

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

Biodiversity survey for The Sundarbans

Interim Results
Not for public distribution



Credit: Sher-e-Bangla Agricultural University.

About eDNA Expeditions

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 extracting this DNA from water samples, and subsequently multiplying and sequencing specific regions within these DNA strands, a wide variety of species can be detected without removing any organisms from their environment. The regions in the DNA targeted for sequencing 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 such as fish. The UNESCO eDNA Expeditions initiative combines a number of markers tailored for marine vertebrates (fish, mammals, and turtles). A general marker was added to also allow detection of 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 and a press kit are available on the UNESCO website: <https://www.unesco.org/en/edna-expeditions>.

Results

Sampling and sample processing

eDNA sampling was conducted in The Sundarbans in January 2023. A total of 20 samples were collected at 5 locations in the park covering different habitats: sundarbans_1, sundarbans_2, sundarbans_3, sundarbans_4, and sundarbans_5 (Figure 1). Up to 1,800 mL of seawater was filtered through filter cartridges containing a filter with a 0.8 μ m pore size. After sampling, the filter cartridges were flushed with preservation liquid and shipped to UNESCO. DNA from the samples was extracted and amplified, and subsequently 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.

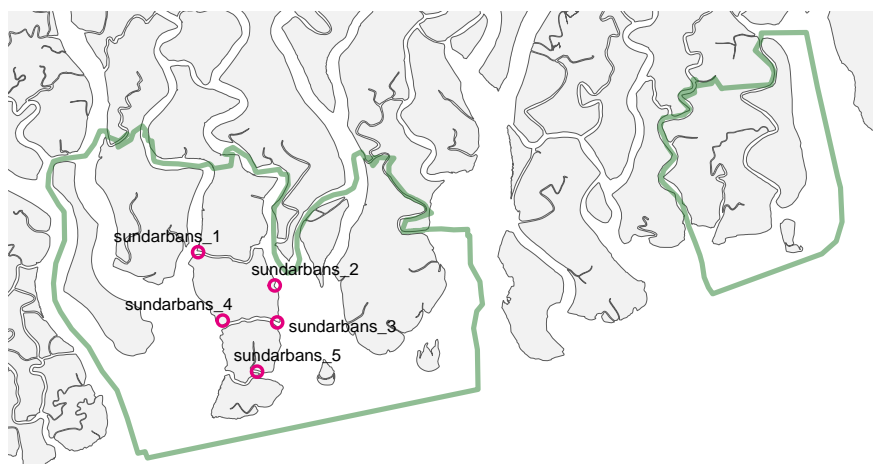


Figure 1: Map of the sampling locations.

DNA sequencing

Sequencing of the DNA from 20 samples resulted in over 25 million sequence reads, from which we collected 15,803 unique sequences or ASVs (Tables 1 and 2).

reads	asvs
25835775	15803

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

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 198 species, including 124 fish species, 5 mammalian species, and 0 turtle species (Table 3). Species from other groups, such as algae, copepods, crustaceans, cnidarians, and molluscs, were also detected (Figures 2 and 3). This number of species detected represents about 22% of the 596 species known from The Sundarbans in the OBIS database. Of the 198 species detected, 102 are not among the species previously reported from The Sundarbans to the UNESCO OBIS database. 9 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.

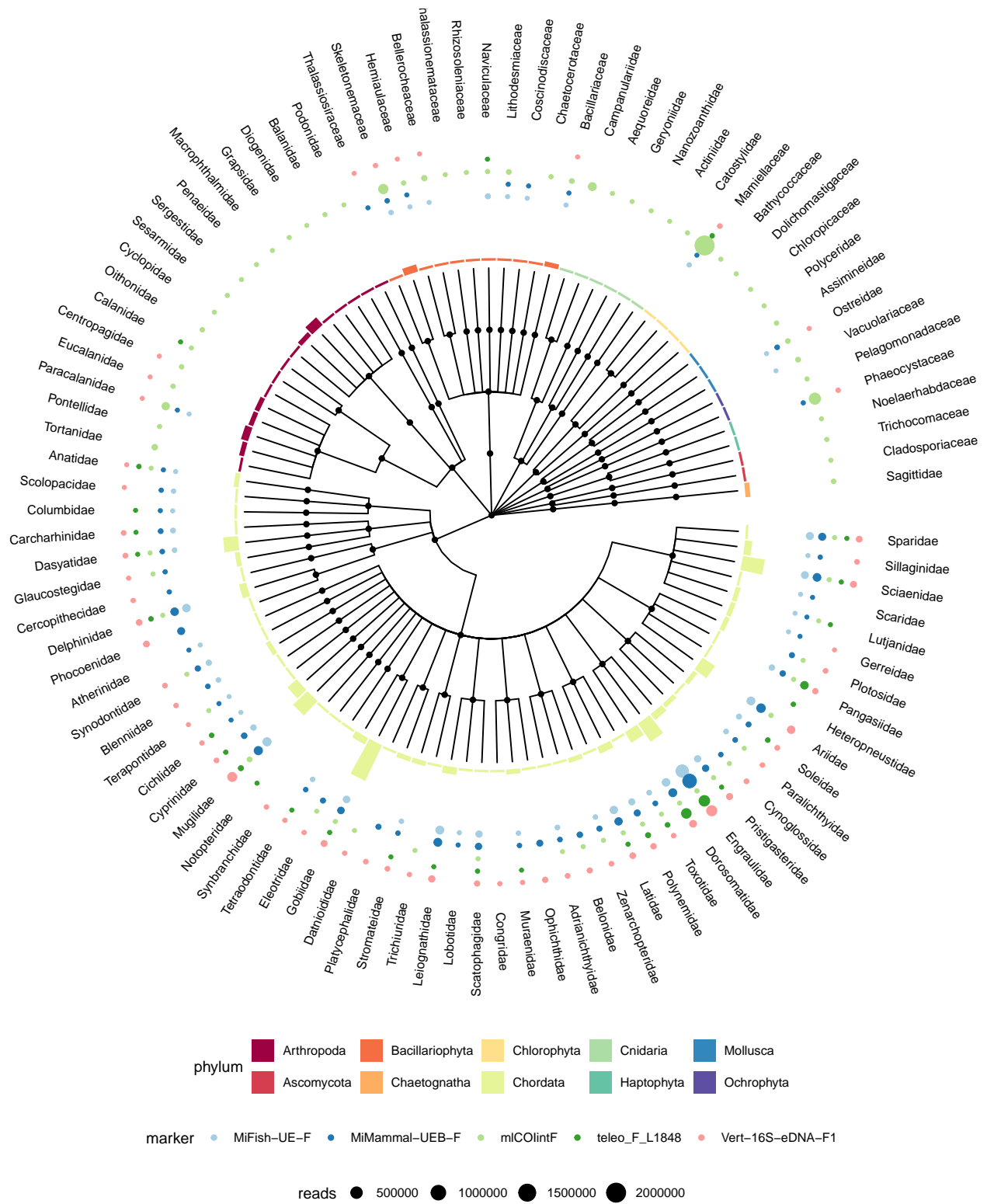


Figure 2: Number of DNA reads (bubbles) and species detected (bars) by family.

locality	materialSampleID	sampleSize	reads	asvs	species
Sundarban West Wildlife Sanctuary	EE0332	1800	1444767	5956	67
Sundarban west wildlife sanctuary	EE0330	1800	1753183	4600	67
Sundarban west wildlife sanctuary	EE0331	1800	767271	4386	64
Sundarban west wildlife sanctuary	EE0334	1800	1869098	4892	86
Sundarban west wildlife sanctuary	EE0337	1800	1438481	4869	61
Sundarban west wildlife sanctuary	EE0347	1800	1895505	6093	85
Sundarban west wildlife sanctuary	EE0360	1800	1067381	5121	68
Sundarban west wildlife sanctuary	EE0361	1800	1427194	4706	64
Sundarban west wildlife sanctuary	EE0363	1800	1459676	371	19
Sundarban west wildlife sanctuary	EE0364	1620	1440685	5154	61
Sundarban's west wildlife sanctuary	EE0351	1800	1348204	5025	78
Sundarbans West Wildlife Sanctuary	EE0345	1800	926509	4463	63
Sundarbans West Wildlife Sanctuary	EE0344	1800	18085	52	3
Sundarbans West Wildlife Sanctuary	EE0350	1800	874715	4310	83
Sundarbans West Wildlife Sanctuary	EE0365	1800	1392590	4346	72
Sundarbans west wildlife sanctuary	EE0336	1800	1786004	5816	83
Sundarbans west wildlife sanctuary	EE0338	1800	1158873	3972	56
Sundarbans west wildlife sanctuary	EE0339	1800	774806	2788	54
Sundarbans west wildlife sanctuary	EE0348	1800	1304487	4783	79
Sundarbans west wildlife sanctuary	EE0362	1800	1688261	5099	76

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

group	eDNA species	total species	fraction
fish	124	569	0.22
mammals	5	6	0.83

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	2	14	0.14
EN	5	14	0.36
VU	2	22	0.09

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

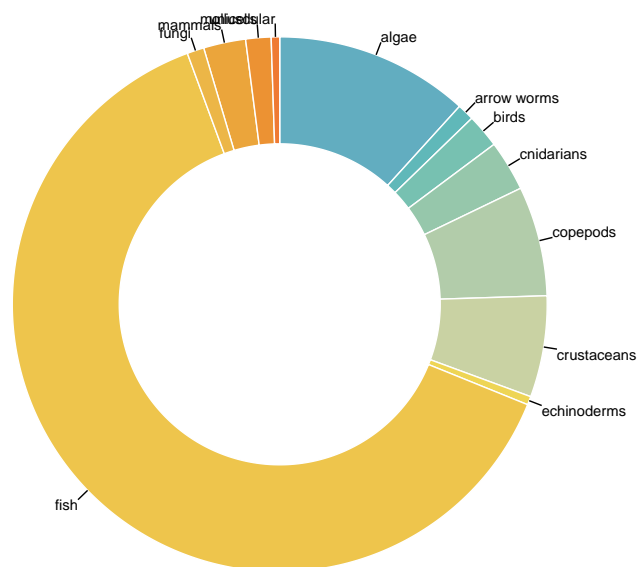


Figure 3: Distribution of detected species across groups.

phylum	class	species	group	category	new	vernacular
Arthropoda	Branchiopoda	Pseudevadne tergestina	crustaceans		yes	
Arthropoda	Copepoda	Bestiolina similis	copepods		yes	
Arthropoda	Copepoda	Canthocalanus pauper	copepods		yes	
Arthropoda	Copepoda	Centropages furcatus	copepods		yes	
Arthropoda	Copepoda	Centropages tenuiremis	copepods		yes	
Arthropoda	Copepoda	Labidocera pectinata	copepods		yes	
Arthropoda	Copepoda	Labidocera rotunda	copepods		yes	
Arthropoda	Copepoda	Oithona attenuata	copepods		yes	
Arthropoda	Copepoda	Paracalanus indicus	copepods		yes	
Arthropoda	Copepoda	Parvocalanus crassirostris	copepods		yes	
Arthropoda	Copepoda	Subeucalanus subcrassus	copepods		yes	
Arthropoda	Copepoda	Subeucalanus subtenuis	copepods		yes	
Arthropoda	Copepoda	Thermocyclops decipiens	copepods		yes	
Arthropoda	Copepoda	Tortanus (Tortanus) forcipatus	copepods		yes	
Arthropoda	Malacostraca	Acetes indicus	crustaceans		yes	
Arthropoda	Malacostraca	Acetes sibogae	crustaceans		yes	
Arthropoda	Malacostraca	Clibanarius padavensis	crustaceans		yes	
Arthropoda	Malacostraca	Episesarma versicolor	crustaceans		yes	
Arthropoda	Malacostraca	Macrophthalmus (Mareotis) tomentosus	crustaceans		yes	
Arthropoda	Malacostraca	Metapenaeus brevicornis	crustaceans		yes	yellow prawn
Arthropoda	Malacostraca	Metopograpsus latifrons	crustaceans		yes	
Arthropoda	Malacostraca	Mierspenaeopsis hardwickii	crustaceans			
Arthropoda	Malacostraca	Mierspenaeopsis sculptilis	crustaceans		yes	
Arthropoda	Malacostraca	Penaeus indicus	crustaceans			Indian white shrimp
Arthropoda	Thecostraca	Amphibalanus rhizophorae	crustaceans		yes	
Ascomycota	Dothideomycetes	Cladosporium herbarum	fungi		yes	
Ascomycota	Eurotiomycetes	Penicillium digitatum	fungi		yes	
Bacillariophyta	Bacillariophyceae	Bellerochea polymorpha	algae		yes	
Bacillariophyta	Bacillariophyceae	Chaetoceros socialis	algae		yes	
Bacillariophyta	Bacillariophyceae	Coscinodiscus granii	algae		yes	
Bacillariophyta	Bacillariophyceae	Cylindrotheca closterium	algae		yes	
Bacillariophyta	Bacillariophyceae	Ditylum brightwellii	algae		yes	
Bacillariophyta	Bacillariophyceae	Hemiaulus sinensis	algae		yes	
Bacillariophyta	Bacillariophyceae	Navicula tsukamotoi	algae		yes	
Bacillariophyta	Bacillariophyceae	Pseudo-nitzschia cuspidata	algae		yes	
Bacillariophyta	Bacillariophyceae	Skeletonema ardens	algae		yes	
Bacillariophyta	Bacillariophyceae	Skeletonema costatum	algae		yes	
Bacillariophyta	Bacillariophyceae	Skeletonema menzelii	algae		yes	
Bacillariophyta	Bacillariophyceae	Sundstroemia setigera	algae		yes	
Bacillariophyta	Bacillariophyceae	Thalassionema nitzschioides	algae		yes	
Bacillariophyta	Bacillariophyceae	Thalassiosira rotula	algae		yes	
Basidiomycota	Agaricomycetes	Schizophyllum commune			yes	
Chaetognatha	Sagittoidea	Flaccisagitta enflata	arrow worms		yes	
Chaetognatha	Sagittoidea	Zonosagitta bedoti	arrow worms		yes	
Chlorophyta	Chloropicophyceae	Chloroparvula pacifica	algae		yes	
Chlorophyta	Mamiellophyceae	Bathycoccus prasinus	algae		yes	
Chlorophyta	Mamiellophyceae	Dolichomastix tenuilepis	algae		yes	
Chlorophyta	Mamiellophyceae	Micromonas pusilla	algae		yes	
Chlorophyta		Chloropicon roscoffensis	algae		yes	
Chordata	Aves	Anas poecilorhyncha	birds	LC	yes	

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phylum	class	species	group	category	new	vernacular
Chordata	Aves	Anser cygnoides	birds		yes	
Chordata	Aves	Columba livia	birds	LC	yes	
Chordata	Aves	Numenius phaeopus	birds	LC		whimbrel, courlis corllieu
Chordata	Elasmobranchii	Brevitrygon imbricata	fish	VU		
Chordata	Elasmobranchii	Brevitrygon walga	fish	NT		
Chordata	Elasmobranchii	Glaucostegus granulatus	fish	CR		
Chordata	Elasmobranchii	Glaucostegus obtusus	fish	CR		
Chordata	Elasmobranchii	Himantura uarnak	fish	EN		honeycomb stingray
Chordata	Elasmobranchii	Himantura undulata	fish	EN		
Chordata	Elasmobranchii	Pastinachus solocirostris	fish	EN	yes	
Chordata	Elasmobranchii	Scoliodon laticaudus	fish	NT		spadenose shark
Chordata	Elasmobranchii	Urogymnus polylepis	fish	EN	yes	
Chordata	Mammalia	Macaca mulatta	mammals	LC	yes	
Chordata	Mammalia	Neophocaena phocaenoides	mammals	VU	yes	Indian dolphin, marsouin aptere
Chordata	Mammalia	Orcaella brevirostris	mammals	EN		Irrawaddy dolphin, orcelle
Chordata	Mammalia	Stenella coeruleoalba	mammals	LC	yes	striped dolphin, dauphin bleu et blanc
Chordata	Mammalia	Tursiops aduncus	mammals	NT	yes	Indo-Pacific bottlenose dolphin, tursiops abusalam
Chordata	Teleostei	Acanthopagrus datnia	fish			
Chordata	Teleostei	Acentrogobius viridipunctatus	fish	LC		
Chordata	Teleostei	Anodontostoma chacunda	fish	LC		
Chordata	Teleostei	Apocryptes bato	fish	LC		
Chordata	Teleostei	Arius arius	fish	LC		
Chordata	Teleostei	Atherinomorus lacunosus	fish	LC	yes	robust silverside
Chordata	Teleostei	Bangana dero	fish	LC	yes	
Chordata	Teleostei	Bathygobius cocosensis	fish	LC	yes	
Chordata	Teleostei	Bathygobius fuscus	fish	LC		dusky frill-goby
Chordata	Teleostei	Boleophthalmus boddarti	fish	LC		
Chordata	Teleostei	Boleophthalmus pectinirostris	fish		yes	
Chordata	Teleostei	Butis humeralis	fish			
Chordata	Teleostei	Butis koilomatodon	fish	LC	yes	mud sleeper
Chordata	Teleostei	Butis melanostigma	fish			
Chordata	Teleostei	Chlorurus sordidus	fish	LC	yes	
Chordata	Teleostei	Chrysochir aureus	fish	LC		
Chordata	Teleostei	Cirrhinus mrigala	fish	LC	yes	
Chordata	Teleostei	Coilia dussumieri	fish	LC		
Chordata	Teleostei	Coilia ramcarati	fish	DD		
Chordata	Teleostei	Coilia reynaldi	fish	LC		
Chordata	Teleostei	Coptodon zillii	fish	LC	yes	
Chordata	Teleostei	Cynoglossus lingua	fish	LC		
Chordata	Teleostei	Datnioides polota	fish	LC		
Chordata	Teleostei	Dichotomyctere nigroviridis	fish	LC	yes	
Chordata	Teleostei	Eleutheronema rhadinum	fish		yes	East Asian fourfinger threadfin, barbure à quatre doigts d'Asie
Chordata	Teleostei	Eleutheronema tetradactylum	fish			blunt-nosed salmon, barbure à quatre doigts
Chordata	Teleostei	Entomacrodus striatus	fish	LC	yes	reef margin blenny
Chordata	Teleostei	Escualosa thoracata	fish	LC		
Chordata	Teleostei	Eucinostomus gula	fish	LC	yes	Jenny mojarra
Chordata	Teleostei	Gerres longirostris	fish	LC		
Chordata	Teleostei	Glossogobius aureus	fish	LC	yes	
Chordata	Teleostei	Glossogobius giuris	fish	LC		tank goby
Chordata	Teleostei	Gobiopsis macrostoma	fish			

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phylum	class	species	group	category	new	vernacular
Chordata	Teleostei	Gymnothorax tile	fish	LC		
Chordata	Teleostei	Harpadon nehereus	fish	NT		Bombay-duck
Chordata	Teleostei	Heteropneustes fossilis	fish	LC		
Chordata	Teleostei	Hilsa kelee	fish	LC		Kelee shad
Chordata	Teleostei	Hypophthalmichthys nobilis	fish	DD	yes	bighead
Chordata	Teleostei	Ilisha elongata	fish	LC		elongate ilisha
Chordata	Teleostei	Ilisha melastoma	fish	LC		
Chordata	Teleostei	Johnius amblycephalus	fish	LC	yes	bearded croaker
Chordata	Teleostei	Johnius belangerii	fish	LC		
Chordata	Teleostei	Johnius borneensis	fish	LC		
Chordata	Teleostei	Johnius carouna	fish	LC	yes	
Chordata	Teleostei	Johnius plagiostoma	fish	LC		
Chordata	Teleostei	Labeo bata	fish	LC	yes	
Chordata	Teleostei	Lates calcarifer	fish	LC		barramundi, perche barramundi
Chordata	Teleostei	Leiognathus brevisrostris	fish			
Chordata	Teleostei	Lepturacanthus savala	fish			
Chordata	Teleostei	Lobotes surinamensis	fish	LC		tripletail
Chordata	Teleostei	Lutjanus argentimaculatus	fish	LC		river snapper
Chordata	Teleostei	Lutjanus johnii	fish	LC		John's snapper
Chordata	Teleostei	Mugil cephalus	fish	LC		grey mullet, muge
Chordata	Teleostei	Nemapteryx caelata	fish			
Chordata	Teleostei	Nemapteryx nenga	fish			
Chordata	Teleostei	Notopterus notopterus	fish	LC		
Chordata	Teleostei	Nuclequula blochii	fish			
Chordata	Teleostei	Nuclequula nuchalis	fish		yes	
Chordata	Teleostei	Odontamblyopus rubicundus	fish	LC		
Chordata	Teleostei	Oligolepis acutipennis	fish	LC		sharptail goby
Chordata	Teleostei	Omobranchus zebra	fish	LC	yes	
Chordata	Teleostei	Ophisternon bengalense	fish	LC		
Chordata	Teleostei	Oryzias javanicus	fish	LC	yes	
Chordata	Teleostei	Osteogeneiosus militaris	fish			
Chordata	Teleostei	Osteomugil cunnesius	fish			
Chordata	Teleostei	Otolithoides pama	fish	DD		
Chordata	Teleostei	Pampus argenteus	fish			Silver pomfret
Chordata	Teleostei	Pampus chinensis	fish			
Chordata	Teleostei	Pangasius pangasius	fish	LC		
Chordata	Teleostei	Paramugil parmatius	fish	LC		
Chordata	Teleostei	Paraplagusia blochii	fish		yes	
Chordata	Teleostei	Paraplotosus albilabris	fish		yes	
Chordata	Teleostei	Parapocryptes serperaster	fish	LC		
Chordata	Teleostei	Pellona ditchela	fish	LC		Indian pellona
Chordata	Teleostei	Periophthalmus chrysospilos	fish	LC		
Chordata	Teleostei	Pisodonophis boro	fish	LC		rice-paddy eel
Chordata	Teleostei	Planiliza macrolepis	fish	LC		
Chordata	Teleostei	Planiliza melinoptera	fish			
Chordata	Teleostei	Planiliza planiceps	fish	LC		
Chordata	Teleostei	Planiliza subviridis	fish	LC		
Chordata	Teleostei	Planiliza tade	fish	DD		
Chordata	Teleostei	Platycephalus cultellatus	fish		yes	
Chordata	Teleostei	Platycephalus indicus	fish	DD		Indian flathead

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phylum	class	species	group	category	new	vernacular
Chordata	Teleostei	Plicofollis argyroleuron	fish		yes	
Chordata	Teleostei	Plicofollis platystomus	fish	LC		
Chordata	Teleostei	Plotosus canius	fish			
Chordata	Teleostei	Polynemus paradiseus	fish	LC		paradise threadfin, barbure paradis
Chordata	Teleostei	Protonibea diacanthus	fish	NT		
Chordata	Teleostei	Pseudorhombus arsius	fish	LC		largetooth flounder
Chordata	Teleostei	Pterolithus maculatus	fish	LC		
Chordata	Teleostei	Puntius sophore	fish	LC		
Chordata	Teleostei	Sardinella albella	fish	LC	yes	white sardinella
Chordata	Teleostei	Scartelaos histophorus	fish	LC		
Chordata	Teleostei	Scatophagus argus	fish	LC		
Chordata	Teleostei	Setipinna taty	fish	LC		
Chordata	Teleostei	Setipinna tenuifilis	fish	DD		
Chordata	Teleostei	Sillaginopsis panijus	fish			
Chordata	Teleostei	Sillago maculata	fish		yes	
Chordata	Teleostei	Sillago sihama	fish	LC		silver sillago
Chordata	Teleostei	Solea ovata	fish	LC	yes	
Chordata	Teleostei	Stigmatogobius sadanundio	fish			
Chordata	Teleostei	Stolephorus dubiosus	fish	LC	yes	
Chordata	Teleostei	Stolephorus insularis	fish		yes	Hardenberg's anchovy
Chordata	Teleostei	Strongylura strongylura	fish			
Chordata	Teleostei	Strophidon sathete	fish	LC		slender giant moray
Chordata	Teleostei	Synaptura commersonnii	fish			
Chordata	Teleostei	Taenioides cirratus	fish	DD		
Chordata	Teleostei	Tenualosa ilisha	fish	LC		
Chordata	Teleostei	Therapon theraps	fish	LC		largescaled therapon
Chordata	Teleostei	Thryssa hamiltonii	fish	LC		
Chordata	Teleostei	Thryssa mystax	fish	LC	yes	
Chordata	Teleostei	Toxotes chatareus	fish	LC		
Chordata	Teleostei	Uroconger lepturus	fish	LC		slender conger
Chordata	Teleostei	Zenarchopterus buffonis	fish	LC		
Chordata	Teleostei	Zenarchopterus ectuntio	fish	LC		
Cnidaria	Hexacorallia	Nanozoanthus harenaceus	cnidarians		yes	
Cnidaria	Hexacorallia	Paracondylactis sinensis	cnidarians		yes	
Cnidaria	Hydrozoa	Aequorea conica	cnidarians		yes	
Cnidaria	Hydrozoa	Clytia folleata	cnidarians		yes	
Cnidaria	Hydrozoa	Liriope tetraphylla	cnidarians		yes	
Cnidaria	Scyphozoa	Acromitus flagellatus	cnidarians		yes	
Cryptophyta	Cryptophyta incertae sedis	Leucocryptos marina			yes	
Echinodermata	Ophiuroidea	Amphiplus (Lymanella) laevis	echinoderms		yes	
Haptophyta	Coccolithophyceae	Gephyrocapsa oceanica	algae		yes	
Haptophyta	Coccolithophyceae	Phaeocystis globosa	algae		yes	
Mollusca	Bivalvia	Saccostrea cucullata	molluscs		yes	
Mollusca	Gastropoda	Gymnodoris pattani	molluscs		yes	
Mollusca	Gastropoda	Pseudomphala latericea	molluscs		yes	
Myxozoa	Dinophyceae	Heterocapsa rotundata	unicellular		yes	
Ochrophyta	Pelagophyceae	Pelagomonas calceolata	algae		yes	
Ochrophyta	Raphidophyceae	Fibrocapsa japonica	algae		yes	