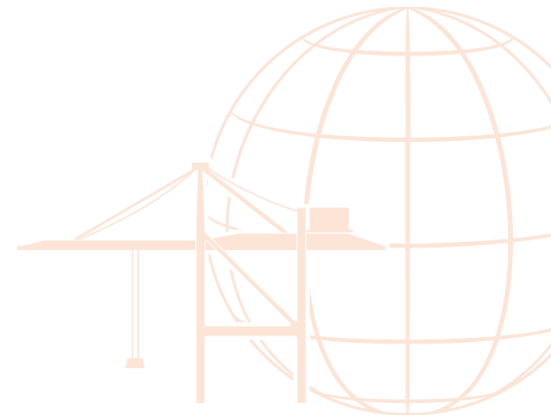


# Monitoring of estuarine fish compositions using an environmental DNA metabarcoding

Shinya Hosokawa

Head of Marine Environmental Information Group,  
Port and Airport Research Institute (PARI), Japan



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## Topics



- What's environmental DNA (eDNA)?
- Advantages of the eDNA technology
- Challenges in using eDNA technology in estuaries and coasts
- What will eDNA technologies contribute?
- Collaborations with open data platform
- Conclusions

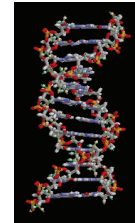
## What's environmental DNA (eDNA)?



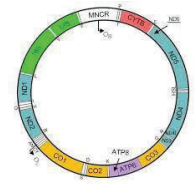
- Environmental DNA (eDNA): DNA released in the environments
- eDNA technology: Technology for detecting trace amounts of eDNA

DNA released from each individuals in the environments

<https://ednasociety.org>



Nuclear DNA  
(Wikipedia)



Mitochondrial DNA  
(Nagoya City University)



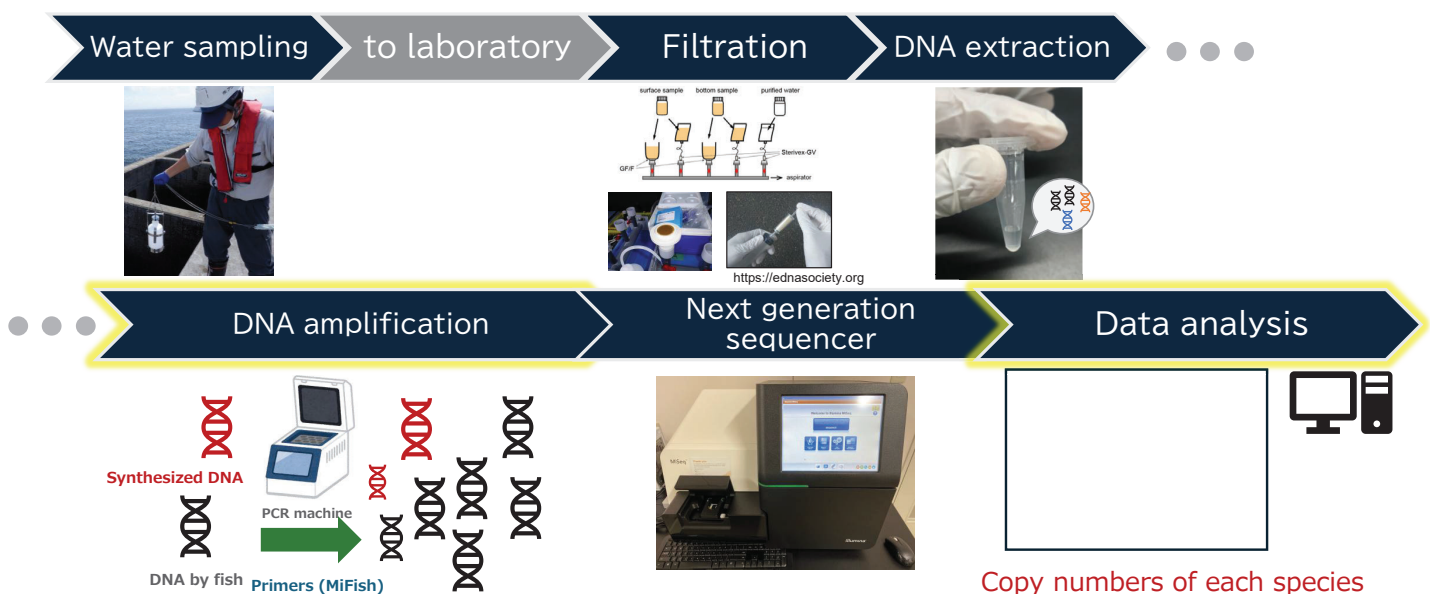
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## What's environmental DNA (eDNA)?



- A process for detecting fish species using eDNA technology



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# What's environmental DNA (eDNA)?



- Detection performance is dependent on “primers.”
- MiFish is a powerful primer set for detecting fish composition.

Mitochondrial DNA

MiFish-U: Universal primer  
MiFish-Ev2: Primer for elasmobranchs  
MiFish-U2: Primer for Pseudoblenius species

Universal region for fish

Hypervariable region (ca 170 bp)

Universal region for fish

first-round tailed PCR to amplify regions of interest

overhang adapter sequence used in second-round PCR and paired-end sequencing on MiSeq

region of interest-specific primer

forward

reverse

PCR 35 cycles

Miya et al. (2015), Royal Society Open Science

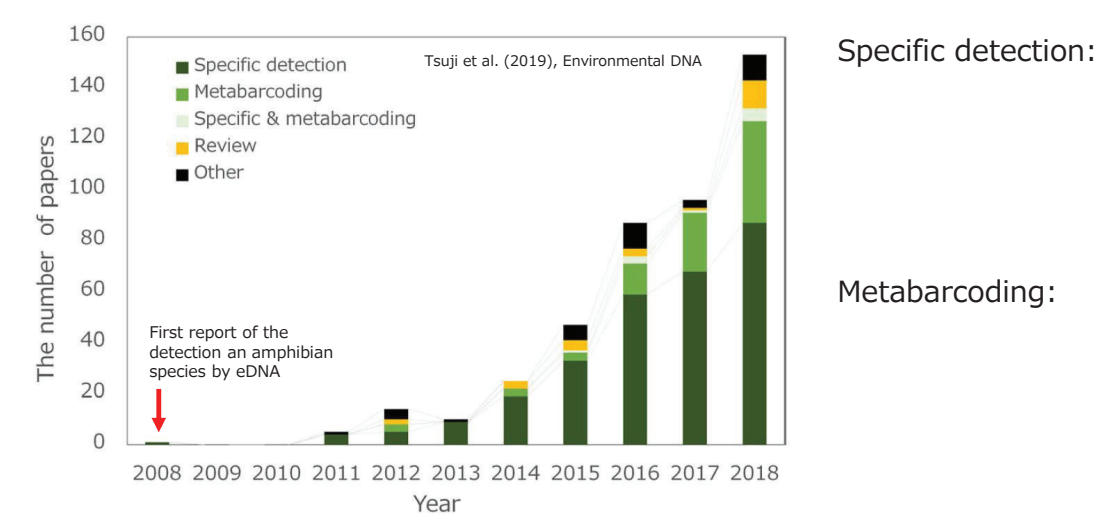
Base (A: Adenine, T: Thymine, G: Guanine, C: Cytosine)  
Colors mean the types of base.

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# What's environmental DNA (eDNA)?



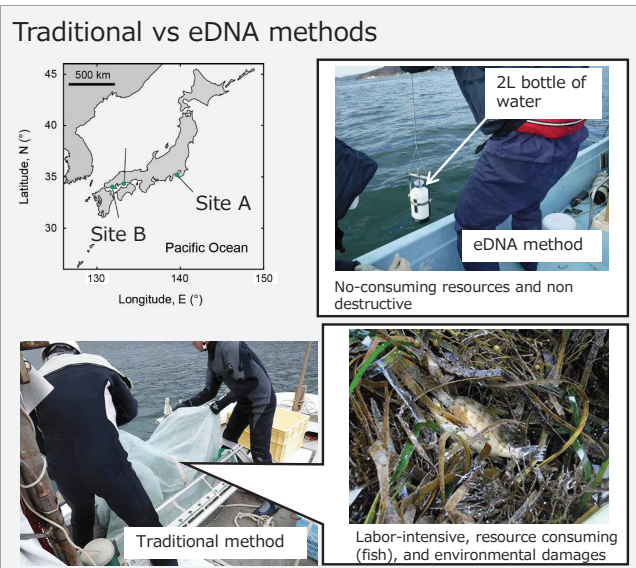
- Widely studied and used in the fields of research and development
- Many suppliers of the eDNA technology (in Japan)



# Advantages of the eDNA technology



- Performance of the eDNA metabarcoding in fish species
- Detection by eDNA metabarcoding >> Catchment by a traditional method



|                              | Site A (Kurihama Bay)                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              | Site B (Kasado Bay)                                                                                                                                                                                                                                                                 |
|------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Traditional and eDNA methods | <div>Acentrogobius pflaumii (21/24)</div> <div>Aulichthys japonicus (1/24)</div> <div>Ditrema viride (24/24)</div> <div>Favonigobius gymnauchen (10/24)</div> <div>Gobiidae (24/24)</div> <div>Heteromycteris japonica (1/24)</div> <div>Paracentropogon rubripinnis (22/24)</div> <div>Plotosus japonicus (2/24)</div> <div>Pseudoblennius cottoides (7/24)</div> <div>Pseudoblennius percoides (7/24)</div> <div>Pterogobius (5/24)</div> <div>Rudarius ercodes (14/24)</div> <div>Sebastes cheni (19/24)</div> <div>Sebastes inermis (19/24)</div> <div>Syngnathus schlegelii (1/24)</div> <div>Takifugu pardalis (23/24)</div> | <div>Acentrogobius pflaumii (13/24)</div> <div>Paracentropogon rubripinnis (1/24)</div> <div>Pseudoblennius cottoides (3/24)</div> <div>Rudarius ercodes (9/12)</div> <div>Sebastes cheni (15/24)</div> <div>Sebastes inermis (15/24)</div> <div>Syngnathus schlegelii (1/24)</div> |
| Only traditional             | <div>Apogonichthyoides niger</div> <div>Hippocampus coronatus</div> <div>Pholis crassispina</div>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                  | <div>Hippocampus coronatus</div>                                                                                                                                                                                                                                                    |
| Only eDNA                    | <div>Carassius cuvieri, ゲンゴロウブナ (1/24)</div> <div>Cyprinus carpio, コイ (21/24)</div> <div>Oncorhynchus sp., タイヘイヨウサケ属の1種 (1/24)</div> <div>Paramisourinus, ドジョウ属 (1/24)</div> <div>Other 58 species</div>                                                                                                                                                                                                                                                                                                                                                                                                                           | <div>Carassius spp., フナの種類 (3/24)</div> <div>Lepomis macrochirus, ブルーギル (1/24)</div> <div>Other 44 species</div>                                                                                                                                                                    |

Hosokawa et al. (2020), Tech Note PARI, No.1373

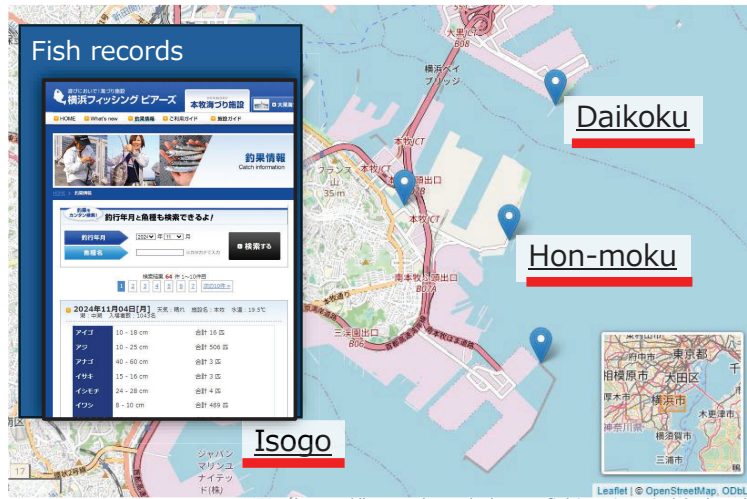


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# Advantages of the eDNA technology

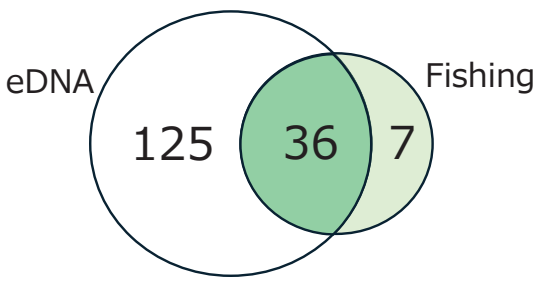


- Performance of the eDNA metabarcoding in fish species
- Detection by eDNA metabarcoding >> Catchment by fishing



- Fish records by the Yokohama Fishing Piers (YFP)
- eDNA sampling and metabarcoding analysis

Between  
January 2021 and March 2023



Munakata et al. (2024), the 71<sup>st</sup> Coastal Engineering, JSCE

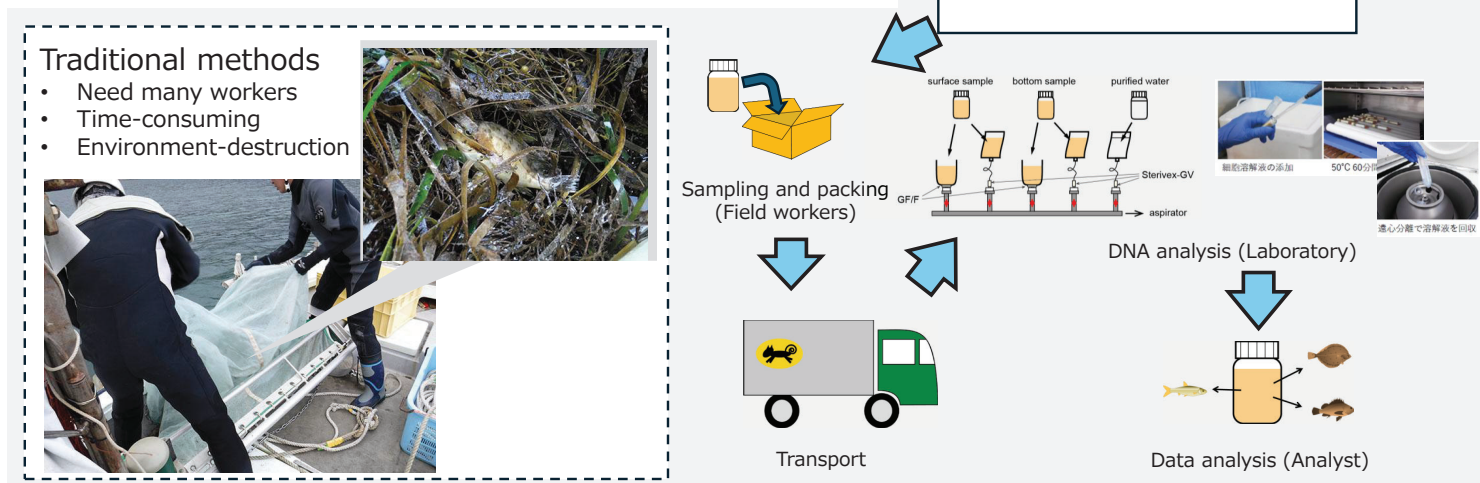


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# Advantages of the eDNA technology



- Good performance
- Non-destructive
- Cost-effective: Need several processes but effective



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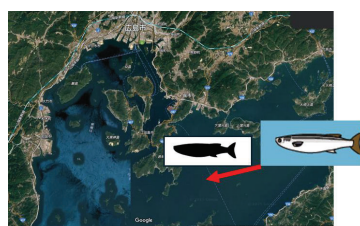
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# Challenges in using eDNA technology in estuaries and coasts

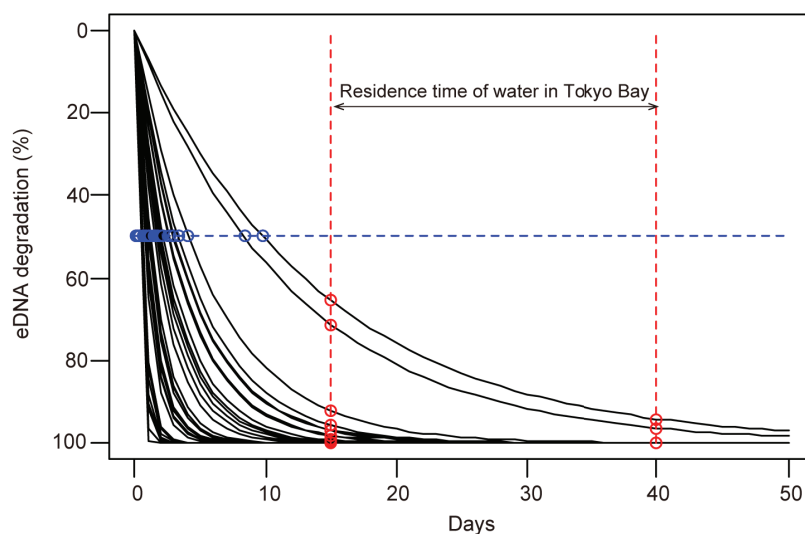
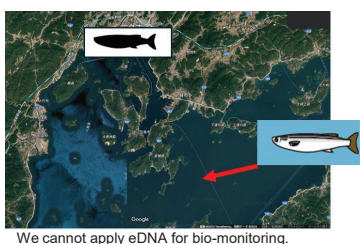


- Does the eDNA distribution mean the fish distribution?

Suitable eDNA degradation:



Without eDNA degradation:



Collins et al. (2018) Communications Biology



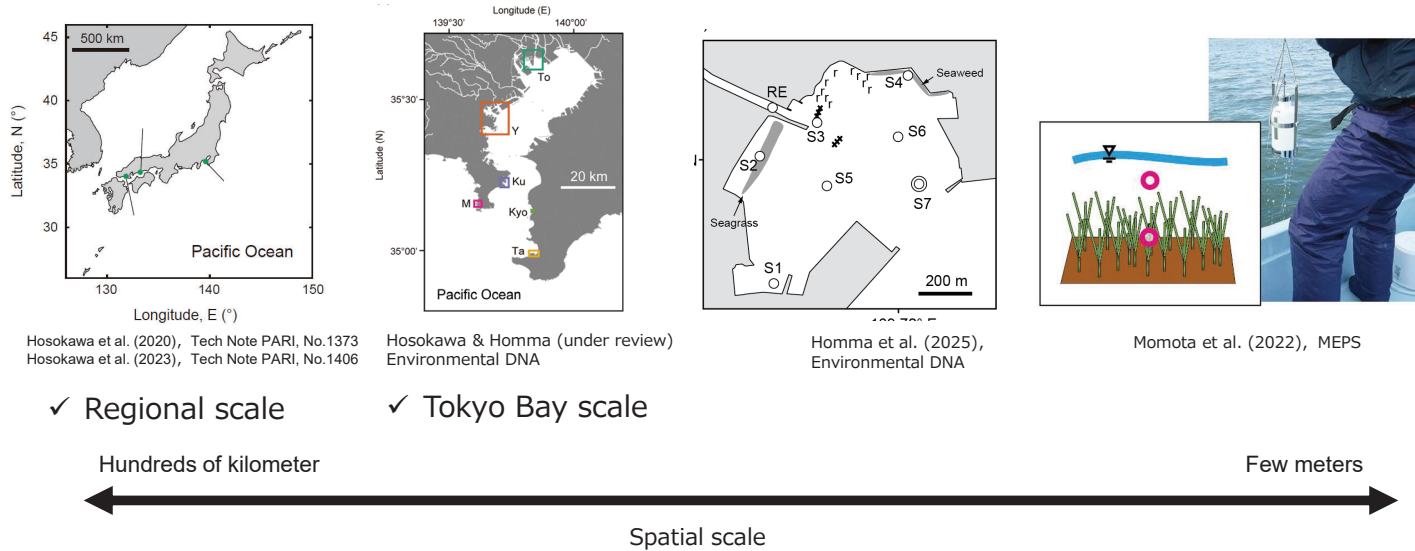
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# Challenges in using eDNA technology in estuaries and coasts



- Thousands of meters of dispersal in marine areas (Jo et al. 2025)

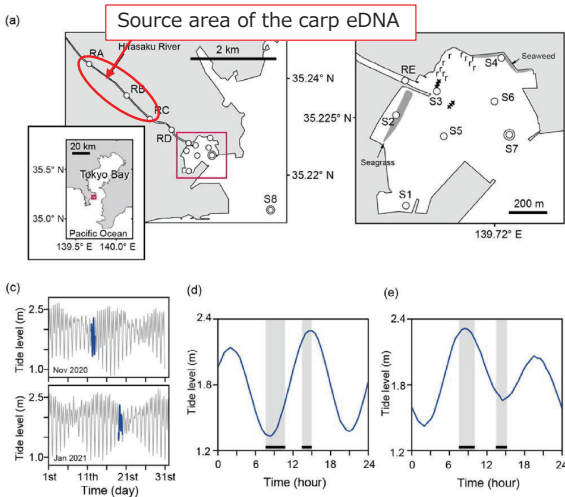


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# Challenges in using eDNA technology in estuaries and coasts

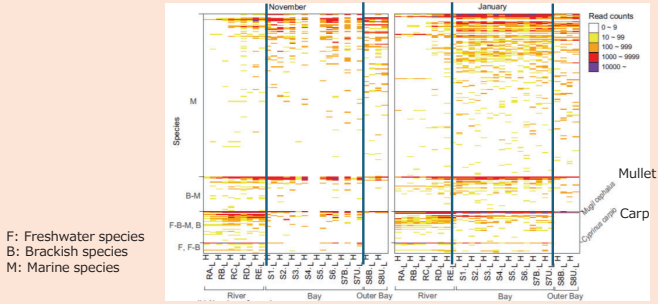


- Topographical changes govern the distribution of eDNA.

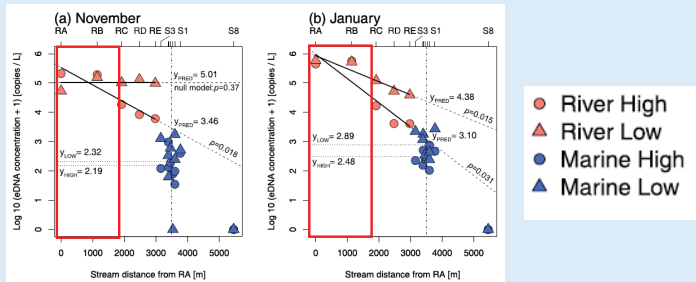


Homma et al. (2025), Environmental DNA

Metabarcoding: Species compositions in river, bay, and outer bay



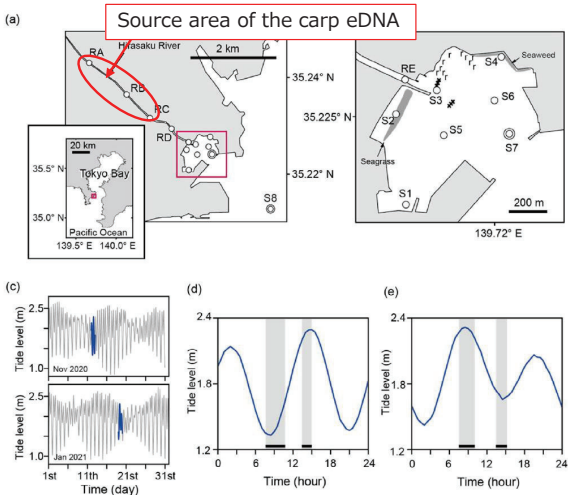
Species-specific analysis: The eDNA conc. of carp decreased in the bay.



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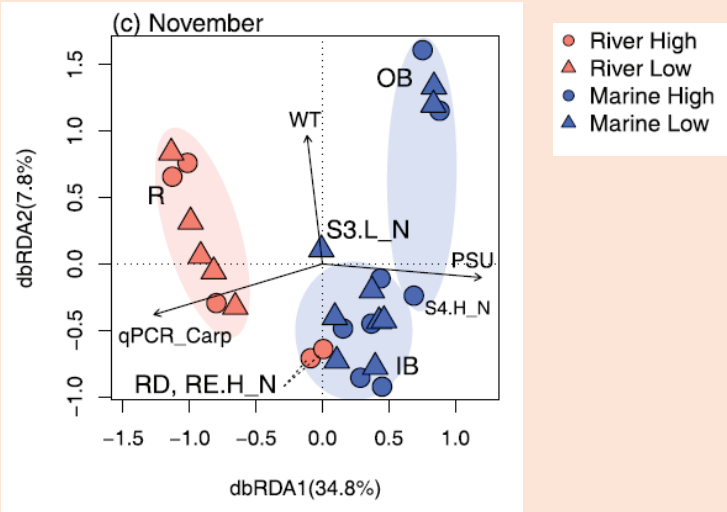
# Challenges in using eDNA technology in estuaries and coasts

- Geographical transition zone is not suitable for eDNA monitoring.



Homma et al. (2025), Environmental DNA

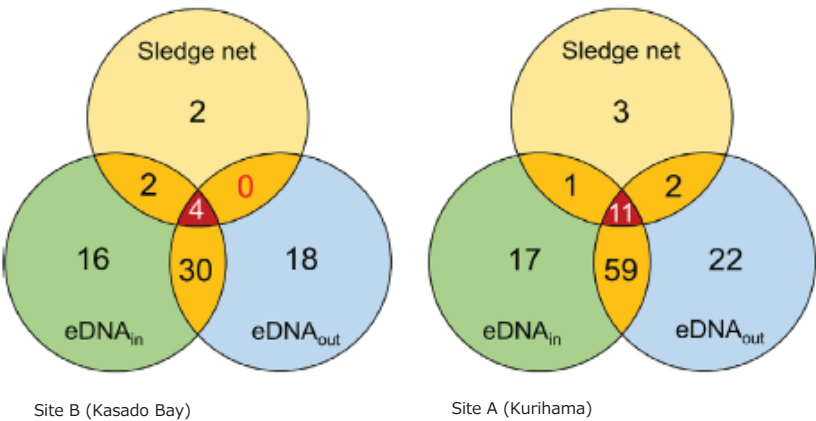
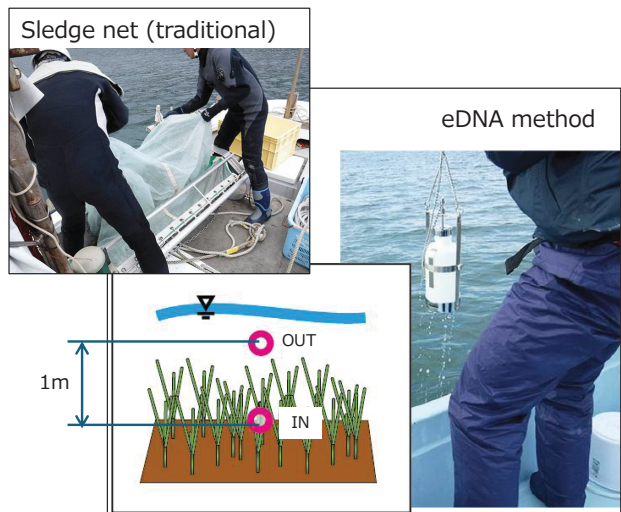
Metabarcoding: Species compositions in river, bay, and outer bay



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# Challenges in using eDNA technology in estuaries and coasts

- eDNA captures the small-scale heterogeneity of fish diversity in the vegetations.



Momota et al. (2022), MEPS

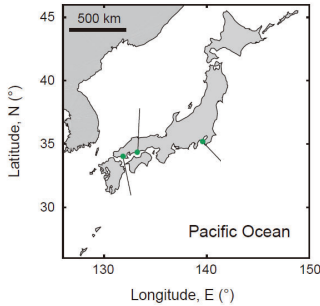


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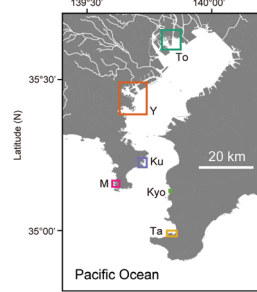
# Challenges in using eDNA technology in estuaries and coasts



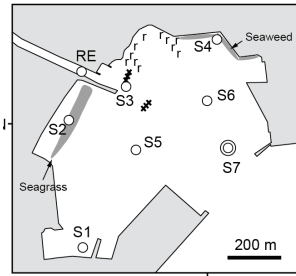
- Thousands of meters of dispersal in marine areas (Jo et al. 2025)
- Topography and vegetations are key factors of the eDNA distributions in fine scale.



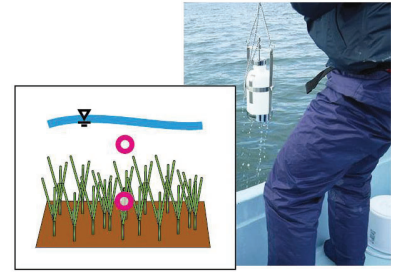
Hosokawa et al. (2020), Tech Note PARI, No.1373  
Hosokawa et al. (2023), Tech Note PARI, No.1406



Hosokawa & Homma (under review)  
Environmental DNA



Homma et al. (2025),  
Environmental DNA



Momota et al. (2022), MEPS

✓ Regional scale

✓ Tokyo Bay scale

Hundreds of kilometer

Few meters

Spatial scale



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# Challenges in using eDNA technology in estuaries and coasts

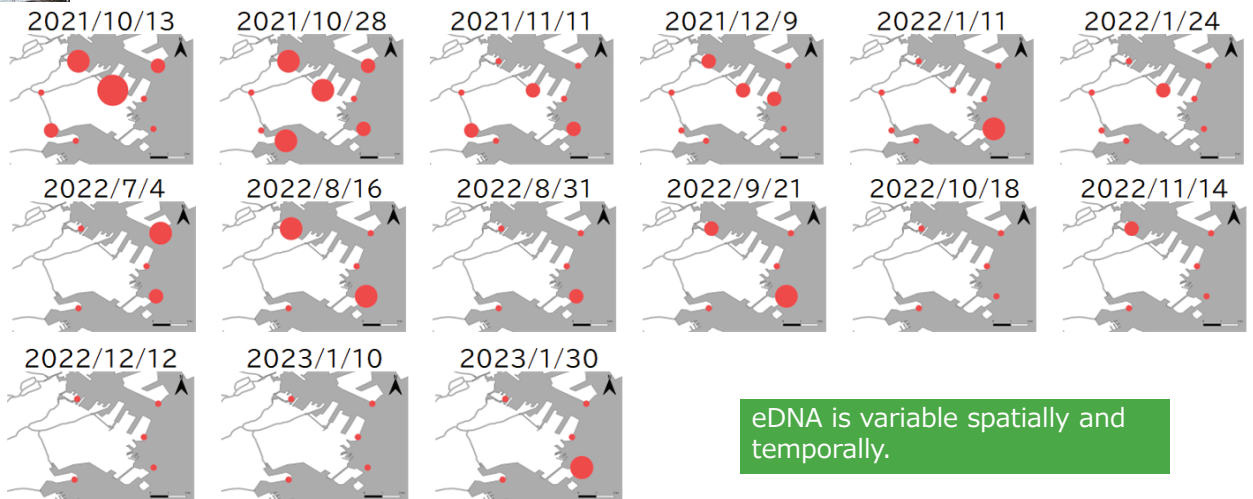


- DNA concentration of Black sea bream in Yokohama Port (yy/mm/dd)



*Acanthopagrus* sp. (Black sea bream )

eDNA (copies/uL) : 0 ●●●●● 603  
(quantified by qMiFish)



eDNA is variable spatially and temporally.

Munakata et al. (2024), the 71<sup>st</sup> Coastal Engineering, JSCE



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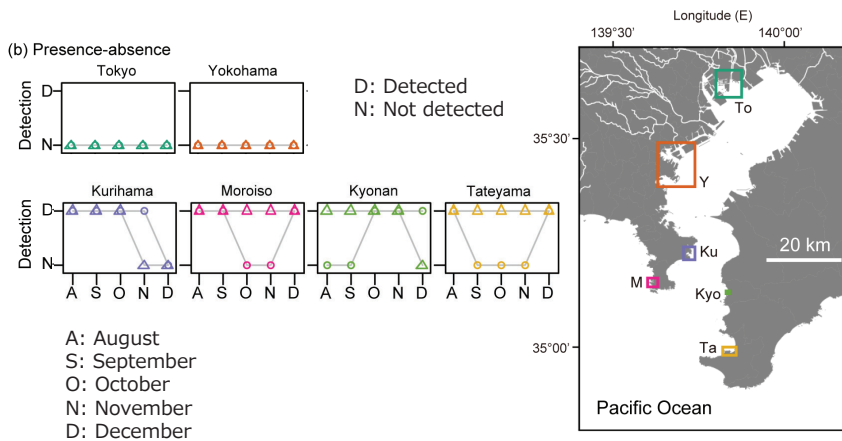
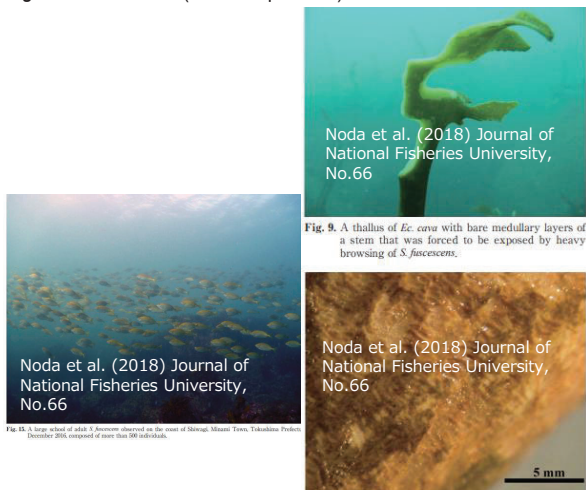
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# What will eDNA technologies contribute?



- Vegetations have been damaged by the mottled spinefoot in recent years.
- eDNA can detect the presence of the mottled spinefoot along the Pacific Ocean.

*Siganus fuscescens* (Mottled spinefoot)



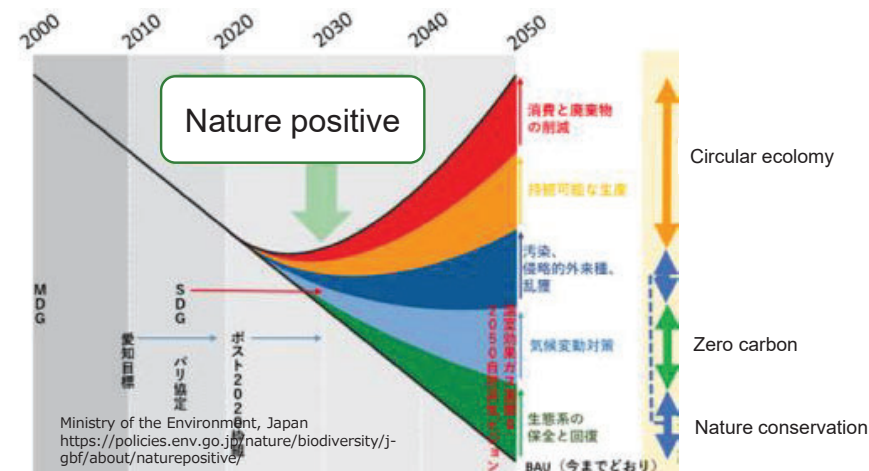
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# What will eDNA technologies contribute?



- Monitoring for the biodiversity change is important. However, targets are unclear.
- eDNA metabarcoding can capture the biodiversity and is suitable for the monitoring many species and biodiversity changes.



Kunming-Montreal Global Biodiversity Framework  
2050 Vision

A world of living harmony with nature

Ministry of the Environment, Japan  
<https://policies.env.go.jp/nature/biodiversity/j-gbf/about/naturepositive/>

2030 Mission

To take urgent action to halt and reverse biodiversity loss to put nature on a path to recovery

Nature positive



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# Collaborations with open data platform



- Visualization tool for eDNA data (UMI-POCHI)

**Biodiversity Database**

Batch download of databases 日本語

**Conditions and agreement**

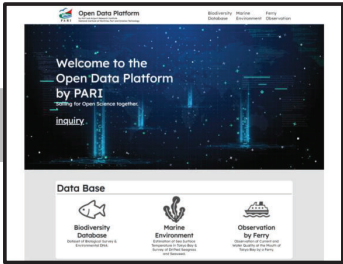
- This site provides a biodiversity database released by the Marine Environmental Information Research Group of the Port and Airport Research Institute.
- You can download the partial data ("download csv") and all data ("Batch download of database") for academic or personal use only.
- We maintain the database carefully. However, some errors may exist. Please use it at your own risk.
- If you wish to use the data from this site in your academic paper, please state the URL of this site and notify us in advance of publication (see [UMI-POCHI](#) to contact us).
- Reproduction of the database is prohibited.

**You can download the all data**

result type condition

**Tokyo Bay Wakasu (2019-1-25)**

2019-1-25 2019-1-25 2019-1-25



<https://pari.mpat.go.jp/biodiversity/>

UMI-POCHI is an open data platform produced in PARI

Visualization of the eDNA results



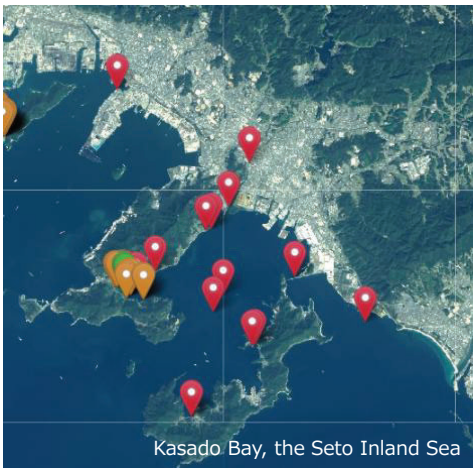
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# Collaborations with open data platform

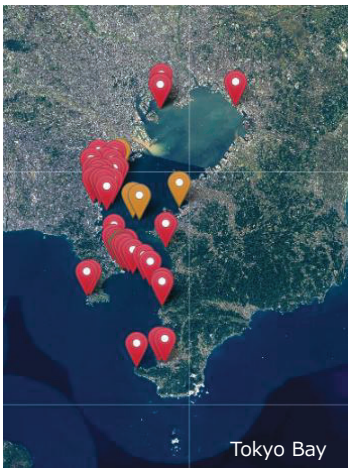


- Dense eDNA data in Tokyo Bay and Kasado Bay
- Why don't you share your eDNA data?

“UMI-POCHI”



<https://pari.mpat.go.jp/biodiversity/>

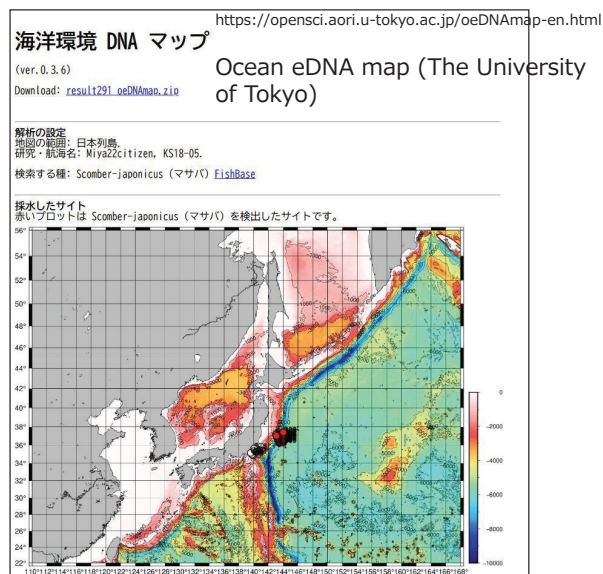
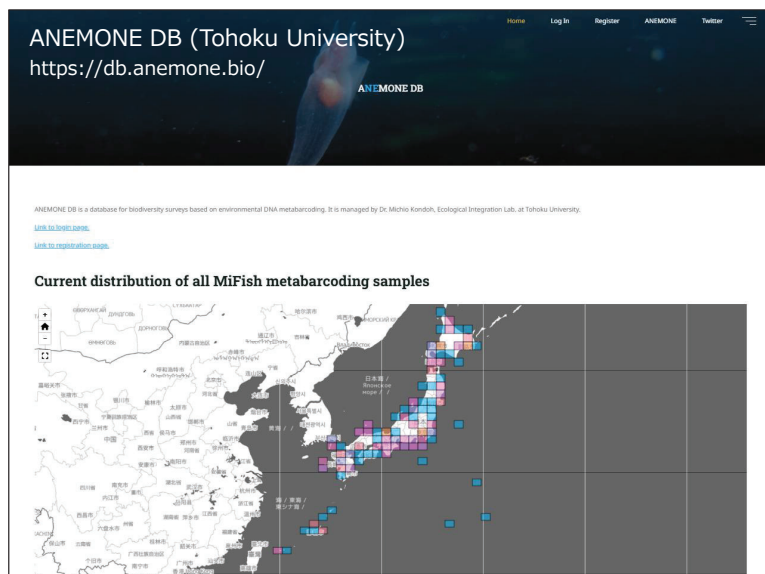


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## Collaborations with open data platform



- Open data platforms that share eDNA data are being launched to accelerate scientific progress.



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## Conclusions



- Environmental DNA (eDNA) technologies are non-destructive and cost-effective, and demonstrate good performance.
- Challenges remain in using eDNA technology in estuaries and coasts. However, eDNA technology is a powerful tool for capturing the spatial scale greater than several kilometers.
- eDNA technologies will serve as a tool for monitoring invasive species and biodiversity changes.
- Open data platforms that share eDNA data are being launched. Collaborating with the platform will accelerate scientific progress based on eDNA data.



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# Thank you for your attention

## Acknowledgements

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