

# UNESCO environmental DNA (eDNA) Expeditions in marine World Heritage sites

## Biodiversity survey for French Austral Lands and Seas

Interim Results Based on 50% of Samples Analyzed  
Not for public distribution

UNESCO's eDNA Expeditions is a global, citizen science initiative that is conducted with the intention to measure marine biodiversity and predict the impact climate change on marine community composition across UNESCO World Heritage marine sites.

eDNA is a cost effective and minimally invasive method to measure biodiversity in any given area. Marine species continuously shed DNA into the water around them in the form of waste, mucus, or cells. By collecting this DNA from water samples, and subsequently amplifying and sequencing specific regions within these DNA strands, experts can detect a wide variety of species without removing any organisms from their environment. The amplified regions are selected depending on the scope of the study: while some markers are suitable to obtain a broad overview of biodiversity across many groups of species, other markers are particularly suited to get high resolution insight for a more specific taxonomic group. The UNESCO eDNA Expeditions initiative combines a number of markers tailored for marine vertebrates (fish, mammals, and turtles). A general marker was added to allow to detect also species from other groups such as invertebrates which can give a broader insight in the marine species diversity in a given area.

Between September 2022 and May 2023, UNESCO organized eDNA sampling campaigns in 21 marine World Heritage sites across 17 countries around the world. At every World Heritage site, about 20 samples were collected from different habitats. Over 250 young people participated in the local sampling expeditions. They were guided by local experts and park management staff, using protocols and eDNA sampling equipment provided by UNESCO.

Biodiversity inventories generated from the samples are being combined with existing species distribution data from public biodiversity databases such as the Ocean Biodiversity Information System (OBIS) in view of obtaining a comprehensive overview of marine life across marine World Heritage sites. Through the use of climate scenarios and species distribution models, an estimate will be made of the impact of climate change, in particular rising temperatures, on local biodiversity and its potential future distribution patterns.

More information about the initiative is available on the UNESCO website: <https://www.unesco.org/en/edna-expeditions>.

## Results

### Sampling and sample processing

eDNA sampling was conducted in French Austral Lands and Seas in December 2022. A total of 20 samples were collected at 3 locations in the park: Ile Haute - Kerguelen, Kerguelen - Armor, and Kerguelen - Suhm (Figure 1). Up to 1500 mL of seawater was filtered through filter cartridges with a 0.8  $\mu\text{m}$  pore size. After sampling, the filter cartridges were flushed with preservation liquid and shipped to UNESCO. From the 20 samples, 8 have now (December 2023) been processed. A second batch of samples will be processed in January 2024. DNA from these samples was extracted and amplified, and then sent to the sequencing facility at KU Leuven in Belgium, a specialized university eDNA lab contracted by UNESCO for this work. PCR amplification was done using five primer pairs selected to capture as much biodiversity as possible in the target groups of the study: fish, mammals, and turtles. The results in this preliminary report are based on a first batch of 8 samples that were analyzed covering all locations that were sampled in the World Heritage site.

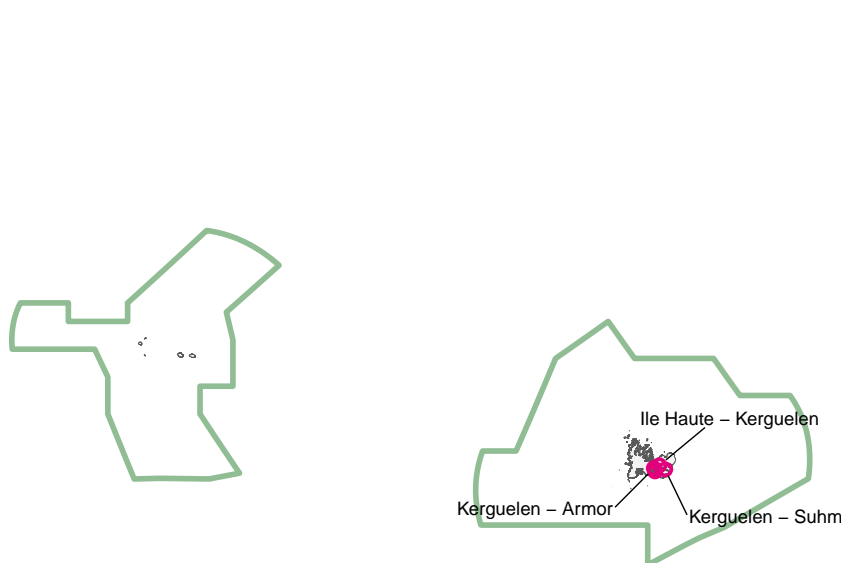


Figure 1: Map of the sampling locations.

### DNA sequencing

Sequencing of the DNA from 8 samples resulted in over 17 million sequence reads (Table 2), from which we collected 8178 unique sequences or ASVs. By matching those sequences with reference sequences in public databases, we were able to detect 112 species. Most of these species are vertebrates, but also species from other groups were detected, including algae, worms, starfish, jellyfish, and molluscs (Figure 2). Of the 112 species detected, 63 are not among the 3173 species previously reported from French Austral Lands and Seas to the UNESCO OBIS database.

reads	asvs
17199365	8178

Table 1: Reads, ASVs, and species across all samples.

locality	materialSampleID	sampleSize	reads	asvs	species
Ile Haute - Kerguelen	EE0013	1500	2309689	3848	49

Ile Haute - Kerguelen	EE0015	1500	1935299	3781	44
Ile Haute - Kerguelen	EE0018	1500	2460092	3926	61
Kerguelen - Armor	EE0007	1500	957024	3303	49
Kerguelen - Armor	EE0016	1500	1865522	3556	48
Kerguelen - Suhm	EE0008	1500	1128457	1446	13
Kerguelen - Suhm	EE0011	1500	1602548	3217	54
Kerguelen - Suhm	EE0020	1500	1283787	3477	49

Table 2: Reads, ASVs, and species by sample.

## Species identification

The marker sequences obtained from sequencing were matched with sequence reference databases built using public data available from the National Center for Biotechnology Information (NCBI). This resulted in the identification of 112 species, of which 14 fish species, 2 mammalian species, and 2 turtle species (Table 3). This represents about NA% of the species known from French Austral Lands and Seas in the OBIS database. 4 of the detected species are listed as threatened on the IUCN Red List (Table 4). A full list of species is added at the end of this report.

group	eDNA species	total species	fraction
fish	14	333	0.04
mammals	2	31	0.06
turtles		1	

Table 3: Number of species in the three target groups, from the OBIS database and from eDNA sampling.

category	eDNA species	total species	fraction
CR		1	
EN	2	13	0.15
VU	2	19	0.11

Table 4: Number of species by IUCN Red List category, from the OBIS database and from eDNA sampling.

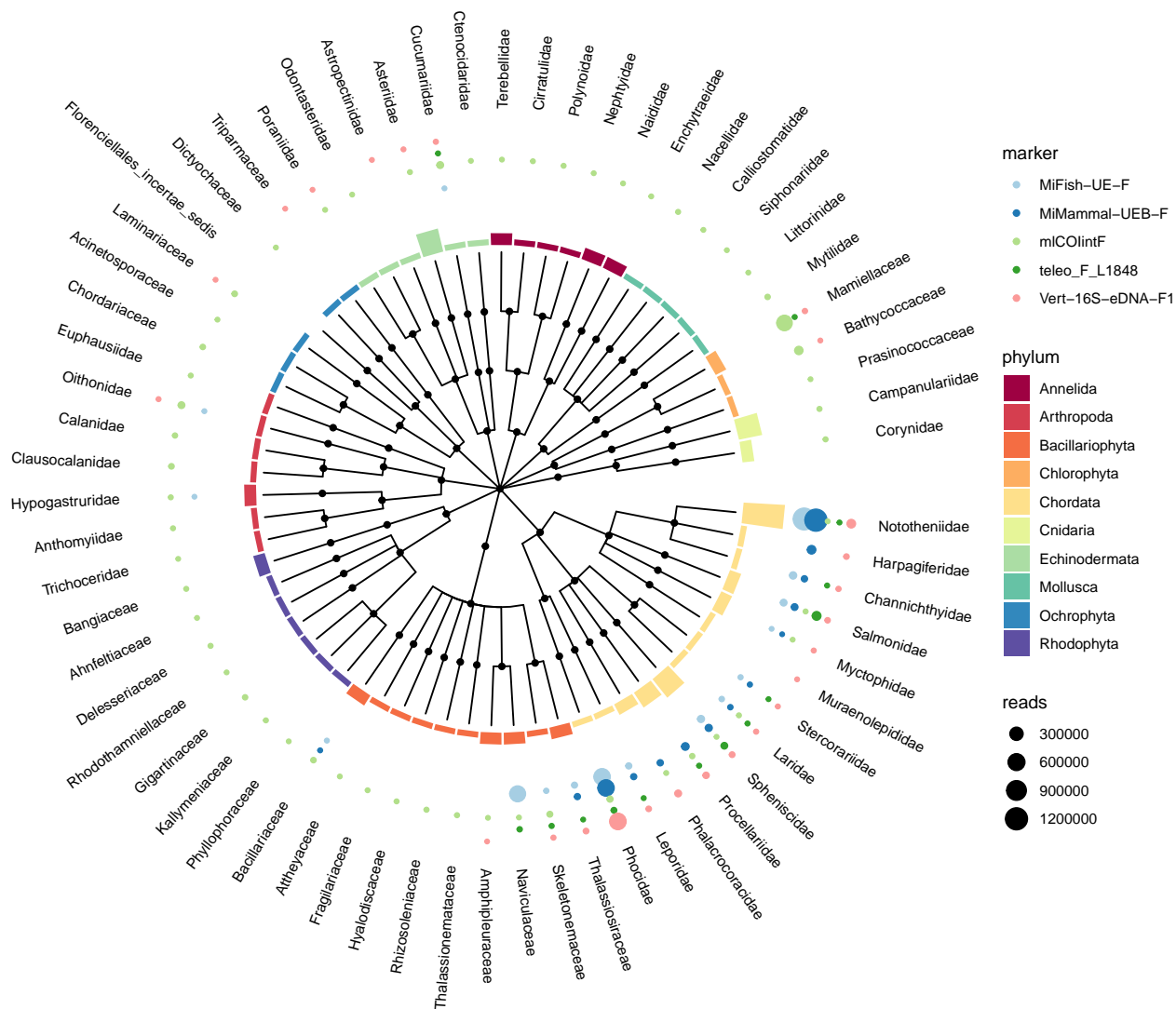


Figure 2: Number of DNA reads (bubbles) and species detected (bars) by family.

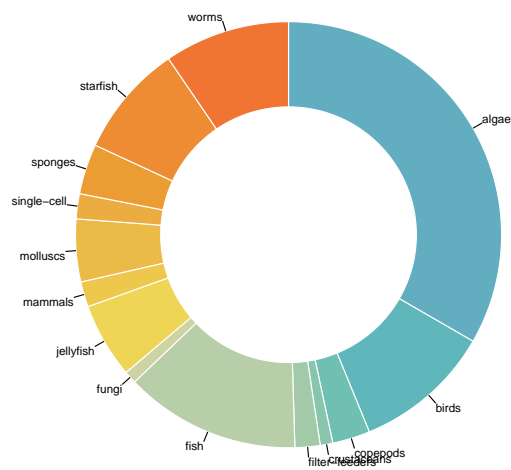


Figure 3: Distribution of detected species across groups.

phylum	class	species	group	category	new	vernacular
Amoebozoa	Discosea	Parvamoeba rugata	single-cell		yes	
Annelida	Clitellata	Cognettia varisetosa	worms		yes	
Annelida	Clitellata	Limnodrilus hoffmeisteri	worms		yes	
Annelida	Clitellata	Lumbricillus antarcticus	worms		yes	
Annelida	Clitellata	Tubifex tubifex	worms		yes	river worm
Annelida	Polychaeta	Aglaophamus trissophyllus	worms			
Annelida	Polychaeta	Amphitrite kerguelensis	worms			
Annelida	Polychaeta	Artacama crassa	worms		yes	
Annelida	Polychaeta	Cirratulus balaenophilus	worms		yes	
Annelida	Polychaeta	Harmothoe magellanica	worms			
Arthropoda	Copepoda	Calanus simillimus	copepods			
Arthropoda	Copepoda	Ctenocalanus citer	copepods		yes	
Arthropoda	Copepoda	Oithona similis	copepods			
Arthropoda	Hexapoda	Fucellia tergina				
Arthropoda	Hexapoda	Hypogastrura purpurescens			yes	
Arthropoda	Hexapoda	Hypogastrura viatica				
Arthropoda	Hexapoda	Trichocera maculipennis			yes	
Arthropoda	Malacostraca	Euphausia vallentini	crustaceans			
Bacillariophyta	Bacillariophyceae	Attheya longicornis	algae		yes	
Bacillariophyta	Bacillariophyceae	Cylindrotheca closterium	algae		yes	
Bacillariophyta	Bacillariophyceae	Frustulia vulgaris	algae		yes	
Bacillariophyta	Bacillariophyceae	Grammonema striatula	algae		yes	
Bacillariophyta	Bacillariophyceae	Halamphora calidilacuna	algae		yes	
Bacillariophyta	Bacillariophyceae	Navicula glaciei	algae		yes	
Bacillariophyta	Bacillariophyceae	Navicula minima	algae		yes	
Bacillariophyta	Bacillariophyceae	Nitzschia palea	algae		yes	
Bacillariophyta	Bacillariophyceae	Podosira stelligera	algae		yes	
Bacillariophyta	Bacillariophyceae	Skeletonema dohrnii	algae		yes	
Bacillariophyta	Bacillariophyceae	Sundstroemia setigera	algae		yes	
Bacillariophyta	Bacillariophyceae	Thalassionema nitzschioides	algae			
Bacillariophyta	Bacillariophyceae	Thalassiosira nordenskiöldii	algae		yes	
Bacillariophyta	Bacillariophyceae	Thalassiosira rotula	algae		yes	
Bigyra	Bicoecea	Cafeteria roenbergensis			yes	
Bryozoa	Gymnolaemata	Antarctothoa dictyota	filter-feeders		yes	
Bryozoa	Gymnolaemata	Austrothoa yagana	filter-feeders		yes	
Chlorophyta	Mamiellophyceae	Bathycoccus prasinos	algae		yes	
Chlorophyta	Mamiellophyceae	Micromonas commoda	algae		yes	
Chlorophyta	Mamiellophyceae	Micromonas pusilla	algae		yes	
Chlorophyta	Pyramimonadophyceae	Prasinoderma coloniale	algae		yes	
Chordata	Aves	Aptenodytes patagonicus	birds	LC		king penguin, manchot royal
Chordata	Aves	Daption capense	birds	LC		Cape petrel, damier du Cap
Chordata	Aves	Eudyptes chrysocome	birds	VU		western rockhopper penguin, gorfou sauteur
Chordata	Aves	Eudyptes filholi	birds			Southern rockhopper penguin, Gorfou de Filhol
Chordata	Aves	Halobaena caerulea	birds	LC		blue petrel, prion bleu
Chordata	Aves	Larus dominicanus	birds	LC		kelp gull, goéland dominicain
Chordata	Aves	Leucocarbo atriceps	birds	LC		
Chordata	Aves	Leucocarbo chalconotus	birds	VU	yes	Otago shag
Chordata	Aves	Pterodroma hasitata	birds	EN	yes	black-capped petrel, Pétrel diabolotin
Chordata	Aves	Pygoscelis papua	birds	LC		gentoo penguin, Manchot papou
Chordata	Aves	Stercorarius skua	birds			great skua, stercoraire brun

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phylum	class	species	group	category	new	vernacular
Chordata	Mammalia	Mirounga leonina	mammals	LC		southern elephant seal
Chordata	Mammalia	Oryctolagus cuniculus	mammals	EN	yes	
Chordata	Teleostei	Channichthys rhinoceratus	fish			unicorn icefish
Chordata	Teleostei	Electrona carlsbergi	fish	LC		
Chordata	Teleostei	Gobionotothen acuta	fish			triangular notothen
Chordata	Teleostei	Gobionotothen gibberifrons	fish		yes	humped rockcod
Chordata	Teleostei	Gymnoscopelus braueri	fish	LC		
Chordata	Teleostei	Harpagifer antarcticus	fish		yes	Antarctic spiny plunderfish
Chordata	Teleostei	Lindbergichthys nudifrons	fish		yes	
Chordata	Teleostei	Muraenolepis marmorata	fish			
Chordata	Teleostei	Notothenia coriiceps	fish			yellowbelly rockcod
Chordata	Teleostei	Notothenia rossii	fish			marbled rockcod
Chordata	Teleostei	Pagothenia borchgrevinki	fish		yes	bald notothen
Chordata	Teleostei	Paranotothenia magellanica	fish			Maori cod
Chordata	Teleostei	Salmo trutta	fish	LC		brown trout, truite brune
Chordata	Teleostei	Salvelinus fontinalis	fish			brook trout, saumon de fontaine
Cnidaria	Hydrozoa	Campanularia lennoxensis	jellyfish		yes	
Cnidaria	Hydrozoa	Clytia gracilis	jellyfish		yes	
Cnidaria	Hydrozoa	Coryne eximia	jellyfish		yes	
Cnidaria	Hydrozoa	Obelia geniculata	jellyfish			knotted thread hydroid, obélie
Cnidaria	Hydrozoa	Silicularia rosea	jellyfish			
Cnidaria	Hydrozoa	Stauridiosarsia producta	jellyfish		yes	
Cryptophyta	Cryptophyceae	Chroomonas placoidea			yes	
Echinodermata	Asteroidea	Anasterias antarctica	starfish		yes	
Echinodermata	Asteroidea	Anasterias suteri	starfish		yes	
Echinodermata	Asteroidea	Diplasterias brucei	starfish			
Echinodermata	Asteroidea	Diplasterias meridionalis	starfish			
Echinodermata	Asteroidea	Glabraster antarctica	starfish			
Echinodermata	Asteroidea	Macroptychaster accrescens	starfish			
Echinodermata	Asteroidea	Odontaster penicillatus	starfish			
Echinodermata	Echinoidea	Aporocidaris milleri	starfish			Miller's sea urchin, oursin de Miller
Echinodermata	Holothuroidea	Staurocucumis liouvillei	starfish			
Foraminifera	Globothalamea	Virgulinea fragilis				
Haptophyta	Coccolithophyceae	Emiliania huxleyi	algae		yes	
Haptophyta	Coccolithophyceae	Phaeocystis antarctica	algae		yes	
Mollusca	Bivalvia	Aulacomya atra	molluscs			
Mollusca	Gastropoda	Laevilitorina caliginosa	molluscs			
Mollusca	Gastropoda	Margarella antarctica	molluscs		yes	
Mollusca	Gastropoda	Nacella macquariensis	molluscs		yes	
Mollusca	Gastropoda	Siphonaria fuegiensis	molluscs		yes	
Myxozoa	Dinophyceae	Heterocapsa rotundata	single-cell		yes	
Nemertea	Pilidiophora	Parborlasia corrugatus	worms			
Ochrophyta	Bolidophyceae	Triparma laevis	algae		yes	
Ochrophyta	Dictyochophyceae	Florenciaella parvula	algae		yes	
Ochrophyta	Dictyochophyceae	Octactis speculum	algae		yes	
Ochrophyta	Dictyochophyceae	Pseudochattonella farcimen	algae		yes	
Ochrophyta	Phaeophyceae	Macrocystis pyrifera	algae			
Ochrophyta	Phaeophyceae	Myrionema strangulans	algae		yes	
Ochrophyta	Phaeophyceae	Pylaiella washingtoniensis	algae		yes	
Oomycota	Peronosporae	Eurychasma dicksonii	fungi		yes	

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phylum	class	species	group	category	new	vernacular
Porifera	Demospongiae	Halichondria (Halichondria) panicea	sponges			breadcrumb sponge
Porifera	Demospongiae	Halisarca dujardini	sponges			soft horny sponge, éponge de Dujardin
Porifera	Demospongiae	Hemigellius fimbriatus	sponges		yes	
Porifera	Demospongiae	Stylocordyla chupachups	sponges		yes	
Rhodophyta	Bangiophyceae	Neoporphyrha haitanensis	algae		yes	
Rhodophyta	Bangiophyceae	Wildemia amplissima	algae		yes	
Rhodophyta	Florideophyceae	Ahnfeltia plicata	algae			
Rhodophyta	Florideophyceae	Callophyllis atrosanguinea	algae			
Rhodophyta	Florideophyceae	Gymnogongrus turquetii	algae			
Rhodophyta	Florideophyceae	Iridaea cordata	algae			
Rhodophyta	Florideophyceae	Neuroglossum multilobum	algae		yes	
Rhodophyta	Florideophyceae	Rhodothamniella floridula	algae		yes	