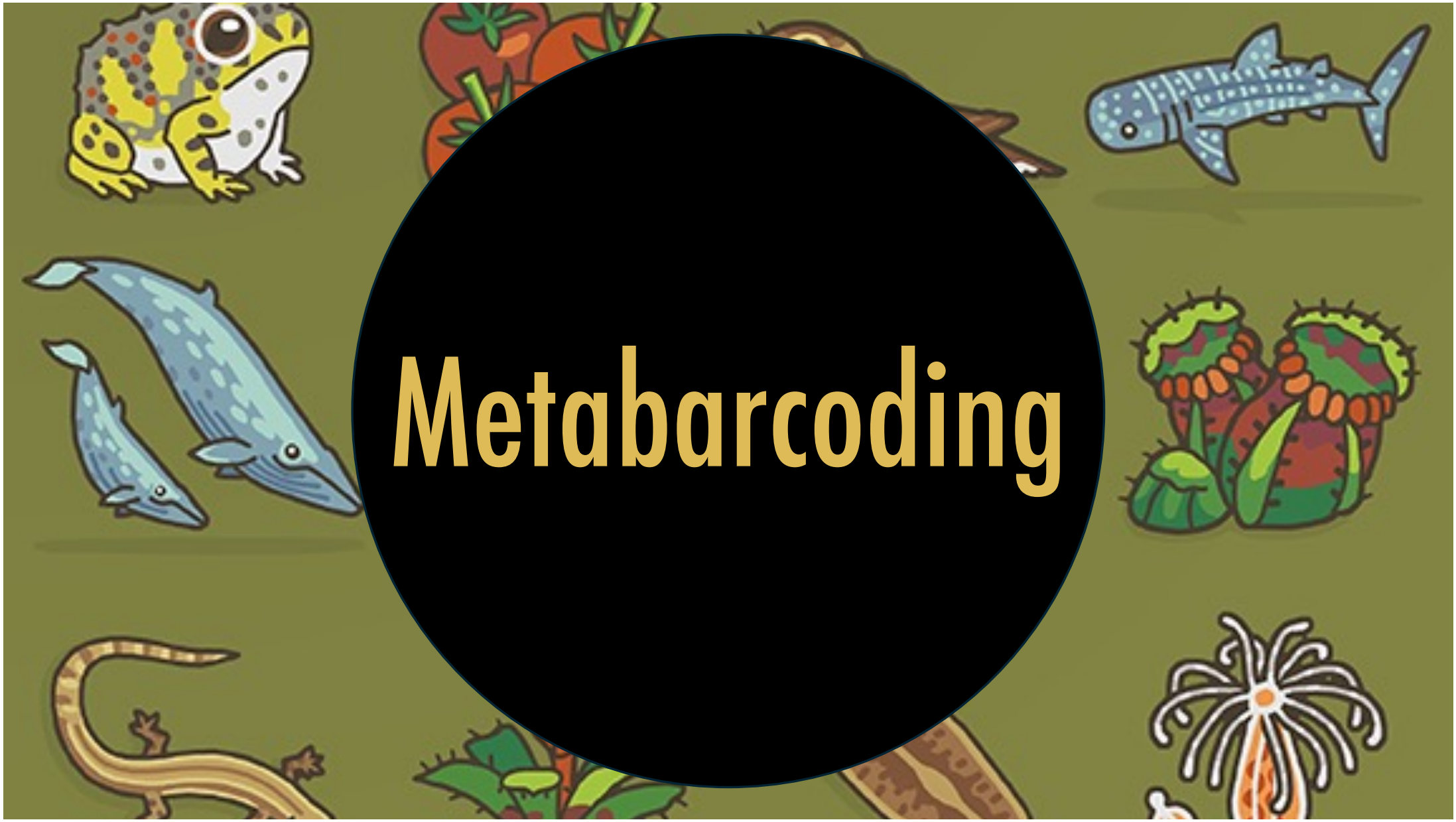


Metabarcoding

Who am I?

Anthropogenic biodiversity change

Community Ecology & Biogeography

Methods in sedaDNA and eDNA

Metabarcoding & metagenomics

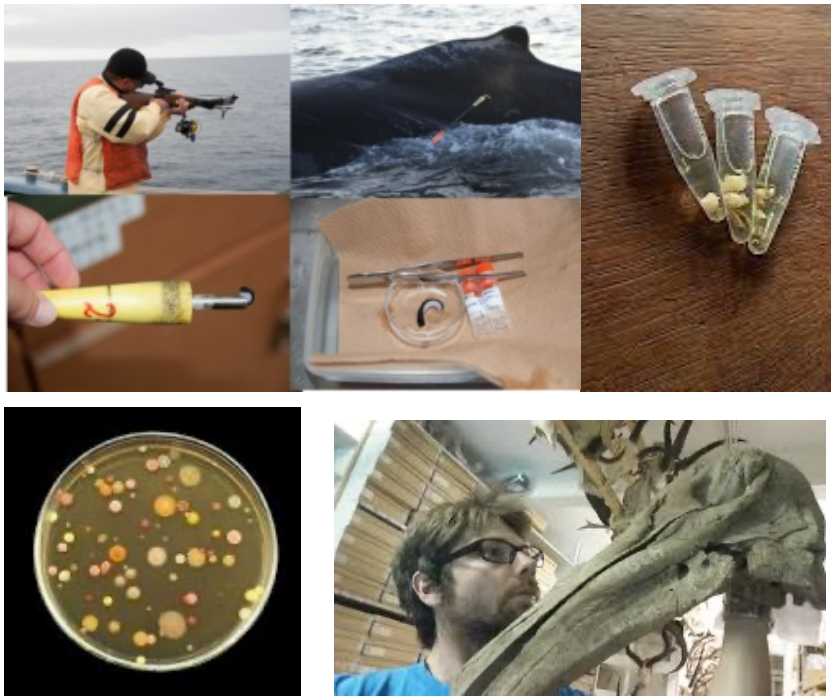
Globe Institute

Today

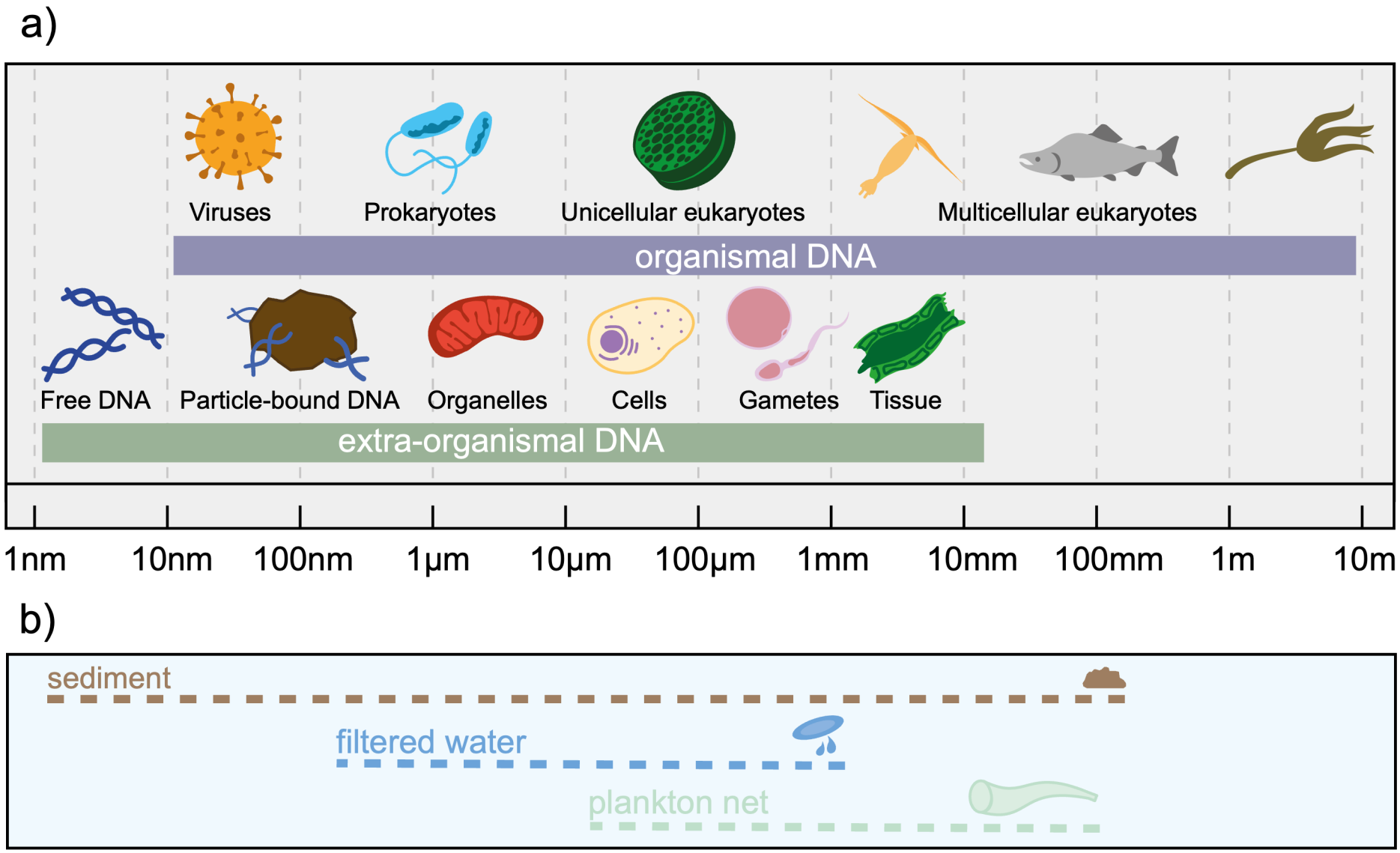
-Sample types
-What is metabarcoding?
-Sequence detective
-Metabarcoding libraries
-Bioinformatics overview

Next Generation Sequencing Sample Sources

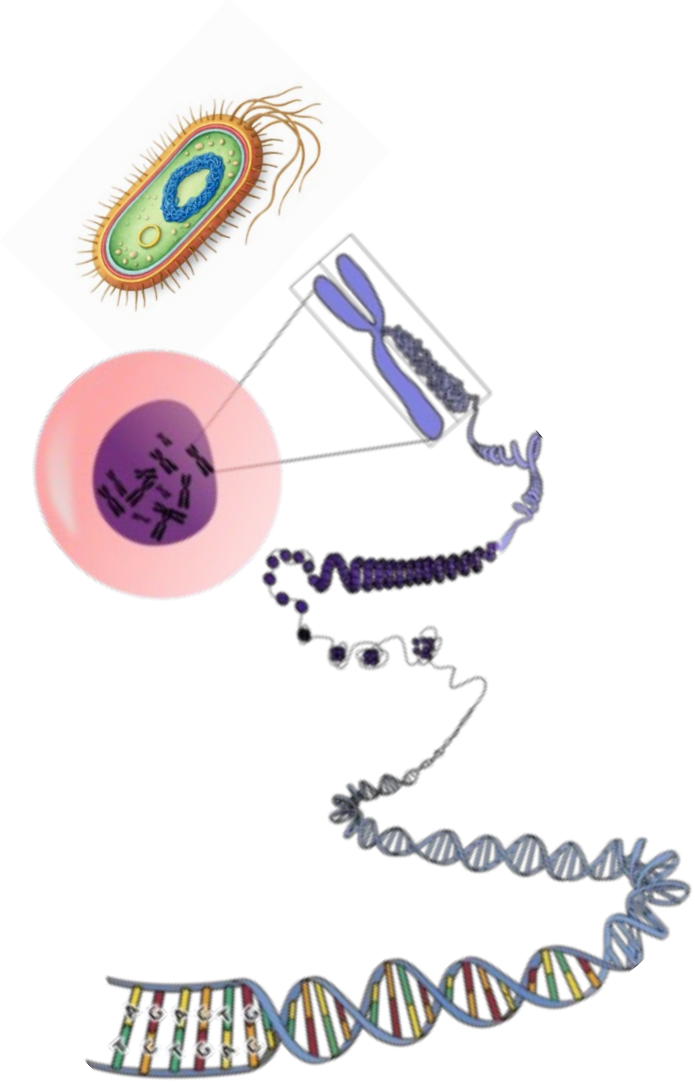
Point Source



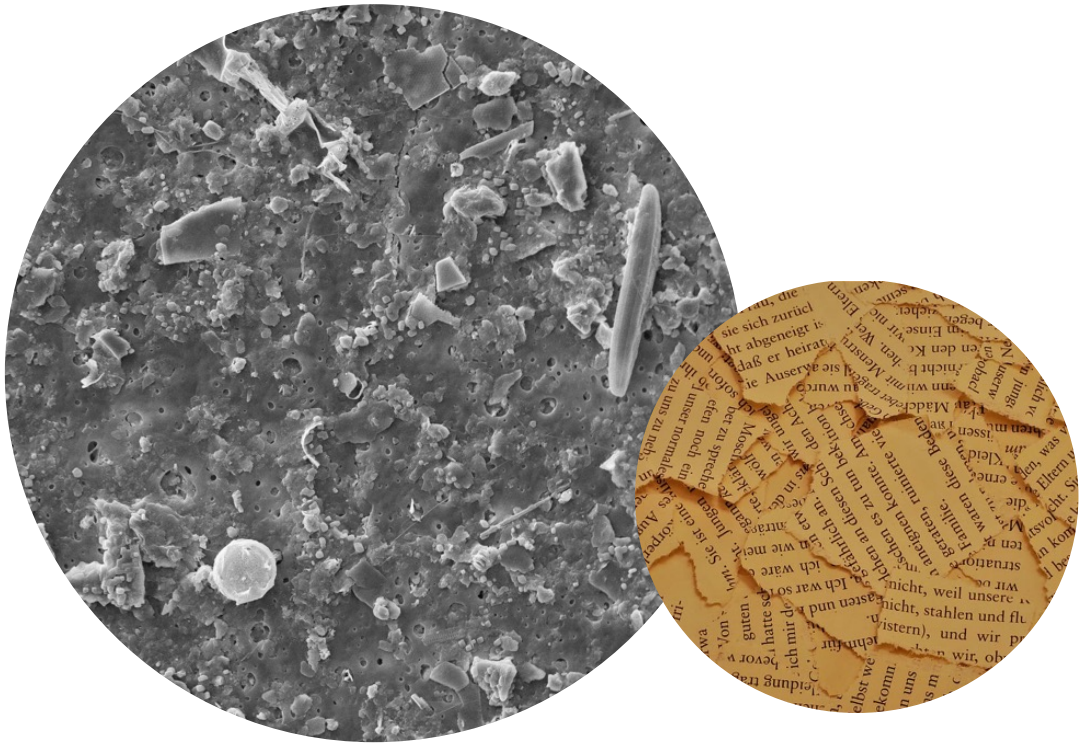
Mixed Source


a)
Viruses
Prokaryotes
Unicellular eukaryotes
Multicellular eukaryotes
organismal DNA
Free DNA
Particle-bound DNA
Organelles
Cells
Gametes
Tissue
extra-organismal DNA
1nm
10nm
100nm
1µm
10µm
100µm
1mm
10mm
100mm
1m
10m
b)
sediment
filtered water
plankton net

What's in these mixed samples?



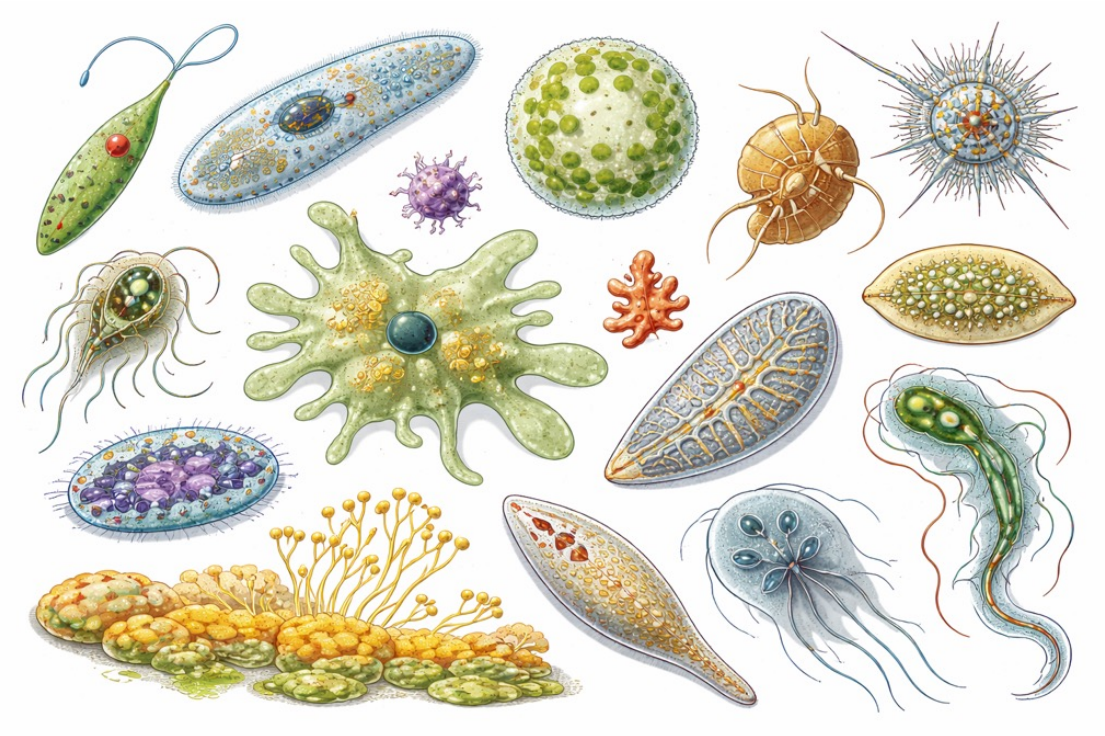
What's in these mixed samples?

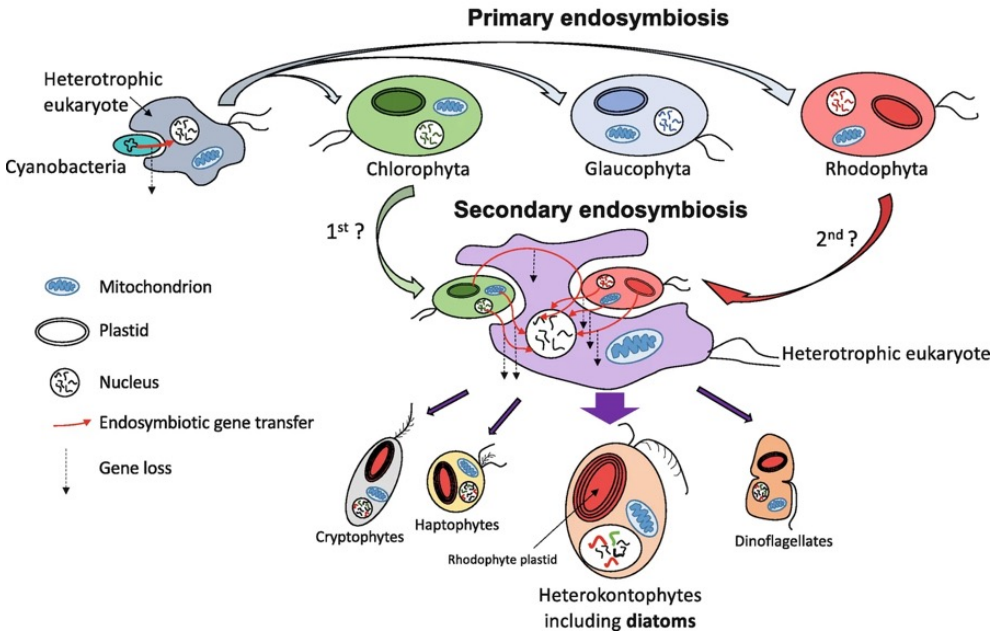


Animal genomes

Mock et al. 2022

What's in these mixed samples?

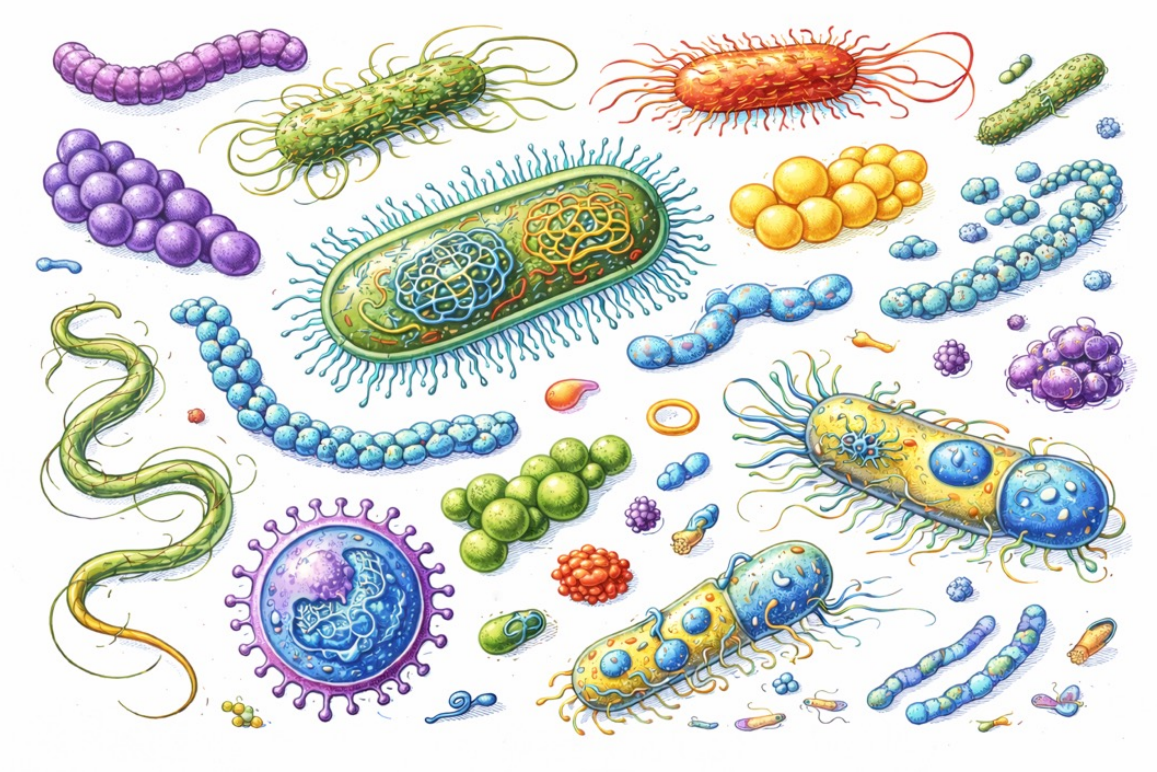


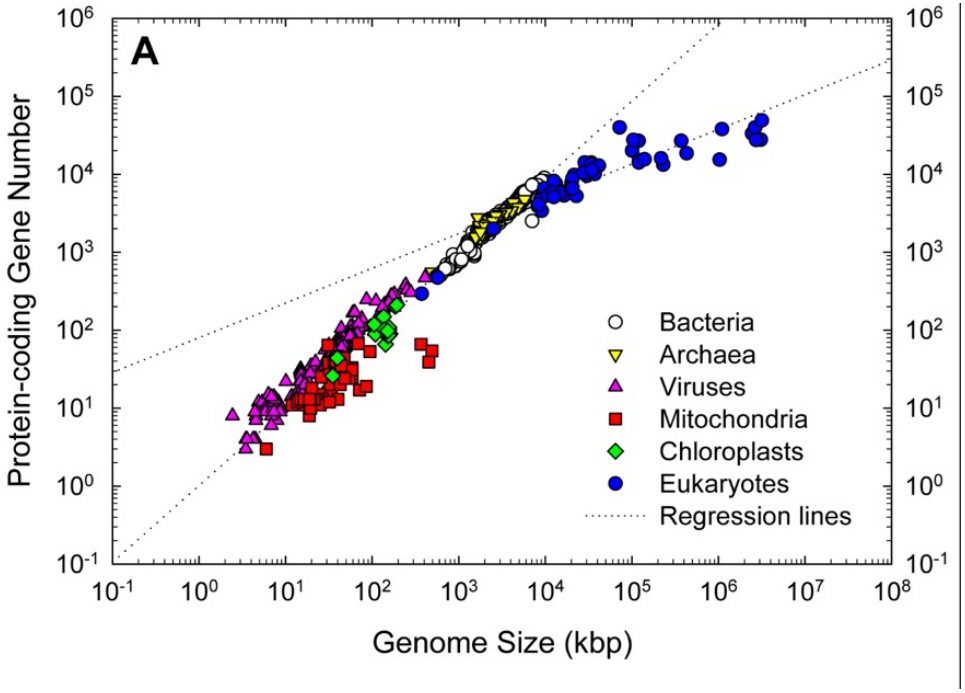


Protists

Mock et al. 2022

What's in these mixed samples?

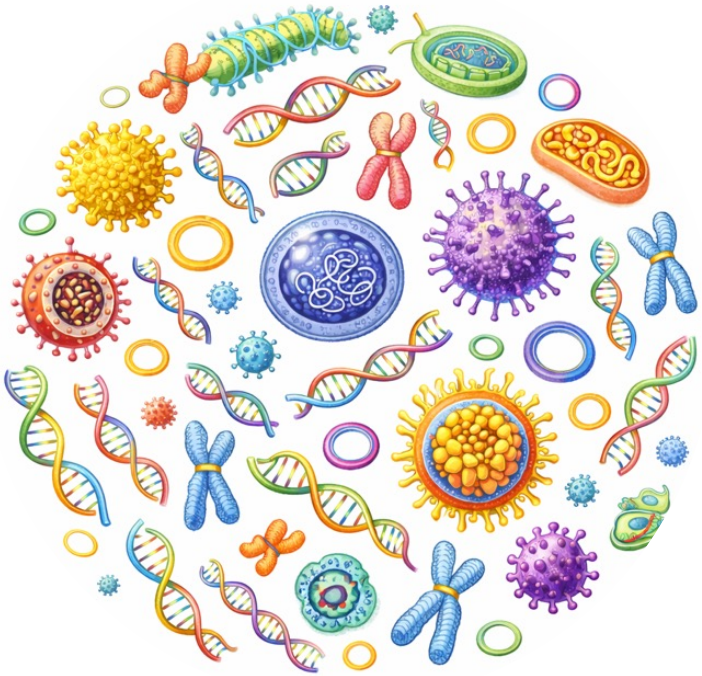




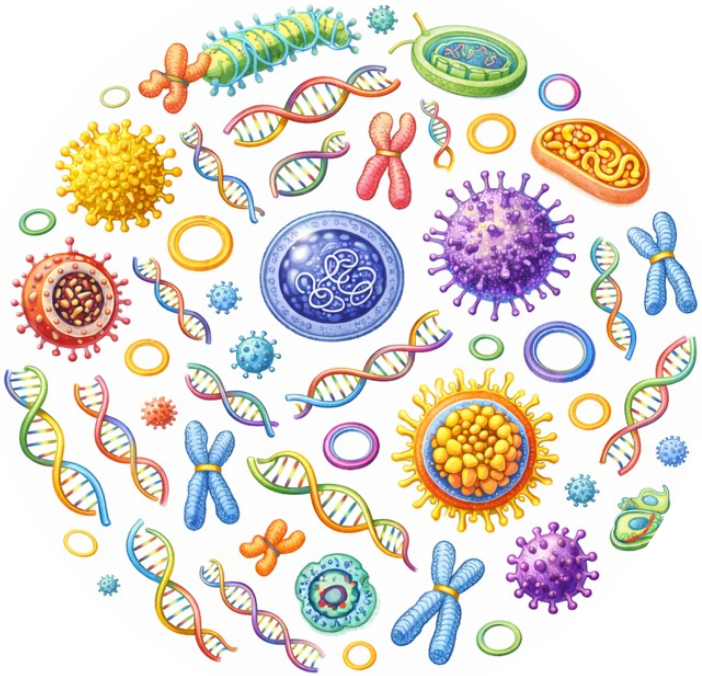
Hou & Lin 2009

Bacteria & Viruses

What taxa are in this sample?

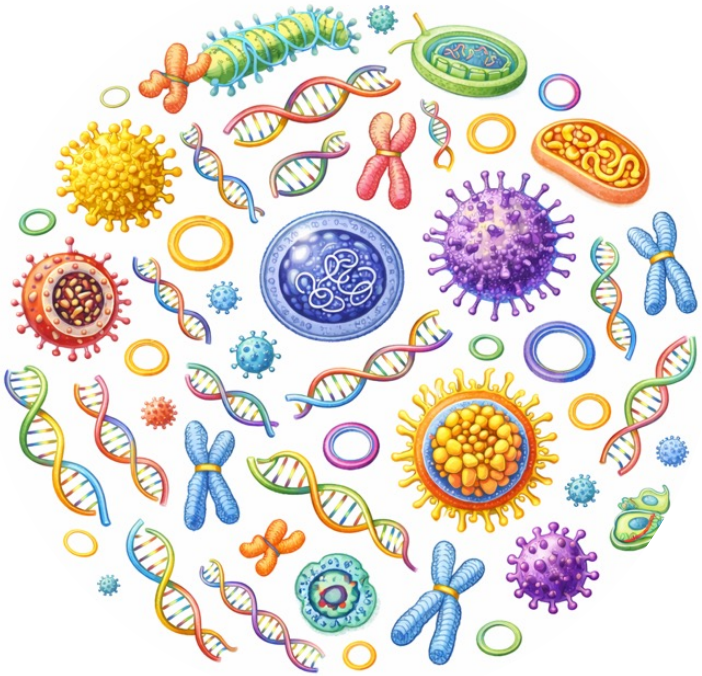


What taxa are in this sample?



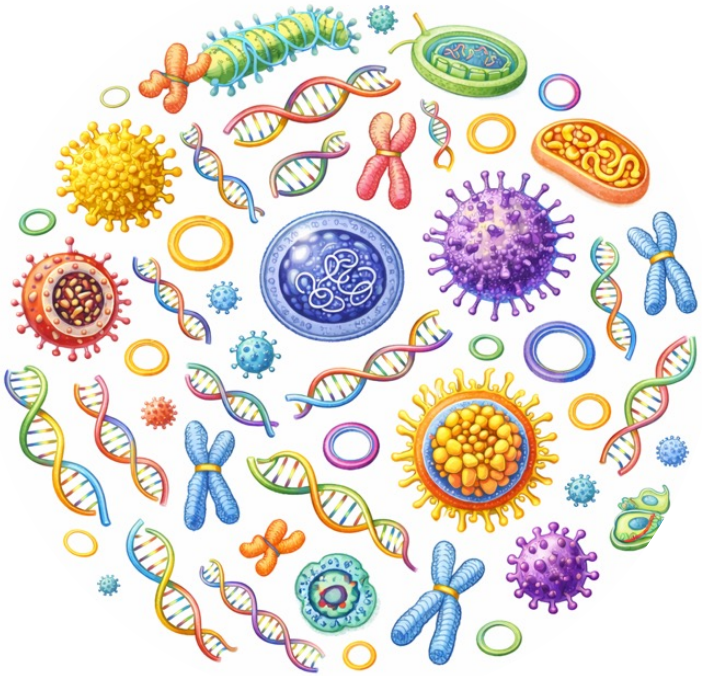
So... what shall we do?

What taxa are in this sample?



So... what shall we do?

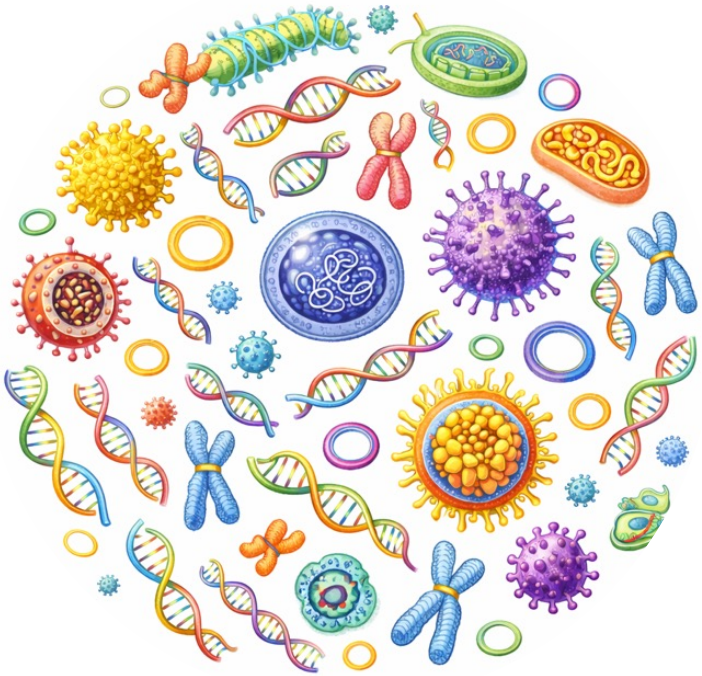
What taxa are in this sample?



Metagenomics

So... what shall we do?

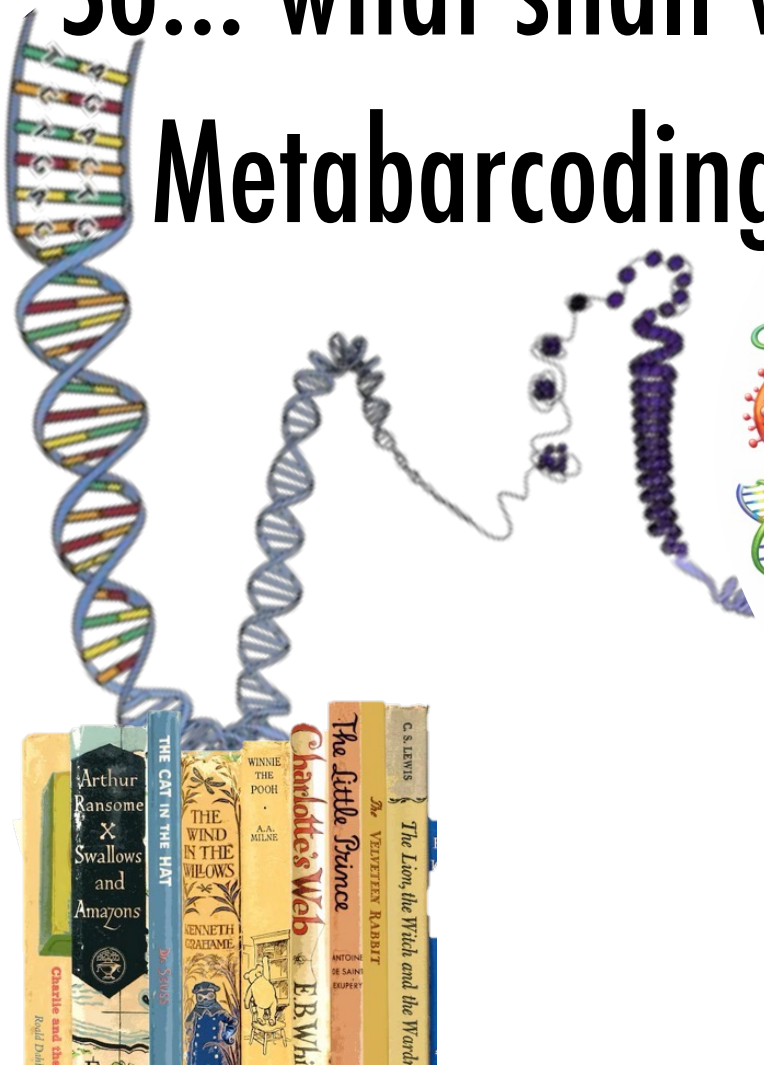
So... what shall we do?

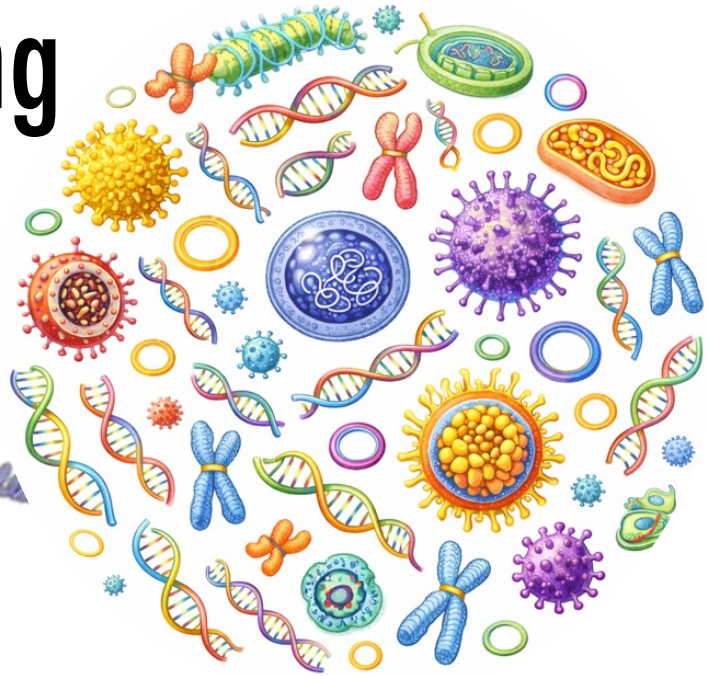


Metagenomics

So... what shall we do?

Metabarcoding



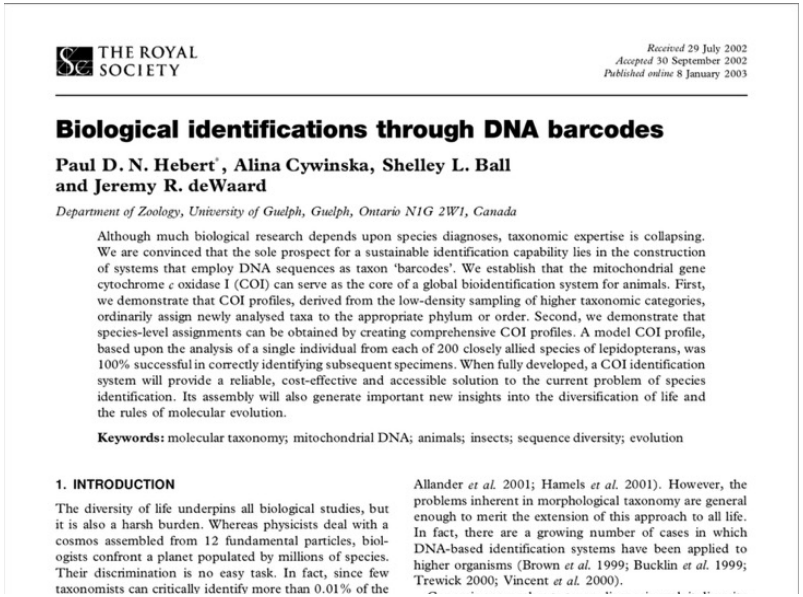


Metagenomics

DNA Barcoding

Prof. Paul Herbert

CBG



THE ROYAL SOCIETY

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Biological identifications through DNA barcodes

Paul D. N. Hebert*, Alina Cywinska, Shelley L. Ball and Jeremy R. deWaard

Department of Zoology, University of Guelph, Guelph, Ontario N1G 2W1, Canada

Although much biological research depends upon species diagnoses, taxonomic expertise is collapsing. We are convinced that the sole prospect for a sustainable identification capability lies in the construction of systems that employ DNA sequences as taxon 'barcodes'. We establish that the mitochondrial gene cytochrome *c* oxidase I (COI) can serve as the core of a global bioidentification system for animals. First, we demonstrate that COI profiles, derived from the low-density sampling of higher taxonomic categories, ordinarily assign newly analysed taxa to the appropriate phylum or order. Second, we demonstrate that species-level assignments can be obtained by creating comprehensive COI profiles. A model COI profile, based upon the analysis of a single individual from each of 200 closely allied species of lepidopterans, was 100% successful in correctly identifying subsequent specimens. When fully developed, a COI identification system will provide a reliable, cost-effective and accessible solution to the current problem of species identification. Its assembly will also generate important new insights into the diversification of life and the rules of molecular evolution.

Keywords: molecular taxonomy; mitochondrial DNA; animals; insects; sequence diversity; evolution

1. INTRODUCTION

The diversity of life underpins all biological studies, but it is also a harsh burden. Whereas physicists deal with a cosmos assembled from 12 fundamental particles, biologists confront a planet populated by millions of species. Their discrimination is no easy task. In fact, since few taxonomists can critically identify more than 0.01% of the

Allander *et al.* 2001; Hamels *et al.* 2001). However, the problems inherent in morphological taxonomy are general enough to merit the extension of this approach to all life. In fact, there are a growing number of cases in which DNA-based identification systems have been applied to higher organisms (Brown *et al.* 1999; Bucklin *et al.* 1999; Trewick 2000; Vincent *et al.* 2000).

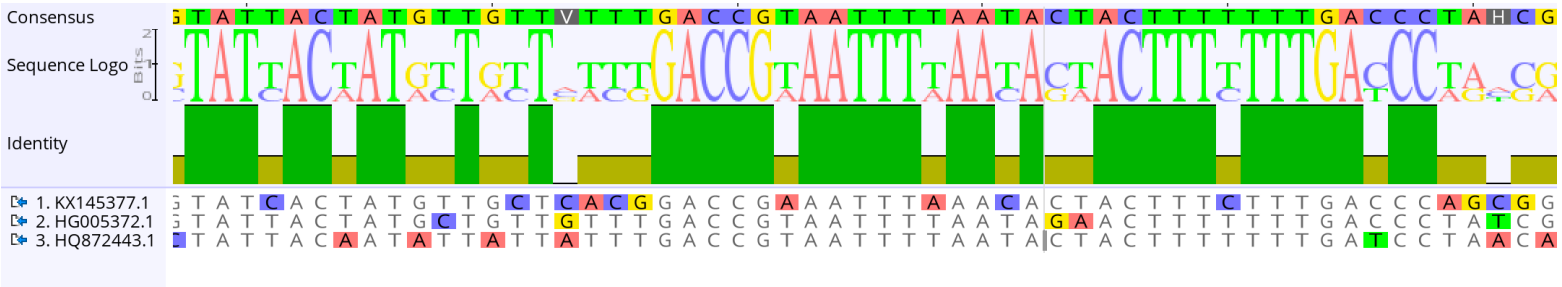
Ciona intestinalis

Arcuatula senhousia

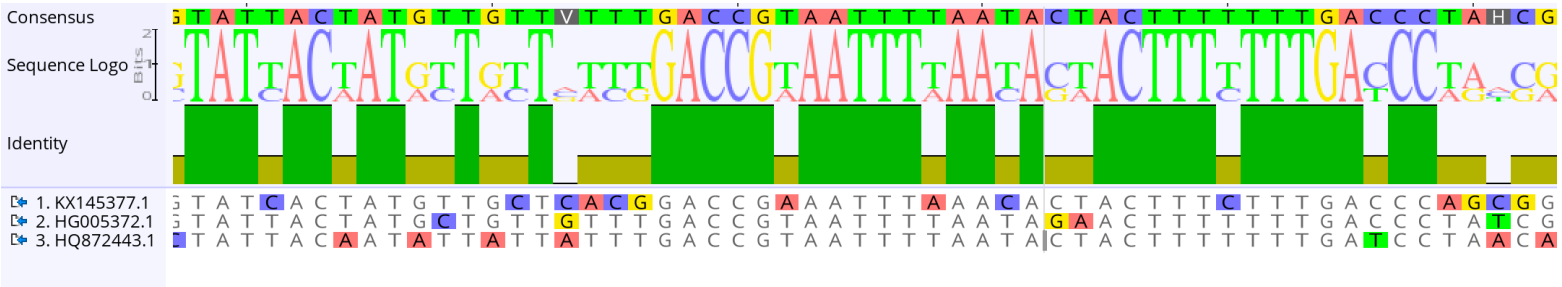
Oncorhynchus gorbuscha

Molecular barcode – a short (>600bp) DNA sequence, from a uniform locality in the genome used for species identification

DNA Barcoding



DNA Barcoding



Metazoan – COI

Herbert et al. 2003

Flowering Plants – rbcL

Hollingsworth et al. 2009

Fungi – ITS

Schoch et al. 2011

Eukaryotes – 18S

Amaral–Zettler et al. 2009

DNA Barcoding

Barcoding Projects

Barcode 500k 2010-2015

BioSCAN 2019-2025

http://www.boldsystems.org
http://ibol.org/projects/project-overview/

Genome Projects

25 High Quality Genomes

https://www.sanger.ac.uk/science/collaboration/25-genomes-25-years

Sequence the 66,000 UK species

Sequence all Eukaryotic diversity
Around $5 billion
https://www.earthbiogenome.org

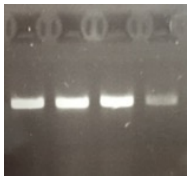
DNA Barcoding

Meta

PCR

Single species sample

A single, clean amplification product

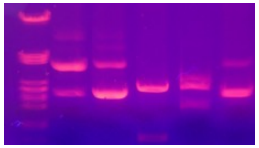


Sanger sequencing

1 seq
(300–1000bp)

Mixed species sample

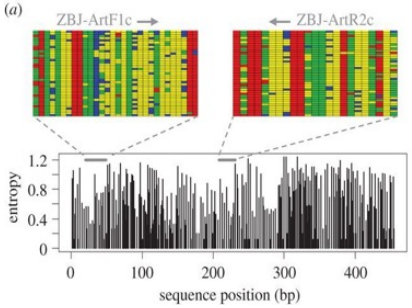
Multiple products from amplification

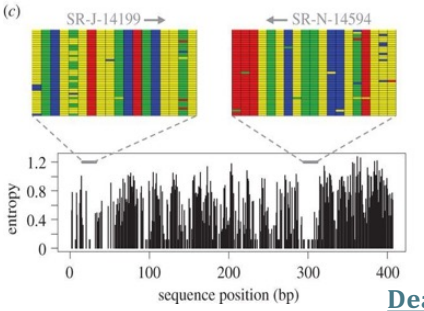


Next-generation Sequencing

10^6 – 10^8 seq
(75–300bp)

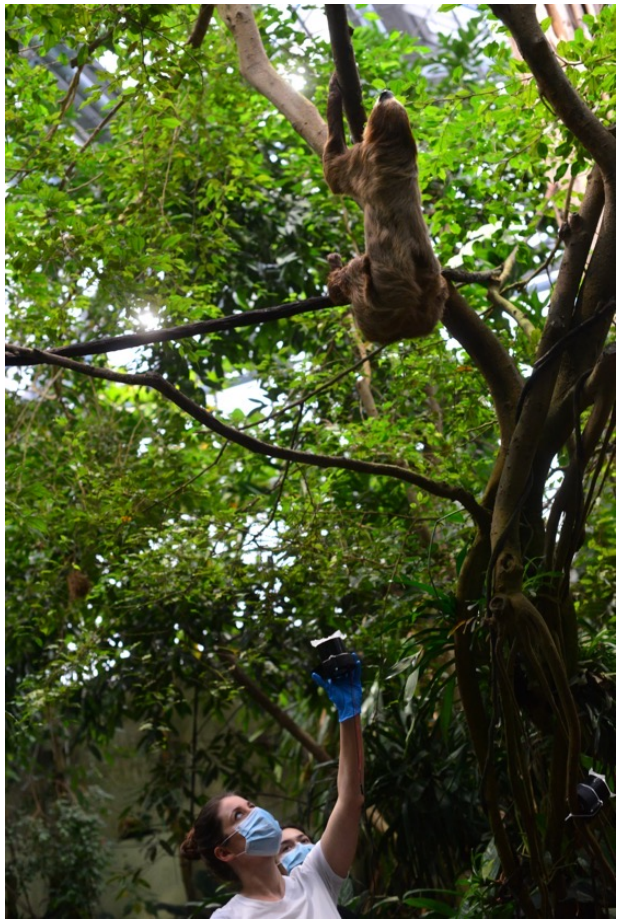
Variable region
Conserved region

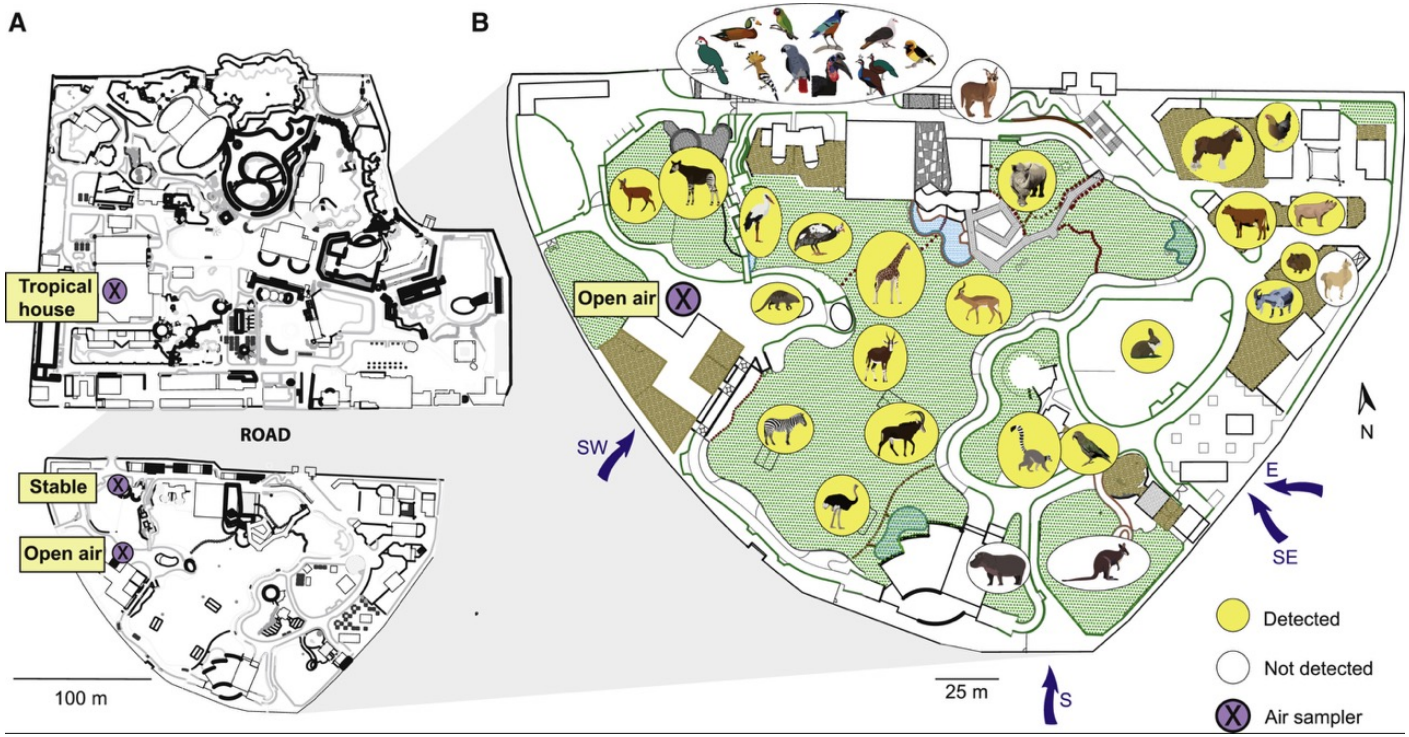




Deagle et al. 2014

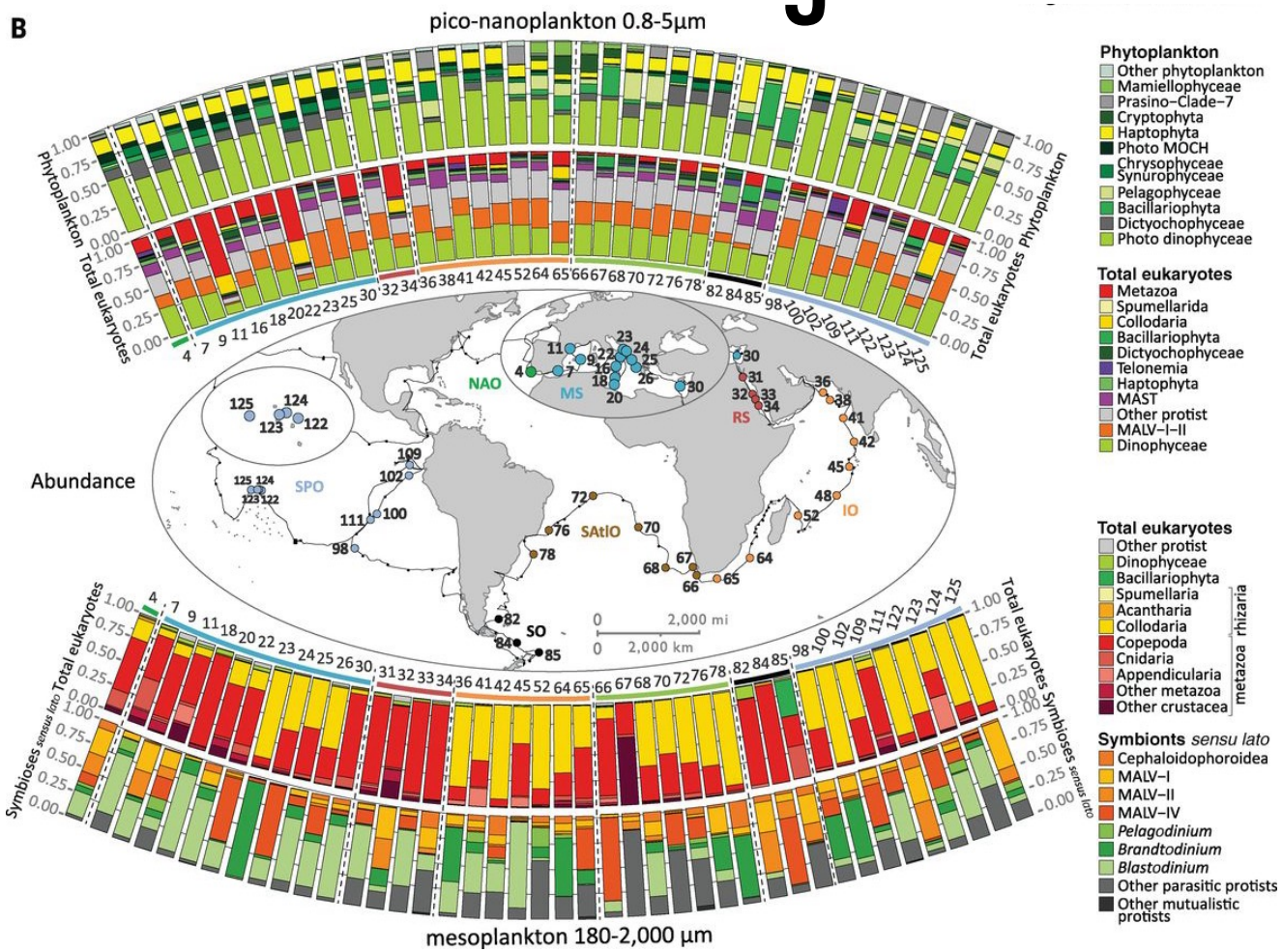
DNA Metabarcoding Studies





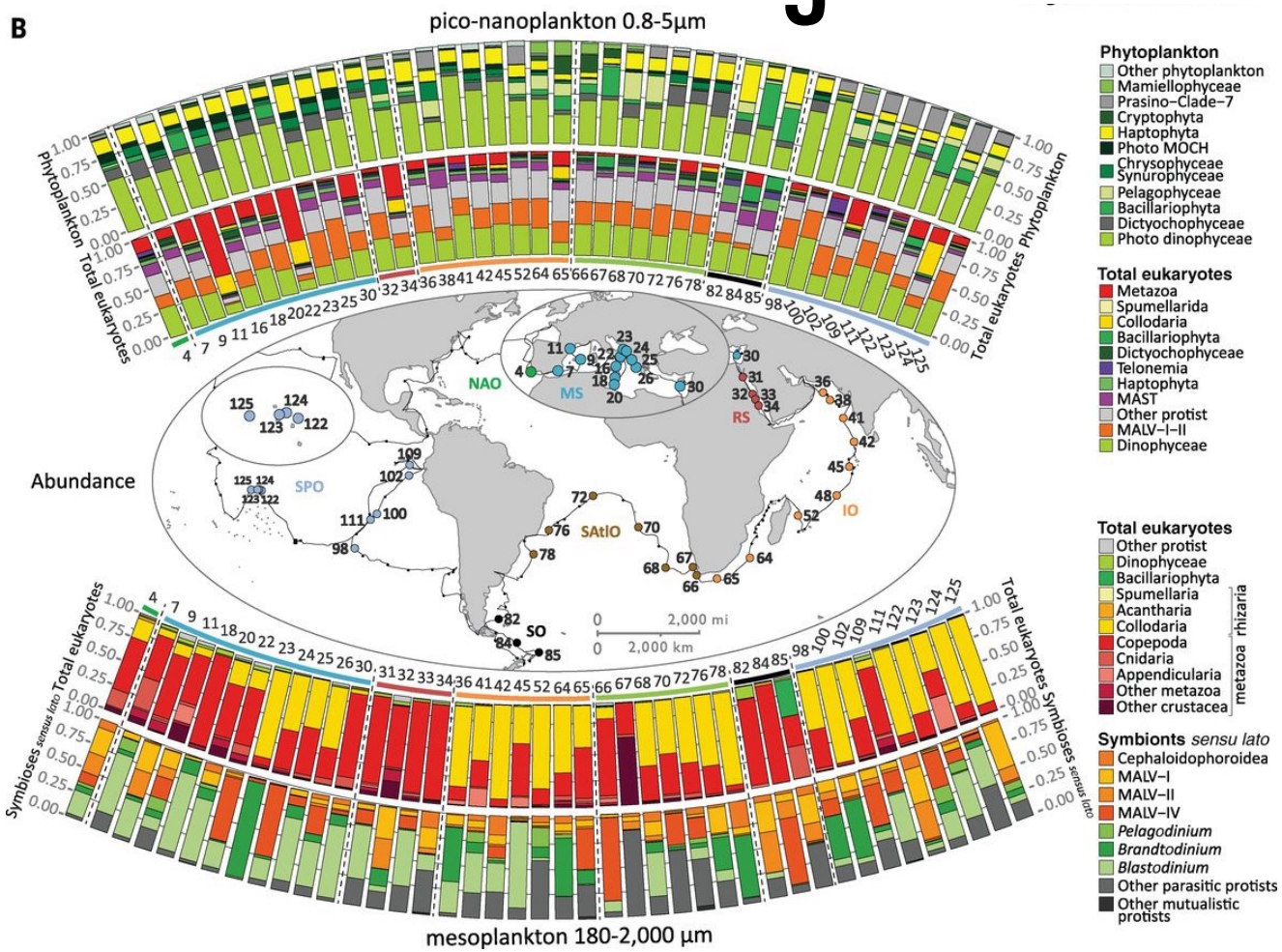
Lynggaard et al. 2022

DNA Metabarcoding Studies



De Vargas et al. 2015

DNA Metabarcoding Studies



De Vargas et al. 2015

Exercise 1 - Metabarcode detective

ACTATCTAGGAACATTGCACATGCCGGAAGGTCCGT
AGACTTTGCCATCTTTTCGTTACATTTAGCCGGAGTTA
GTTCAATTCTTGGGGCAGTAAATTTTATTAGGACTTTA
GGAAACCTTCGAGTATTTGGAATATTACTAGATCGCA
TACCTCTGTTTGCATGAGCTGTATTAATTACTGCTGTA
CTATTACTTCTTTCTTTACCTGTTCTAGCTGGGGCTATT
ACCATATTATTAACCGATCGTAATTTGAATACAACATTT
TATGATGTGGGGGGAGGAGGAGATCCTATTCTATATC
AACACCTATTC

Exercise 1 - Metabarcode detective

ACTATCTAGGAACATTGCACATGCCGGAAGGTCCGT
AGACTTTGCCATCTTTTCGTTACATTTAGCCGGAGTTA
GTTCAATTCTTGGGGCAGTAAATTTTATTAGGACTTTA
GGAAACCTTCGAGTATTTGGAATATTACTAGATCGCA
TACCTCTGTTTGCATGAGCTGTATTAATTACTGCTGTA
CTATTACTTCTTTCTTTACCTGTTCTAGCTGGGGCTATT
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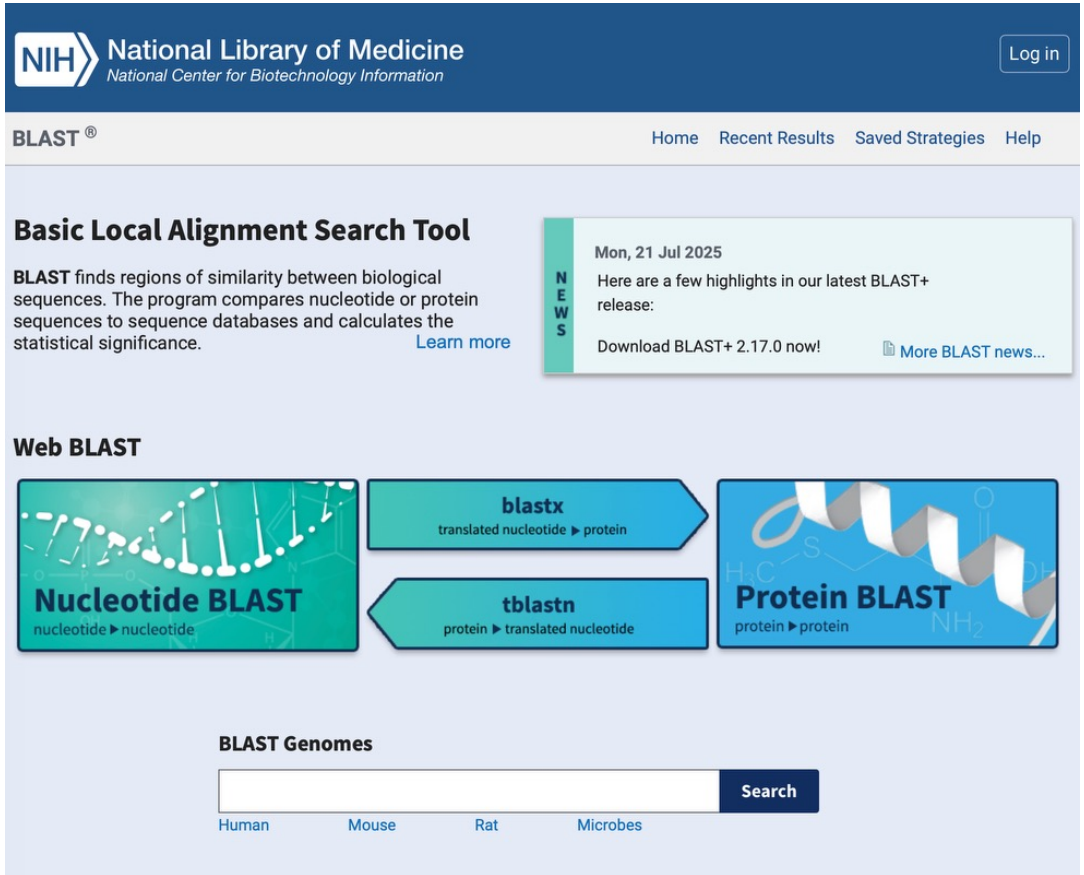

Exercise 1 - Metabarcode detective

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TACCTCTGTTTGCATGAGCTGTATTAATTACTGCTGTA
CTATTACTTCTTTCTTTACCTGTTCTAGCTGGGGCTATT
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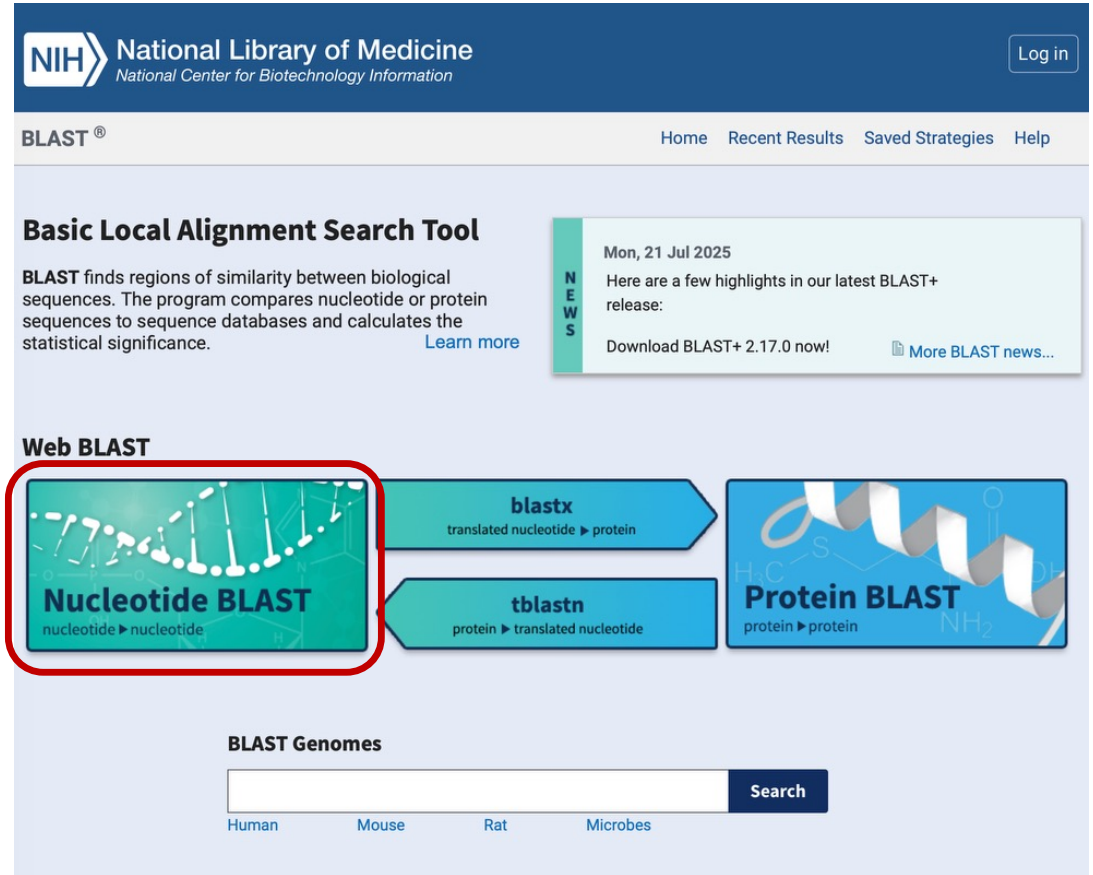

Exercise 1 - Metabarcode detective

ACTATCTAGGAACATTGCACATGCCGGAAGGTCCGT
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CTATTACTTCTTTCTTTACCTGTTCTAGCTGGGGCTATT
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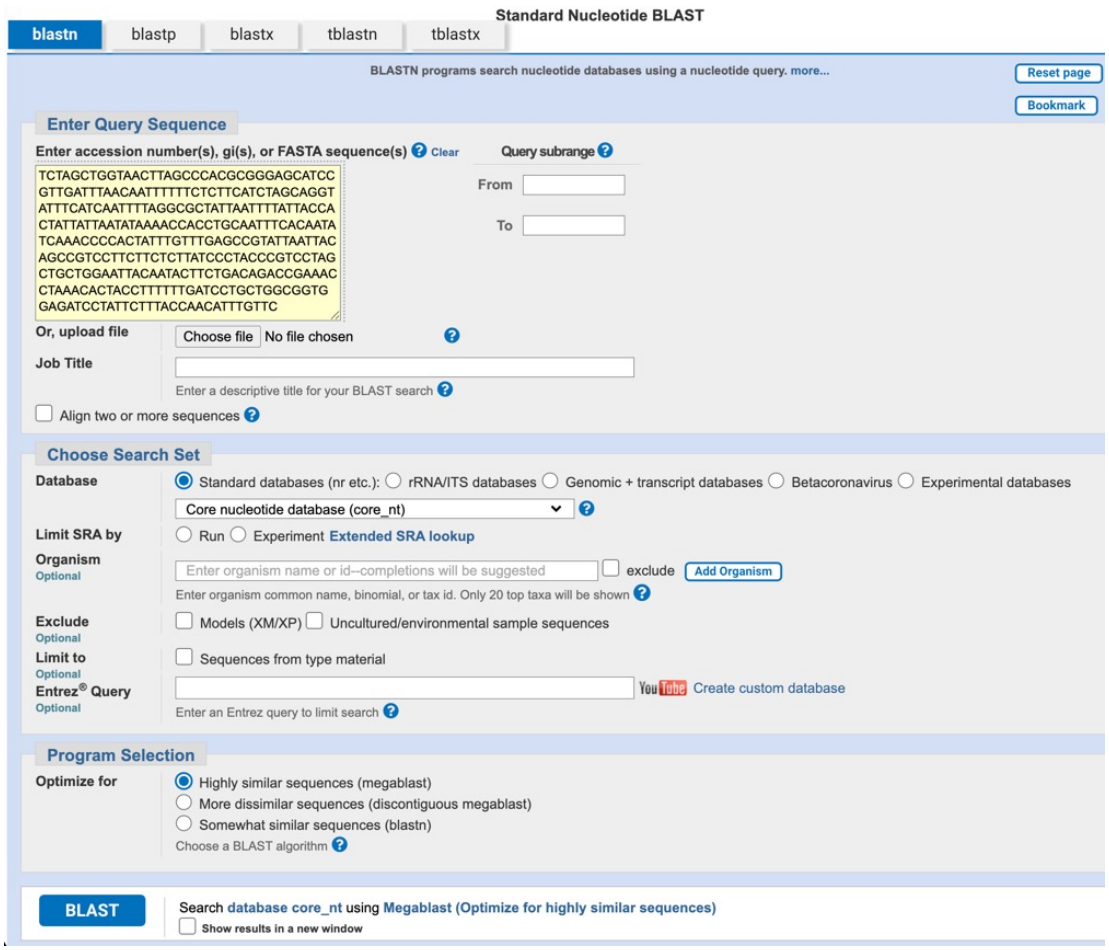

Exercise 1 - Metabarcode detective

ACTATCTAGGAACATTGCACATGCCGGAAGGTCCGT
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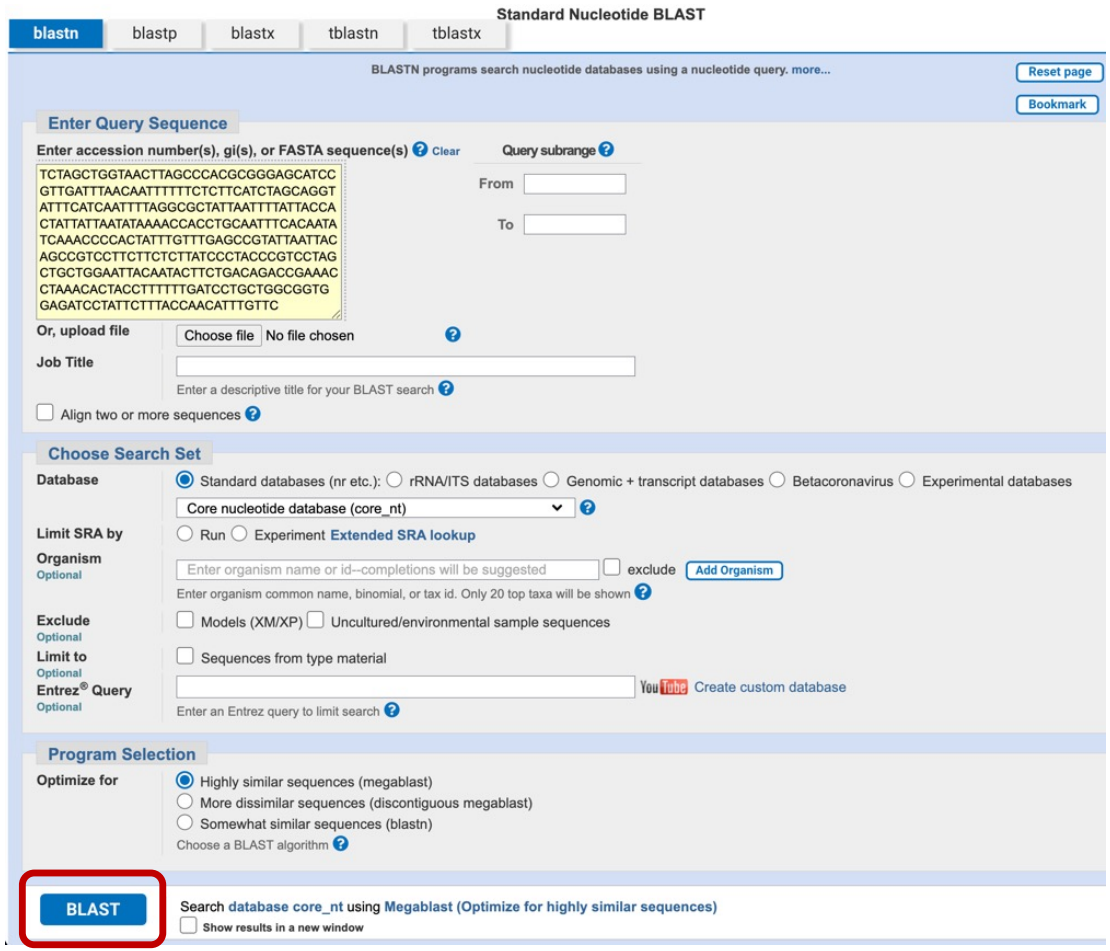

Exercise 1 - Metabarcode detective

Descriptions | Graphic Summary | Alignments | Taxonomy

Sequences producing significant alignments Download Select columns Show 100

select all *100 sequences selected* GenBank Graphics Distance tree of results MSA Viewer

Description	Scientific Name	Max Score	Total Score	Query Cover	E value	Per. Ident	Acc. Len	Accession
Paracalanus parvus voucher P0033 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	657	KP861426.1
Paracalanus parvus isolate IT2_9 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	644	MH818625.1
Paracalanus parvus voucher V082.NEA1a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	646	KF715875.1
Paracalanus parvus isolate P0244 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	644	MH818684.1
Paracalanus parvus isolate MT00828 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	577	577	100%	5e-160	100.00%	657	JX995216.1
Paracalanus parvus voucher PP0406.NEA cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus par...	577	577	100%	5e-160	100.00%	646	MN764491.1
Paracalanus parvus isolate Bul1_7 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	573	573	100%	6e-159	99.68%	644	MH818639.1
Paracalanus parvus isolate Bul3_2 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	573	573	100%	6e-159	99.68%	644	MH818656.1
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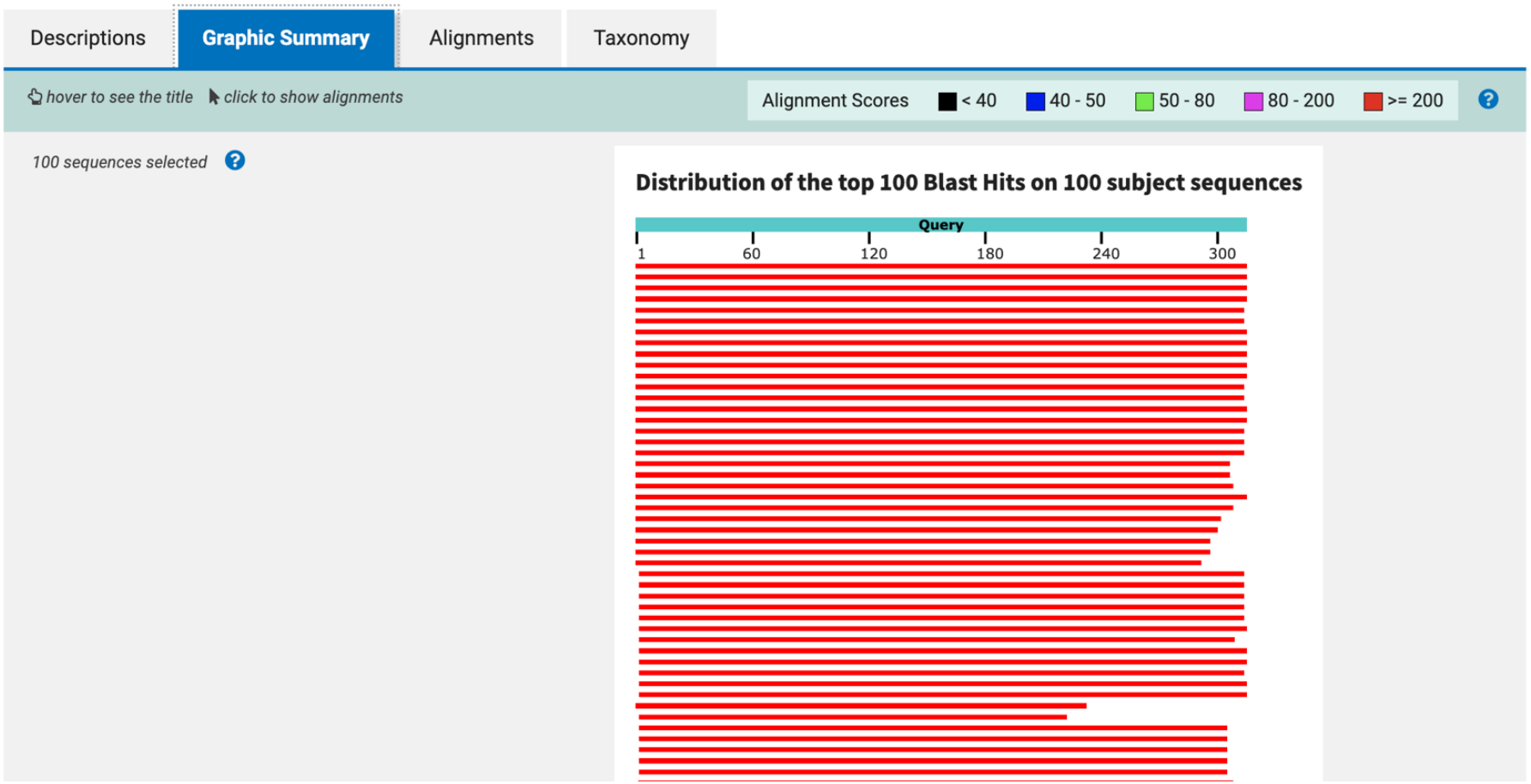
Exercise 1 - Metabarcode detective



Exercise 1 - Metabarcode detective

Descriptions | Graphic Summary | **Alignments** | Taxonomy

Alignment view: Pairwise | CDS feature | Restore defaults | Download

100 sequences selected

Download | GenBank | Graphics | ▼Next | ▲Previous | ◀Descriptions

Paracalanus parvus voucher P0033 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial

Sequence ID: KP861426.1 **Length:** 657 **Number of Matches:** 1

Range 1: 345 to 657 GenBank Graphics ▼Next Match ▲Previous Match

Score	Expect	Identities	Gaps	Strand
579 bits(313)	1e-160	313/313(100%)	0/313(0%)	Plus/Plus

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Sbjct  345  ACTATCTAGGAACATTGCACATGCCGGAAGGTCCGTAGACTTTGCCATCTTTTCGTTACA  404

Query  61   TTTAGCCGGAGTTAGTTCAATTCTTGGGGCAGTAAATTTTATTAGGACTTTAGGAAACCT  120
            ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
Sbjct  405  TTTAGCCGGAGTTAGTTCAATTCTTGGGGCAGTAAATTTTATTAGGACTTTAGGAAACCT  464

Query  121  TCGAGTATTTGGAATATTACTAGATCGCATACCTCTGTTTGCATGAGCTGTATTAATTAC  180
            ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
Sbjct  465  TCGAGTATTTGGAATATTACTAGATCGCATACCTCTGTTTGCATGAGCTGTATTAATTAC  524

Query  181  TGCTGTACTATTACTTCTTTCTTTACCTGTTCTAGCTGGGGCTATTACCATATTATTAAC  240
            ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
Sbjct  525  TGCTGTACTATTACTTCTTTCTTTACCTGTTCTAGCTGGGGCTATTACCATATTATTAAC  584

Query  241  CGATCGTAATTTGAATACAACATTTTATGATGTGGGGGGAGGAGGAGATCCTATTCTATA  300
            ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
Sbjct  585  CGATCGTAATTTGAATACAACATTTTATGATGTGGGGGGAGGAGGAGATCCTATTCTATA  644
```

Exercise 1 - Metabarcode detective

Descriptions | Graphic Summary | Alignments | **Taxonomy**

Reports | **Lineage** | **Organism** | **Taxonomy**

100 sequences selected

Organism	Blast Name	Score	Number of Hits	Description
Arthropoda	arthropods		191	
. Calanoida	crustaceans		188	
. . Paracalanidae	crustaceans		182	
. . . Paracalanus	crustaceans		164	
. . . . Paracalanus parvus	crustaceans	579	96	Paracalanus parvus hits
. . . . Paracalanus sp. F AC-2013	crustaceans	457	20	Paracalanus sp. F AC-2013 hits
. . . . Paracalanus indicus	crustaceans	324	41	Paracalanus indicus hits
. . . . Paracalanus tropicus	crustaceans	320	3	Paracalanus tropicus hits
. . . . Paracalanus aff. indicus AC-2013	crustaceans	318	1	Paracalanus aff. indicus AC-2013 hi
. . . . Paracalanus quasimodo	crustaceans	318	1	Paracalanus quasimodo hits
. . . . Paracalanus sp. PP0417.GOM	crustaceans	313	1	Paracalanus sp. PP0417.GOM hits
. . . . Paracalanus parvus environmental sample	crustaceans	296	1	Paracalanus parvus environmental s
. . . Acrocalanus monachus	crustaceans	309	2	Acrocalanus monachus hits
. . . Acrocalanus gibber	crustaceans	307	2	Acrocalanus gibber hits

Exercise 1 - Metabarcode detective

Descriptions | Graphic Summary | Alignments | Taxonomy

Sequences producing significant alignments Download Select columns Show 100

select all *100 sequences selected* GenBank Graphics Distance tree of results MSA Viewer

Description	Scientific Name	Max Score	Total Score	Query Cover	E value	Per. Ident	Acc. Len	Accession
Paracalanus parvus voucher P0033 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	657	KP861426.1
Paracalanus parvus isolate IT2_9 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	644	MH818625.1
Paracalanus parvus voucher V082.NEA1a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	646	KF715875.1
Paracalanus sp. F AC-2013 voucher V001.SEA2 cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus sp. ...	448	448	99%	4e-121	92.60%	647	KF715968.1
Paracalanus sp. F AC-2013 voucher V420.SEA5a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus sp. ...	448	448	99%	4e-121	92.60%	647	KF715973.1
Paracalanus parvus voucher PaPa1 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	446	446	99%	1e-120	92.60%	625	KC287780.1
Paracalanus sp. F AC-2013 voucher V001.SEA1 cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus sp. ...	442	442	99%	2e-119	92.28%	647	KF715967.1
Paracalanus sp. F AC-2013 voucher V420.SEA4a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus sp. ...	436	436	99%	8e-118	91.96%	647	KF715970.1
Paracalanus parvus isolate IT2_26 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	422	422	74%	2e-113	99.57%	562	MH818634.1
Paracalanus sp. F AC-2013 isolate IT2_29 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus sp. ...	329	329	70%	1e-85	93.61%	552	MH818800.1
Paracalanus indicus voucher V341.PI2a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus indi...	324	324	96%	7e-84	86.05%	647	KF715901.1
Paracalanus indicus voucher V071.PI1a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus indi...	324	324	96%	7e-84	86.05%	647	KF715892.1
Paracalanus parvus isolate Ppar_Ok cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial gene for...	Paracalanus par...	324	324	96%	7e-84	86.05%	656	AF474111.1
Paracalanus parvus voucher PaPa3 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	324	324	96%	7e-84	86.05%	646	KC287782.1

Exercise 1 - Metabarcode detective

Descriptions | Graphic Summary | Alignments | Taxonomy

Sequences producing significant alignments Download Select columns Show 100

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Paracalanus parvus isolate IT2_9 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	644	MH818625.1
Paracalanus parvus voucher V082.NEA1a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	646	KF715875.1
Paracalanus sp. F AC-2013 voucher V001.SEA2 cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus sp. ...	448	448	99%	4e-121	92.60%	647	KF715968.1
Paracalanus sp. F AC-2013 voucher V420.SEA5a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus sp. ...	448	448	99%	4e-121	92.60%	647	KF715973.1
Paracalanus parvus voucher PaPa1 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	446	446	99%	1e-120	92.60%	625	KC287780.1
Paracalanus sp. F AC-2013 voucher V001.SEA1 cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus sp. ...	442	442	99%	2e-119	92.28%	647	KF715967.1
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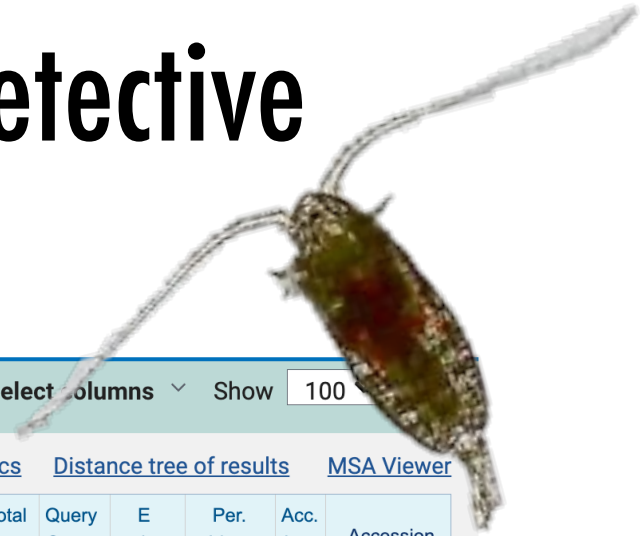
Paracalanus sp. F AC-2013 isolate IT2_29 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus sp. ...	329	329	70%	1e-85	93.61%	552	MH818800.1
Paracalanus indicus voucher V341.PI2a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus indi...	324	324	96%	7e-84	86.05%	647	KF715901.1
Paracalanus indicus voucher V071.PI1a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus indi...	324	324	96%	7e-84	86.05%	647	KF715892.1
Paracalanus parvus isolate Ppar_Ok cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial gene for...	Paracalanus par...	324	324	96%	7e-84	86.05%	656	AF474111.1
Paracalanus parvus voucher PaPa3 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	324	324	96%	7e-84	86.05%	646	KC287782.1

Exercise 1 - Metabarcode detective



Descriptions | Graphic Summary | Alignments | Taxonomy

Sequences producing significant alignments Download Select columns Show 100

select all *100 sequences selected* GenBank Graphics Distance tree of results MSA Viewer

Description	Scientific Name	Max Score	Total Score	Query Cover	E value	Per. Ident	Acc. Len	Accession
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Paracalanus parvus isolate IT2_9 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	644	MH818625.1
Paracalanus parvus voucher V082.NEA1a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	646	KF715875.1
Paracalanus parvus isolate P0244 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	579	579	100%	1e-160	100.00%	644	MH818684.1
Paracalanus parvus isolate MT00828 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	577	577	100%	5e-160	100.00%	657	JX995216.1
Paracalanus parvus voucher PP0406.NEA cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus par...	577	577	100%	5e-160	100.00%	646	MN764491.1
Paracalanus parvus isolate Bul1_7 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	573	573	100%	6e-159	99.68%	644	MH818639.1
Paracalanus parvus isolate Bul3_2 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	573	573	100%	6e-159	99.68%	644	MH818656.1
Paracalanus parvus isolate Ukr1_4 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	573	573	100%	6e-159	99.68%	644	MH818669.1
Paracalanus parvus isolate IT7_29 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	573	573	100%	6e-159	99.68%	644	MH818637.1
Paracalanus parvus isolate IT2_14 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	573	573	100%	6e-159	99.68%	644	MH818626.1
Paracalanus parvus isolate MT03859 cytochrome c oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	571	571	100%	2e-158	99.68%	657	JX995220.1
Paracalanus parvus voucher PP0400.NEA cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus par...	571	571	100%	2e-158	99.68%	646	MN764495.1
Paracalanus parvus voucher V106.NEA2a cytochrome oxidase subunit I gene, partial cds; mitochondrial	Paracalanus par...	568	568	100%	3e-157	99.36%	646	KF715881.1
Paracalanus parvus isolate Bul3_1 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	Paracalanus par...	568	568	100%	3e-157	99.36%	644	MH818655.1

Exercise 1 - Metabarcode detective

TCTAGCTGGTAACTTAGCCCACGCGGGAG
CATCCGTTGATTTAACAATTTTTTCTCTTCATC
TAGCAGGTATTTCATCAATTTTAGGCGCTATT
AATTTTATTACCACTATTATTAATATAAAACCA
CCTGCAATTTCACAATATCAAACCCCACTAT
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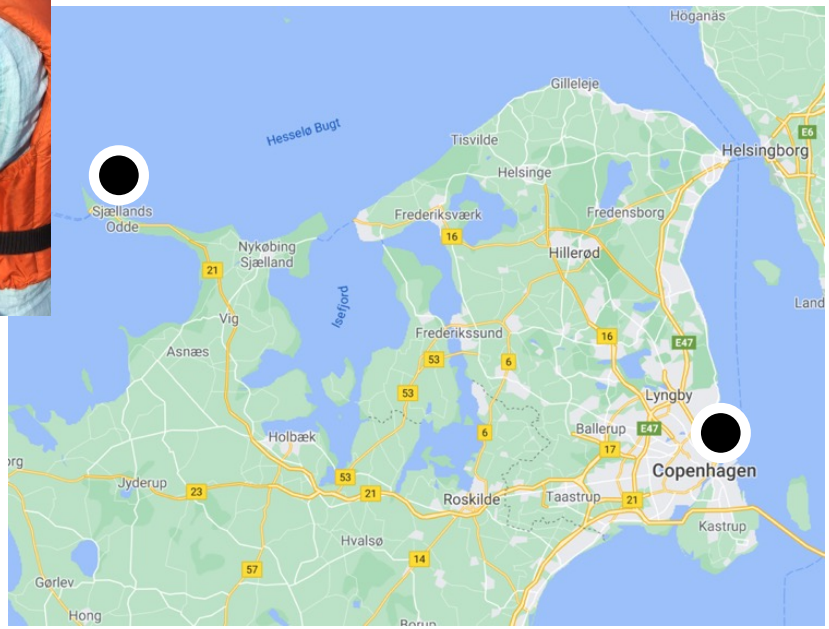

Exercise 1 - Metabarcode detective

ACTATCTAGGAACATTGCACATGCCGG
AAGGTCCGTAGACTTTGCCATCTTTTCG
TTACATTTAGCCGGAGTTAGTTCAATTCT
TGGGGCAGTAAATTTTATTAGGACTTTAG
GAAACCTTCGAGTATTTGGAATATTACTA
GATCGCATACCTCTGTTTGCATGAGCTG
TATTAATTACTGCTGTACTATTACTTCTTT
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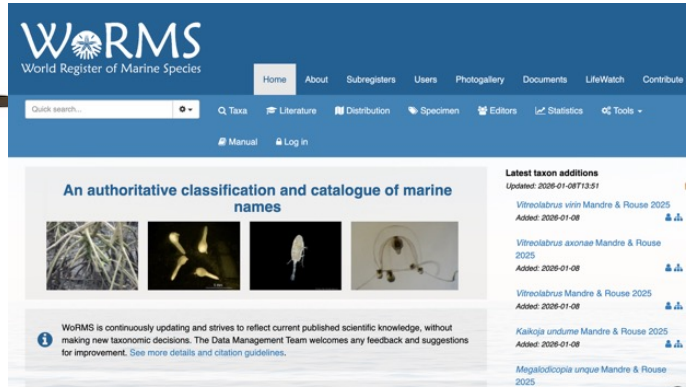
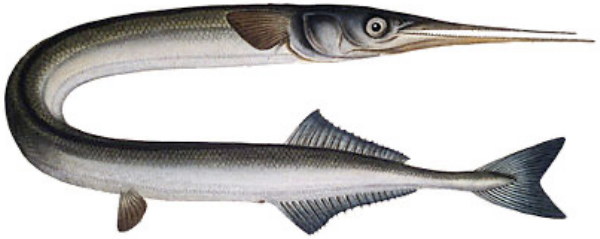


Results

Seq 1

Seq 1 Belone belone

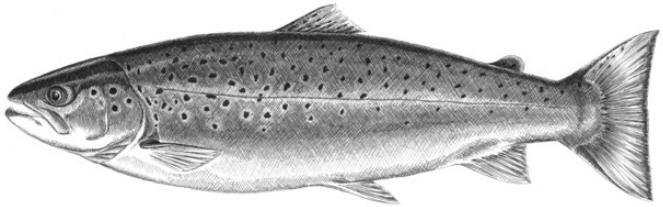
Hornfisk/Garfish



Seq 2

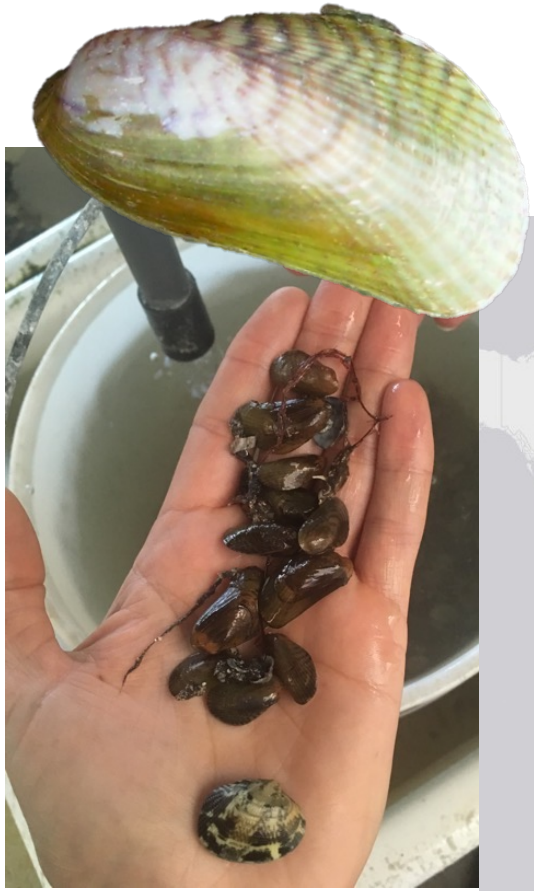
Seq 2 Salmo trutta

Sea trout/Brown trout



Seq 3

Seq 3 Arcuatula senhousia



Seq 3 Arcuatula senhousia

WoRMS
World Register of Marine Species

Home | About | Subregisters | Users | Photogallery | Documents | LifeWatch | Contribute

Quick search... | Taxa | Literature | Distribution | Specimen | Editors | Statistics | Tools | Manual | Log in

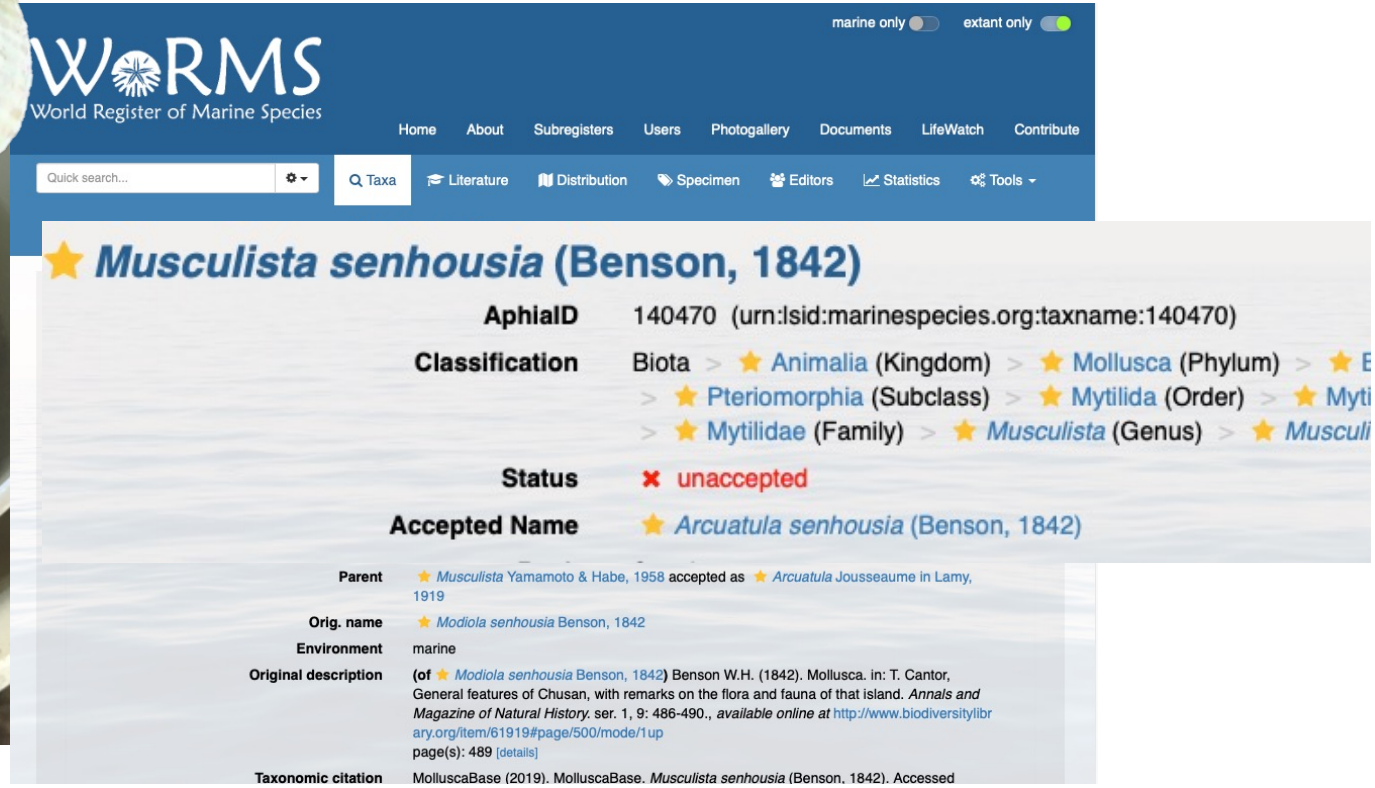
WoRMS name details

Musculista senhousia (Benson, 1842)

AphiaID	140470 (urn:lsid:marinespecies.org:taxname:140470)
Classification	Biota > Animalia (Kingdom) > Mollusca (Phylum) > Bivalvia (Class) > Pteriomorphia (Subclass) > Mytilida (Order) > Mytiloidea (Superfamily) > Mytilidae (Family) > *Musculista* (Genus) > *Musculista senhousia* (Species)
Status	unaccepted
Accepted Name	*Arcuatula senhousia* (Benson, 1842)
Rank	Species
Parent	*Musculista* Yamamoto & Habe, 1958 accepted as *Arcuatula* Jousseaume in Lamy, 1919
Orig. name	*Modiola senhousia* Benson, 1842
Environment	marine
Original description	(of *Modiola senhousia* Benson, 1842) Benson W.H. (1842). Mollusca. in: T. Cantor, General features of Chusan, with remarks on the flora and fauna of that island. *Annals and Magazine of Natural History.* ser. 1, 9: 486-490., *available online at* http://www.biodiversitylibrary.org/item/61919#page/500/mode/1up page(s): 489 [details]
Taxonomic citation	MolluscaBase (2019). MolluscaBase. *Musculista senhousia* (Benson, 1842). Accessed

Seq 3 Arcuatula senhousia

–Taxonomy is a dynamic discipline
–Use the taxonomy matching tool in WoRMS (if marine) or similar databases



Seq 4

Seq 4 Caenus pusilla

–eDNA contains many non–target organisms

–Think critically about transport

Seq 4 Caenus pusilla

–eDNA contains many non–target organisms

–Think critically about transport

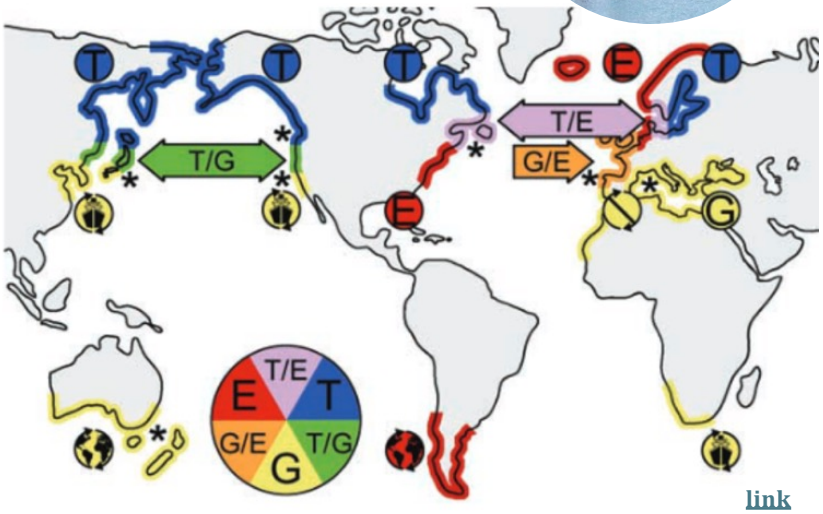
–Use taxonomy to subset data by species or groups of interest.

Seq 5

Seq 5 Mytilus spp.

Mytilus edulis
Mytilus trossulus
Mytilus galloprovincialis

Seq 5 Mytilus spp.



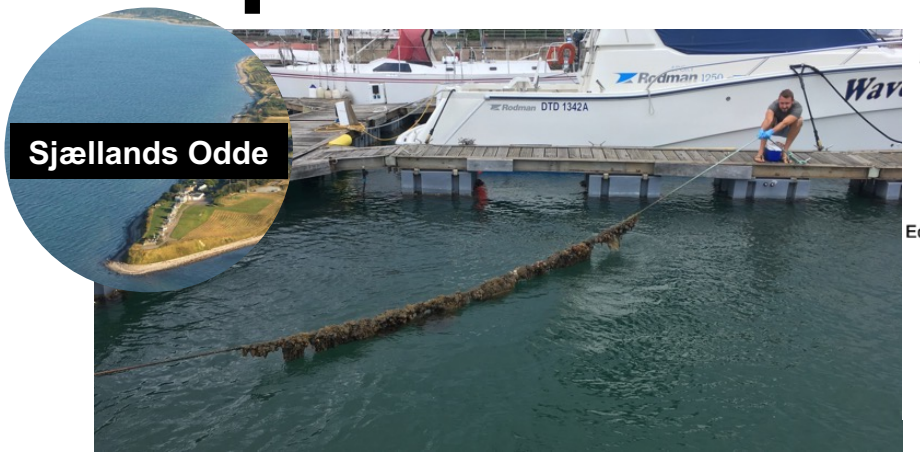
link

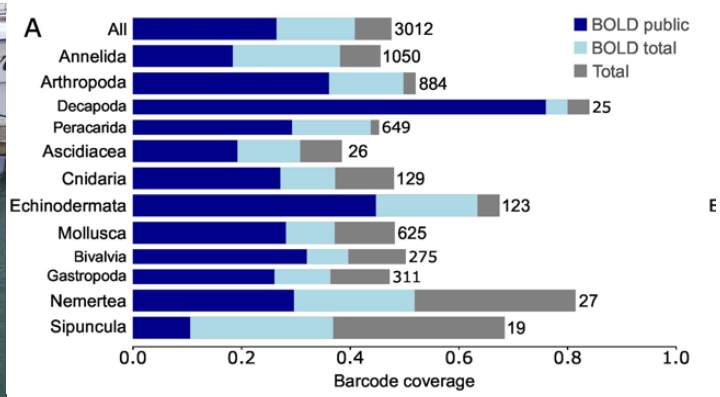
Mytilus edulis
Mytilus trossulus
Mytilus galloprovincialis

–eDNA often blind to hybrids and species complexes

Seq 6

Seq 6 Unknown botryllid ascidian





Weigand et al. 2019

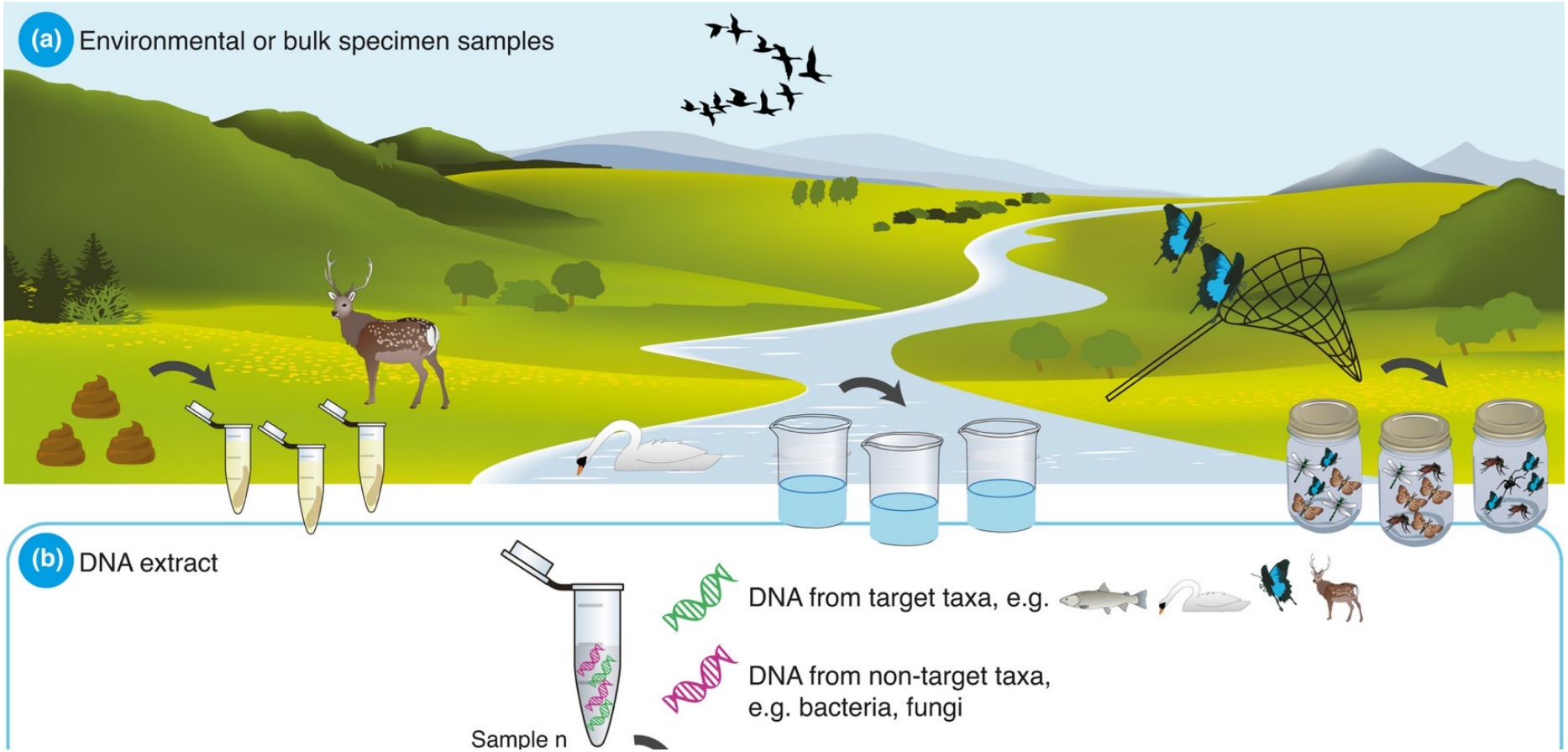
–Less than 50% coverage of barcodes in Europe!

–38% in Ascidiacea ☹

Results

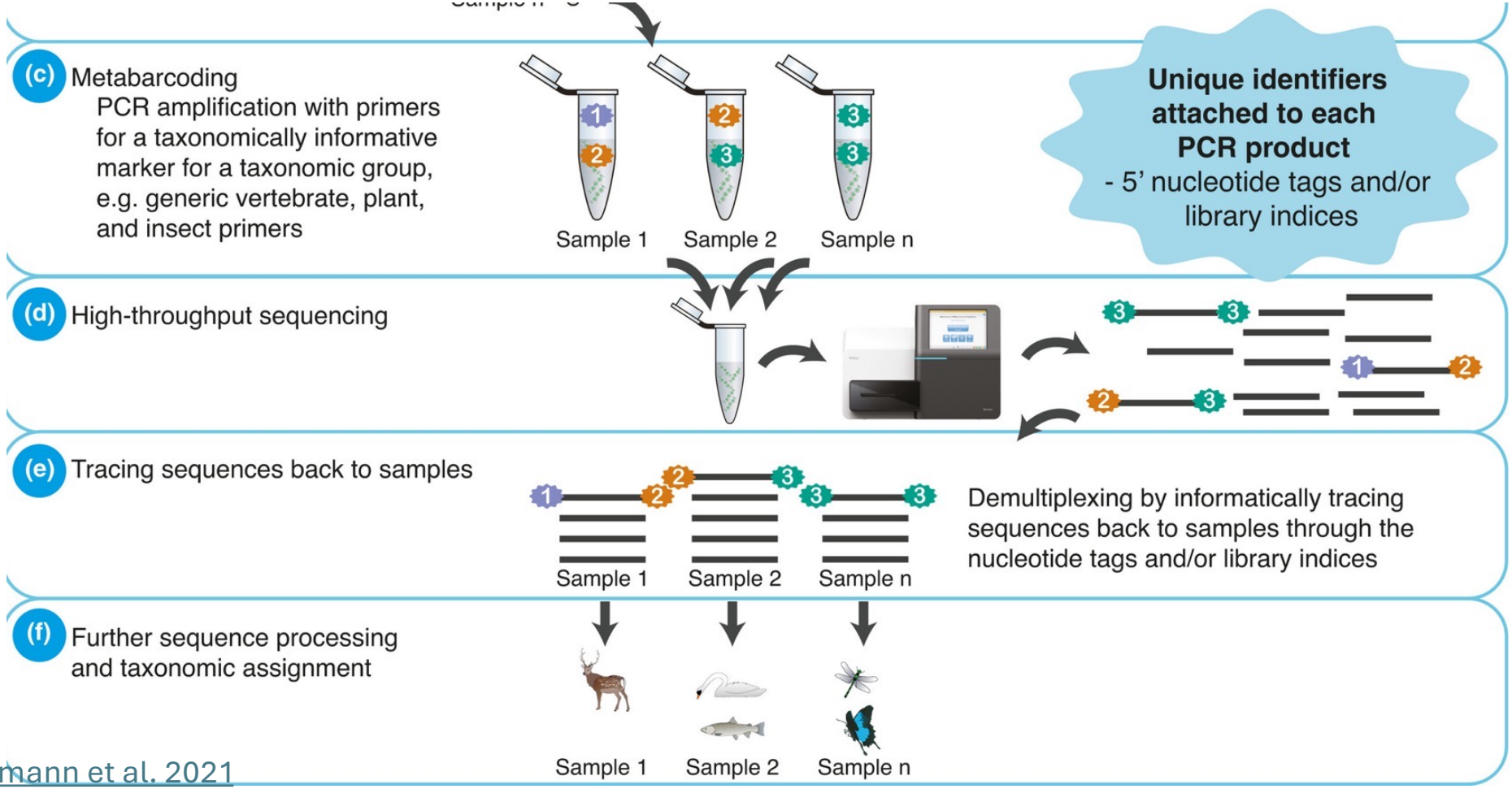
How do we do metabarcoding?

How do we do metabarcoding?



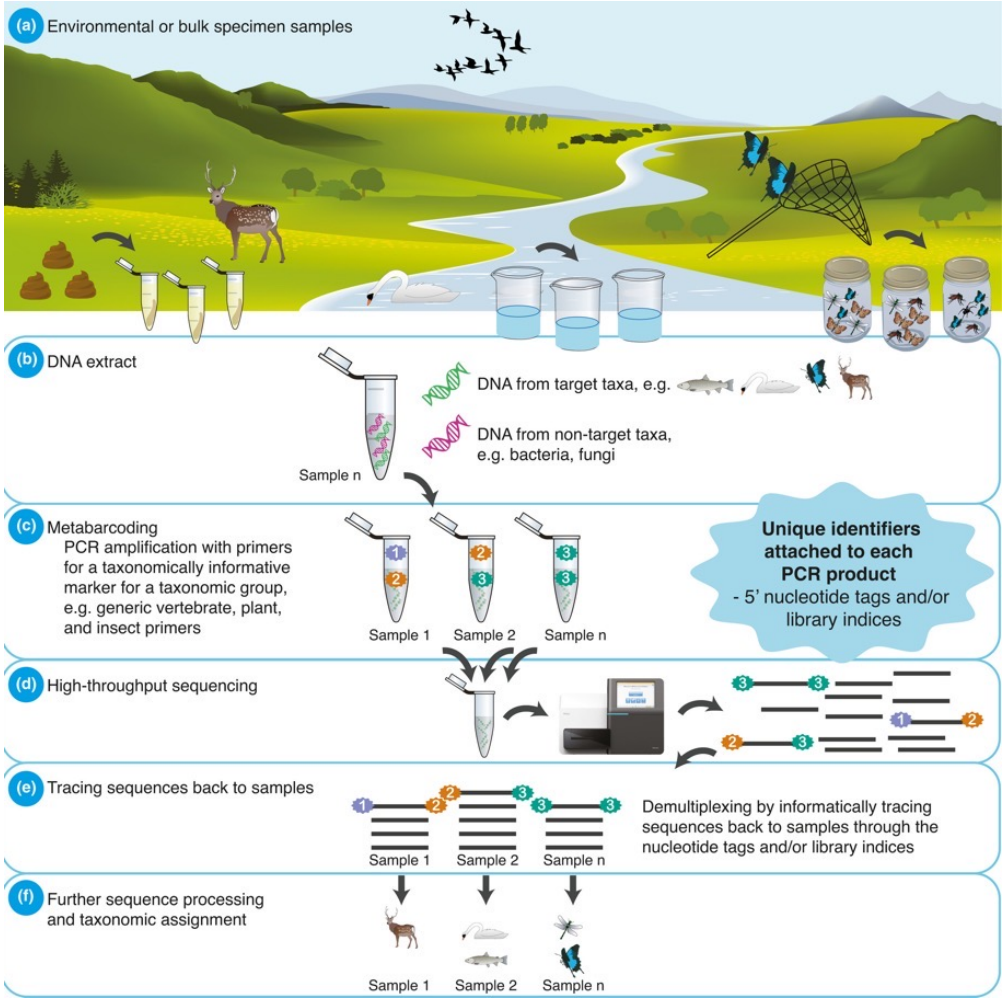
Bohmann et al. 2021

How do we do metabarcoding?



Bohmann et al. 2021

How do we do metabarcoding?



Bohmann et al. 2021

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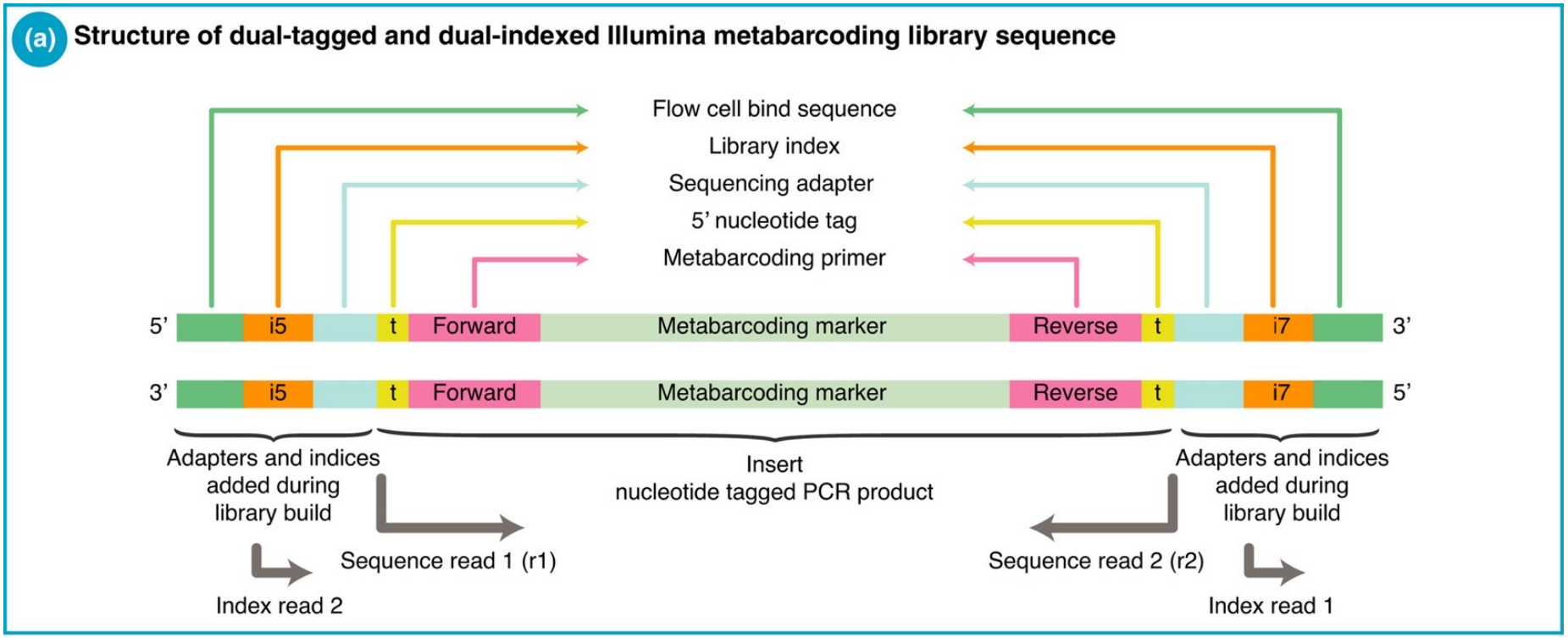
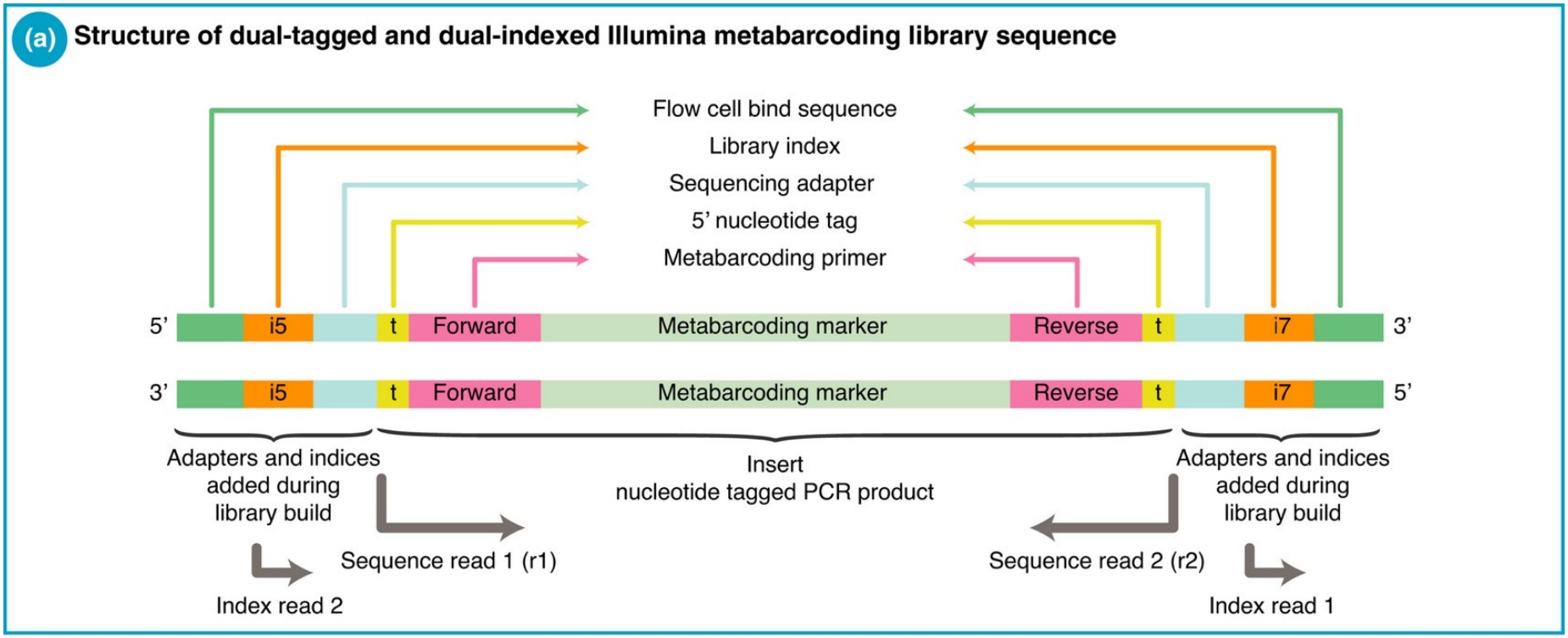


Bohmann et al. 2021

How do we do metabarcoding?



Bohmann et al. 2021

How do we do metabarcoding?

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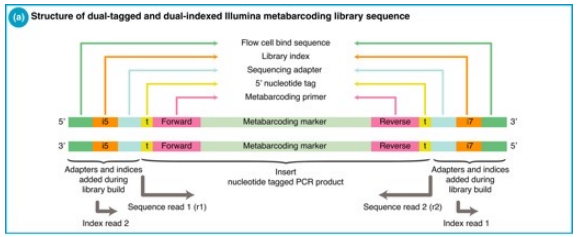

Bohmann et al. 2021

How do we do metabarcoding?

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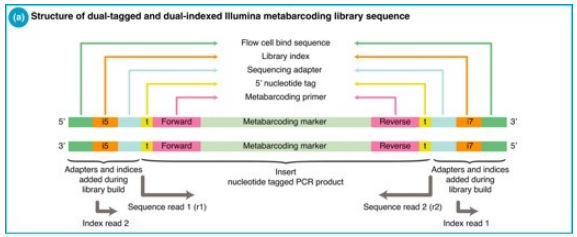

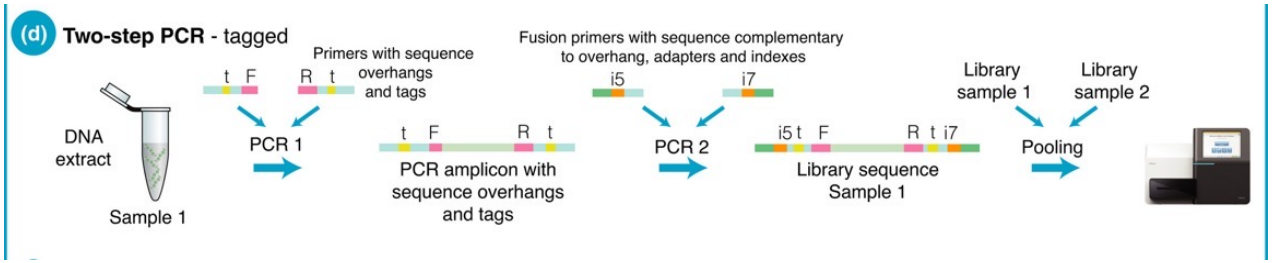

Bohmann et al. 2021

How do we do metabarcoding?

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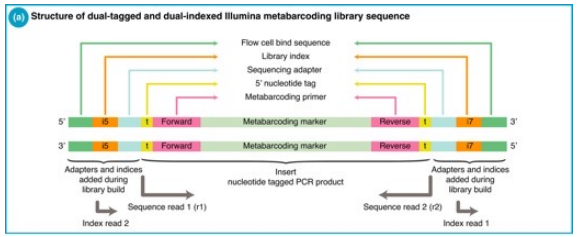
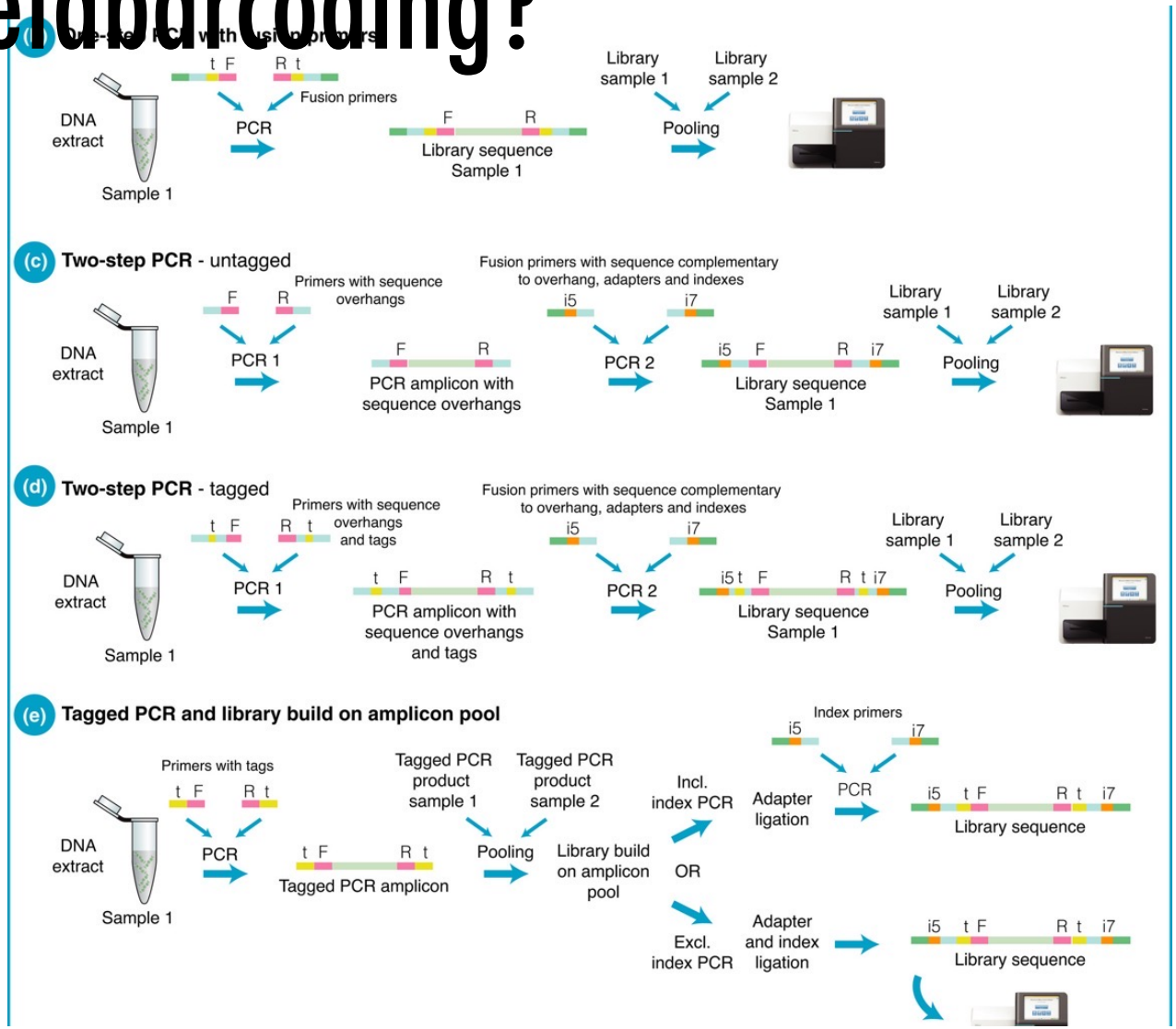
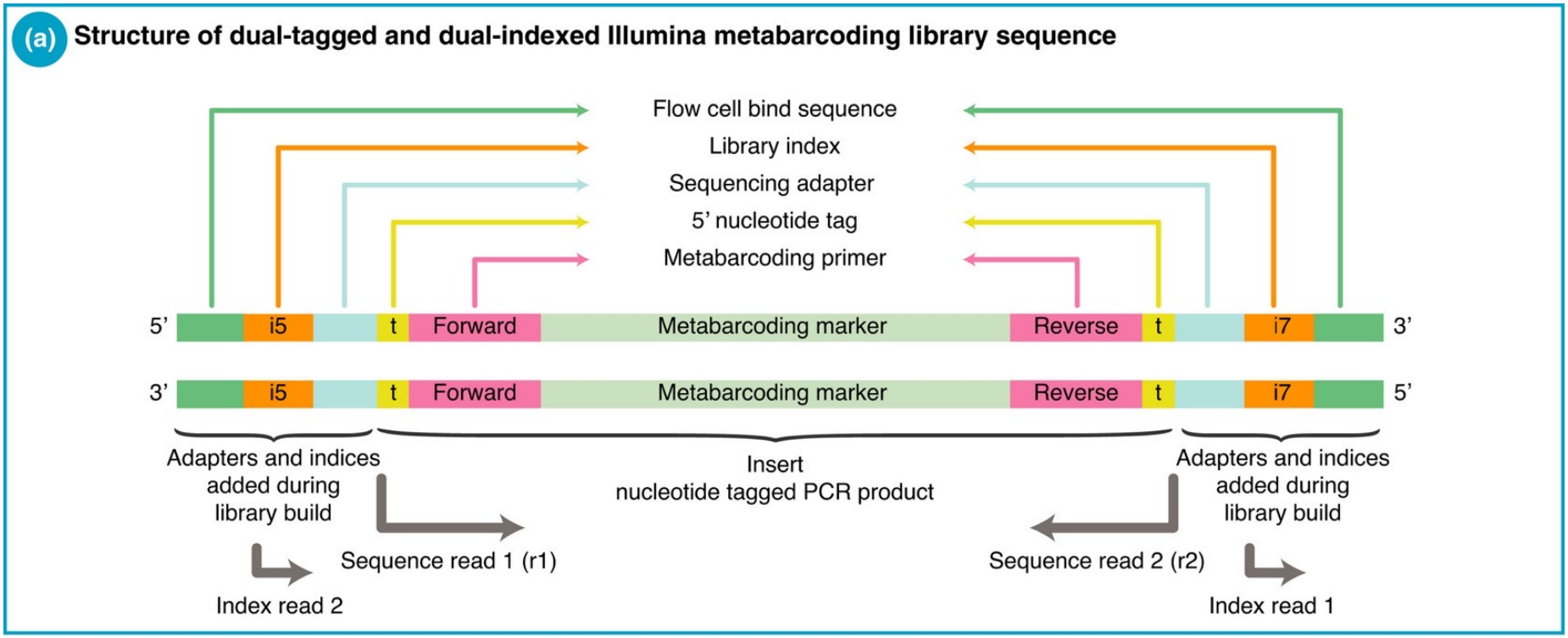




Bohmann et al. 2021

How do we do metabarcoding?

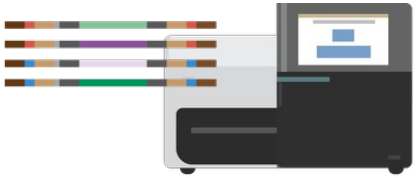


Bohmann et al. 2021

eDNA Metabarcoding Bioinformatics

AIM: To cluster or filter sequences into biologically relevant groupings assigned to samples while minimizing the effect of biases

Sequencing Library



Raw Reads

Quality Control & dereplication

Merging pairs

Sequence by Sample Table

	A	B	C	D
1		Sample_1	Sample_2	Sample_N
2	OTU_1	0	20	0
3	OTU_2	500	20	0
4	OTU_N	100	20	0

Metabarcoding Pipelines

Simple

Complicated

Metabarcoding Pipelines

Simple

Constrained

Complicated

Adaptable

Metabarcoding Pipelines

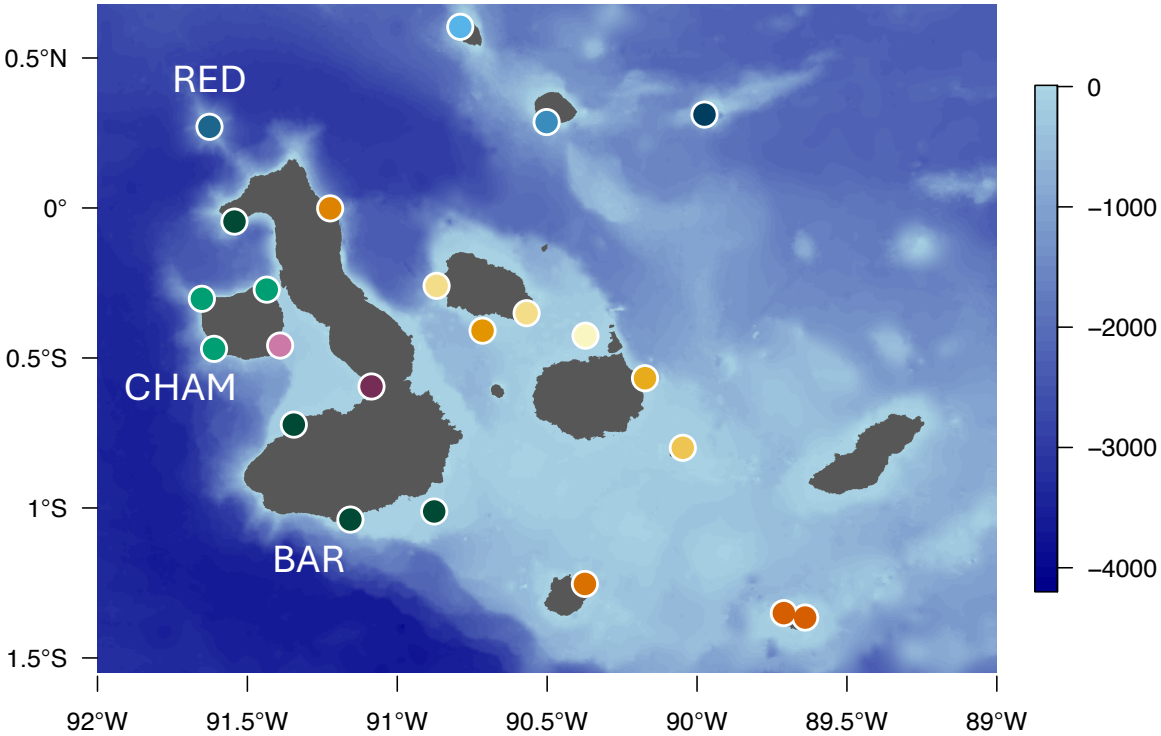
Simple

Constrained

Complicated

Adaptable

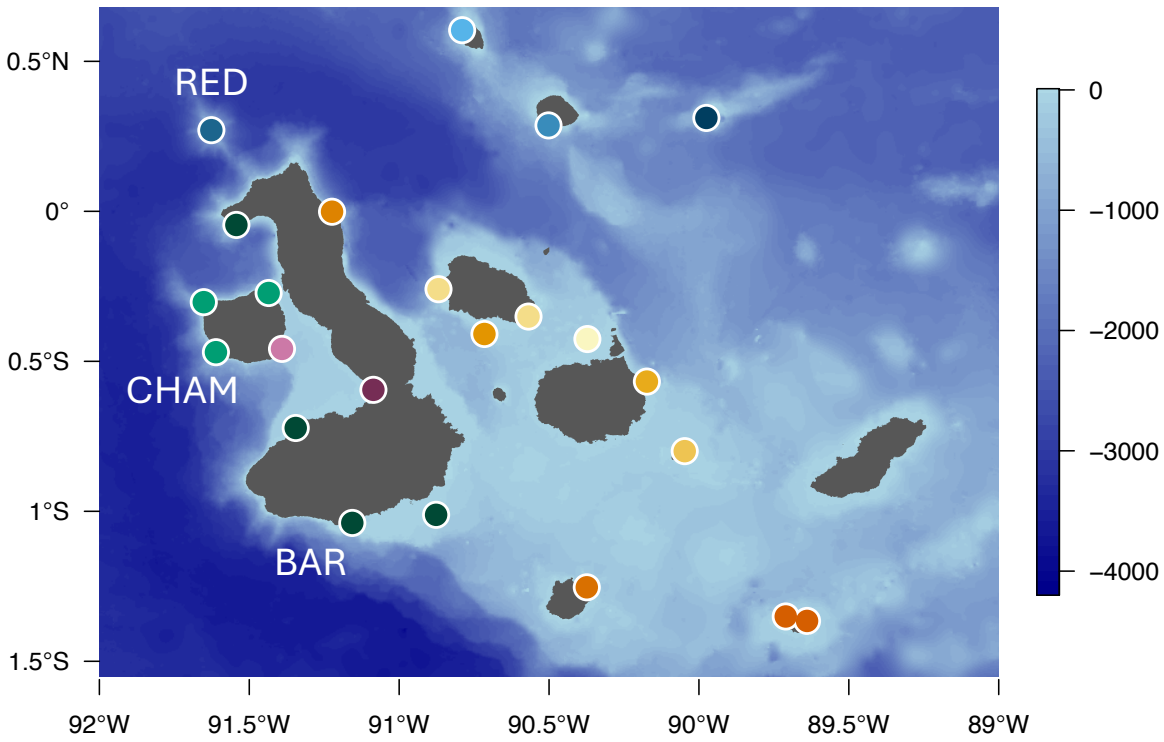
Metabarcoding Dataset



RED - Roca Redonda
CHAM - Cabo Hammond
BAR - Barahona Beach

C1 – Control 1 (extraction control)
C2 – Control 2 (PCR control)

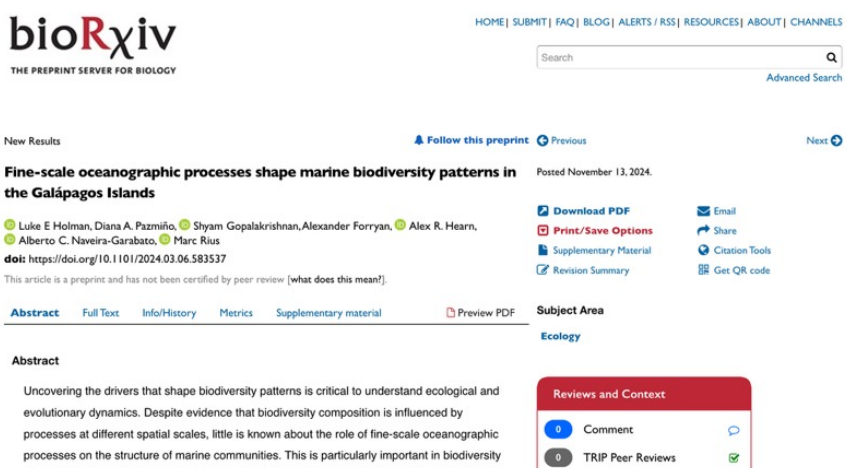
Metabarcoding Dataset



What DNA region is this data from?

What is the most common taxa?

Do we see differences between our samples?



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Fine-scale oceanographic processes shape marine biodiversity patterns in the Galápagos Islands

Posted November 13, 2024.

Luke E Holman, Diana A. Pazmiño, Shyam Gopalakrishnan, Alexander Forryan, Alex R. Hearn, Alberto C. Naveira-Garabato, Marc Rius

doi: https://doi.org/10.1101/2024.03.06.583537

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Subject Area
Ecology

Abstract

Uncovering the drivers that shape biodiversity patterns is critical to understand ecological and evolutionary dynamics. Despite evidence that biodiversity composition is influenced by processes at different spatial scales, little is known about the role of fine-scale oceanographic processes on the structure of marine communities. This is particularly important in biodiversity

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