of the dynamic interplay between these pathogens and the environment they inhabit.

CONCLUSIONS

The pandemic caused by SARS-CoV-2 has underscored the crucial importance of health knowledge, surveillance systems, and the integration of human, environmental, and animal health in effectively addressing present and future challenges. In this context, the endeavors undertaken by Fiocruz in Antarctica assume paramount significance. These initiatives embrace the concept of One Health, recognizing the interconnectedness of human and non-human health and the environment. "On a planet that is already experiencing the consequences of climate change, working to build knowledge and responses to face the health emergencies that are already knocking

at our door and affecting public health is our strategic duty on behalf of future generations" (Nísia Trindade Lima 2019).

Drawing upon a century-long legacy of research and scientific expertise, Fiocruz has embarked on pioneering studies in the Antarctic region. These studies assume strategic importance for Brazil, as signatory of the Antarctic Treaty, as they secure the nation's presence in the decision-making processes concerning the future of the continent. Through the FioAntar project, Fiocruz has contributed to the PROANTAR program, under the coordination of the CIRM and the Brazilian Ministry of Science, Technology and Innovation (MCTI).

By venturing into Antarctic studies, Fiocruz expands its scientific footprints and reinforces the commitment to global research collaborations. This milestone marks a significant step forward, allowing Fiocruz to actively contribute to the international scientific community's efforts in understanding and preserving the Antarctic ecosystem. With an extensive expertise and multidisciplinary approach, Fiocruz is poised to address critical health and environmental challenges in this pristine and fragile region.

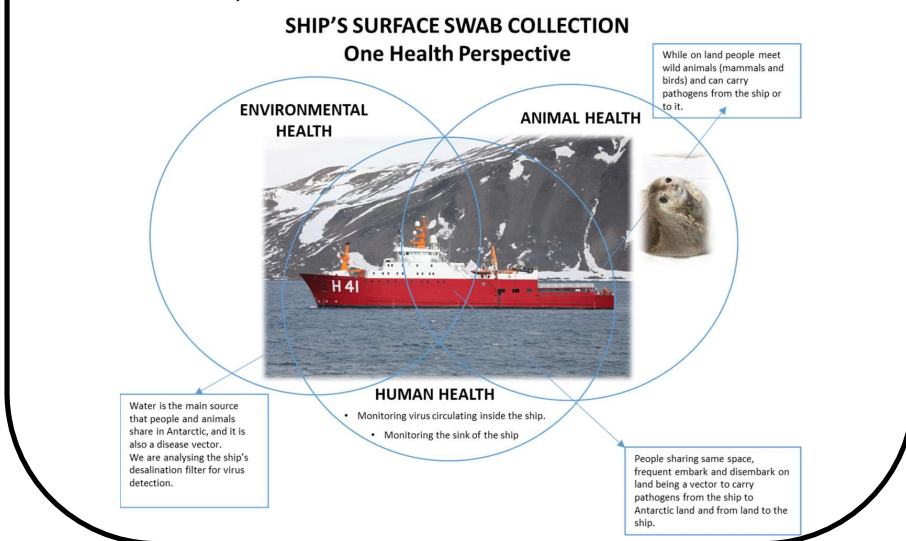
By aligning research initiatives with PROANTAR, Fiocruz demonstrates the dedication to promoting the sustainable development and responsible stewardship of Antarctica. By actively participating in the decision-making

Figure 7. Are we carrying any pathogen to Antarctica in the Navy? Are we bringing anything back?

In line with the One Health concept, which integrates various strategies to promote health, we conducted a study on the occurrence of human adenovirus (HAdV). HAdV serves as a crucial viral marker for tracing human fecal contamination within a ship's facilities. Our investigation aimed to evaluate environmental contamination arising from the movement of people during the different stages of an Antarctic expedition.

The objective of this study was to monitor human contamination by detecting the presence of these viral pathogens (HAdV) on surfaces and fomites aboard the polar vessel at various times, reflecting the high level of human activity over the course of the seven-month journey. We collected and analyzed a total of 128 environmental surface samples, all of which tested negative for HAdV. Routine cleaning and disinfection practices on the expedition were among the proactive measures implemented to prevent the spread of pathogens in enclosed spaces.

During polar expeditions, however, researchers also gather samples from soil, sea, lakes, ice, and animal feces. Through interaction with these elements, there is potential for microorganisms from the polar ecosystem to be transferred to the ship. A significant concern is the possible spread of certain microorganisms, especially pathogens, to other continents. As a result, we plan to employ additional molecular detection methods, such as metagenomics, to investigate the microbial diversity typically found on indoor surfaces of polar vessels."



processes, Fiocruz ensures that the perspectives and expertise of Brazil are well-represented, helping shape the future of the continent in a manner that respects its ecological significance and safeguards its immense scientific value.

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Conception of the study: MB, LM, TL, AV, MC, SS, HGM, MM, MO, MS, WD. Sample collection: LT, AV, HMG, TP, LM, MM, HM, RV, MO, MB. Methodology, data analysis and interpretation: WD, TP, MB, RV, LT, AV, MS, MM, MO, MS, FG, TP, MB. The initial draft of the manuscript was written by MB, LM, WD. Contributed with manuscript revision: RV, TP, TP MS, MO, GFD, WD. All authors read and approved the final manuscript.

