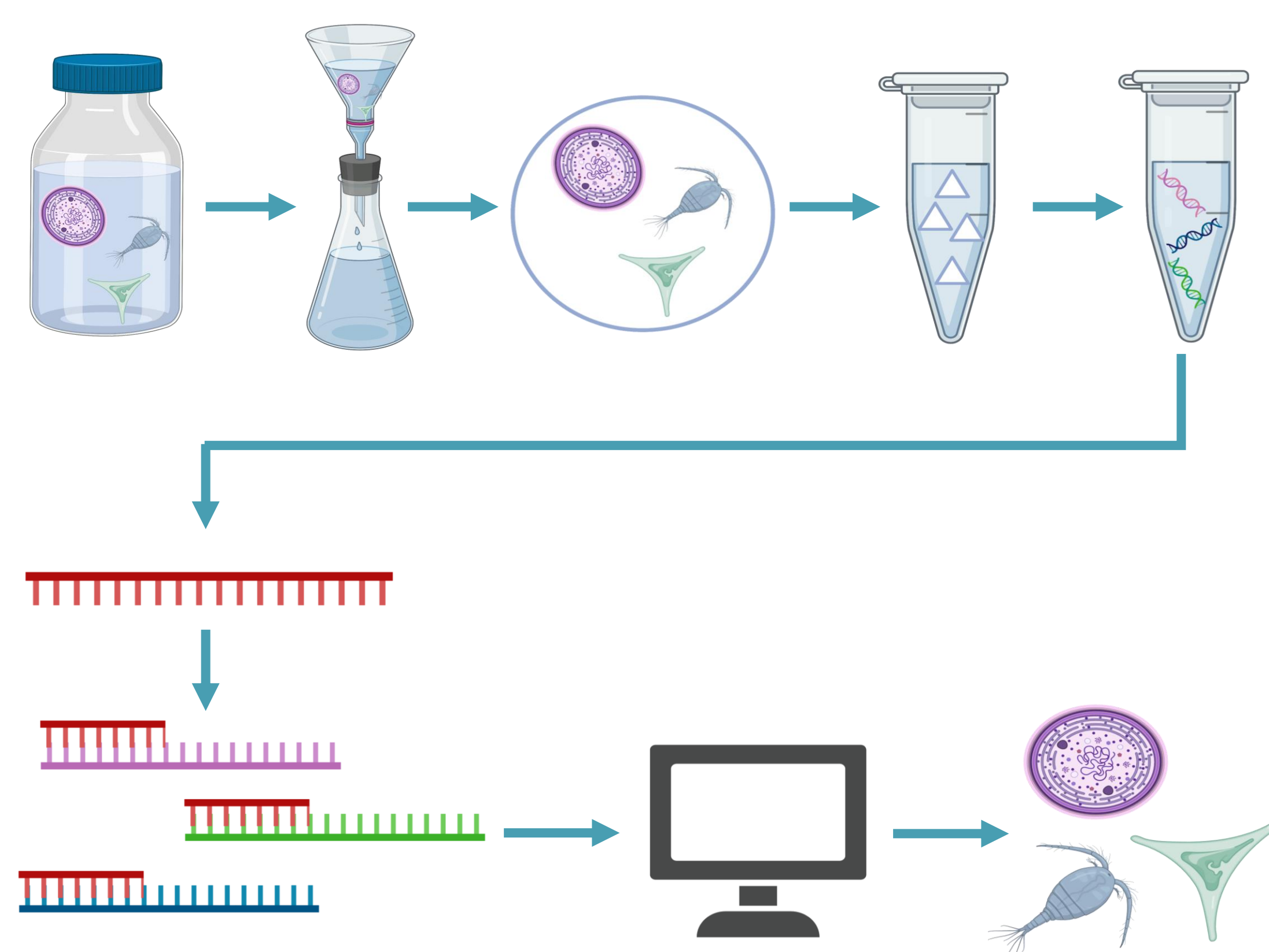


## BACKGROUND

Plankton communities are key components of estuarine ecosystems and serve as bioindicators of environmental changes. DNA metabarcoding is a tool that can efficiently expand upon traditional plankton monitoring methods.

## OBJECTIVE

Optimize a DNA metabarcoding protocol for monitoring plankton communities in the Guana Tolomato Matanzas (GTM) National Estuarine Research Reserve.



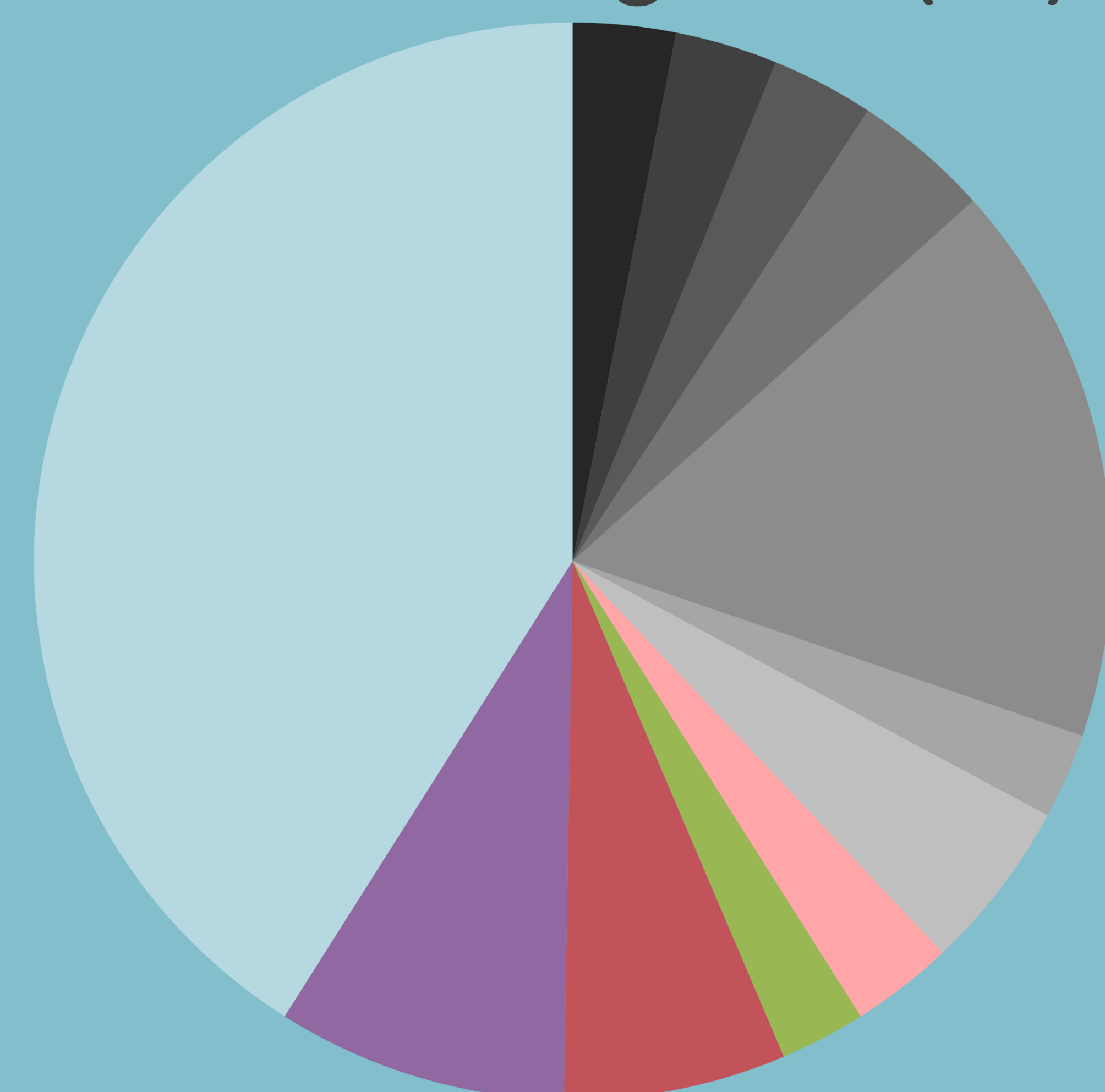
## METHODS

Samples were collected at Ft. Matanzas (FM) and Lake Middle (LM) for metabarcoding and microscopy analysis:

1. DNA was extracted using 7 different kits, which differ in lysis method, PCR inhibitor removal, and cost.
2. The 18S V9 rRNA region was amplified and sequenced following a two-step PCR protocol and Illumina MiSeq.
3. Amplicon sequence variants (ASVs; unique sequences) were determined using an in-house bioinformatics pipeline.
4. Taxa were compared between one metabarcoding and microscopy sample from FM.

# DNA metabarcoding detects similar plankton communities to microscopy.

## Metabarcoding ASVs (≥5)



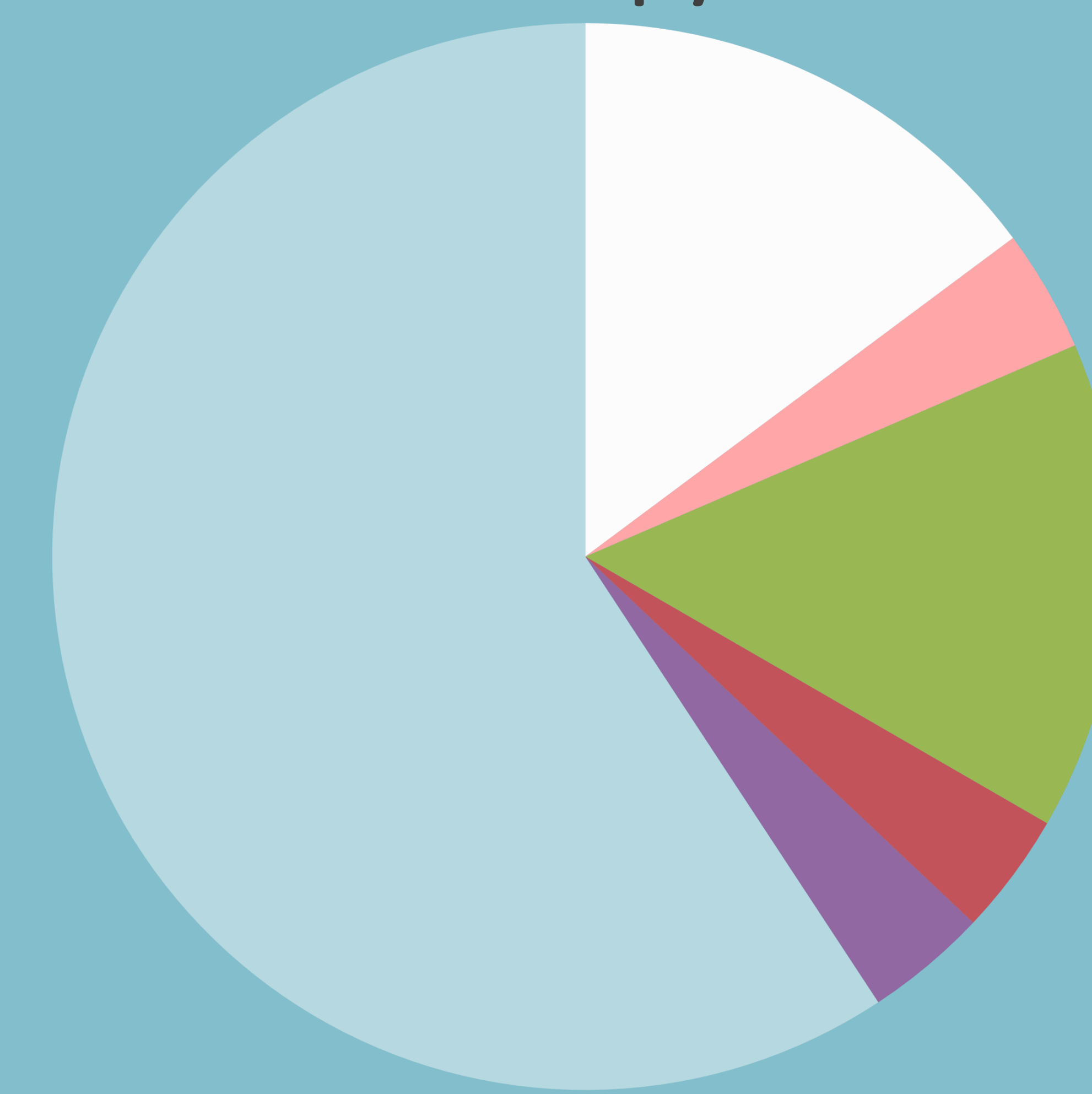
### Shared Phyla

- Bacillariophyta
- Chlorophyta
- Cryptophyta
- Myxozoa
- Porifera

### Other Metabarcoding Phyla (≥5 ASVs)

- Annelida
- Cnidaria
- Bigyra
- Mollusca
- Oomycota
- Cercozoa
- Ochrophyta

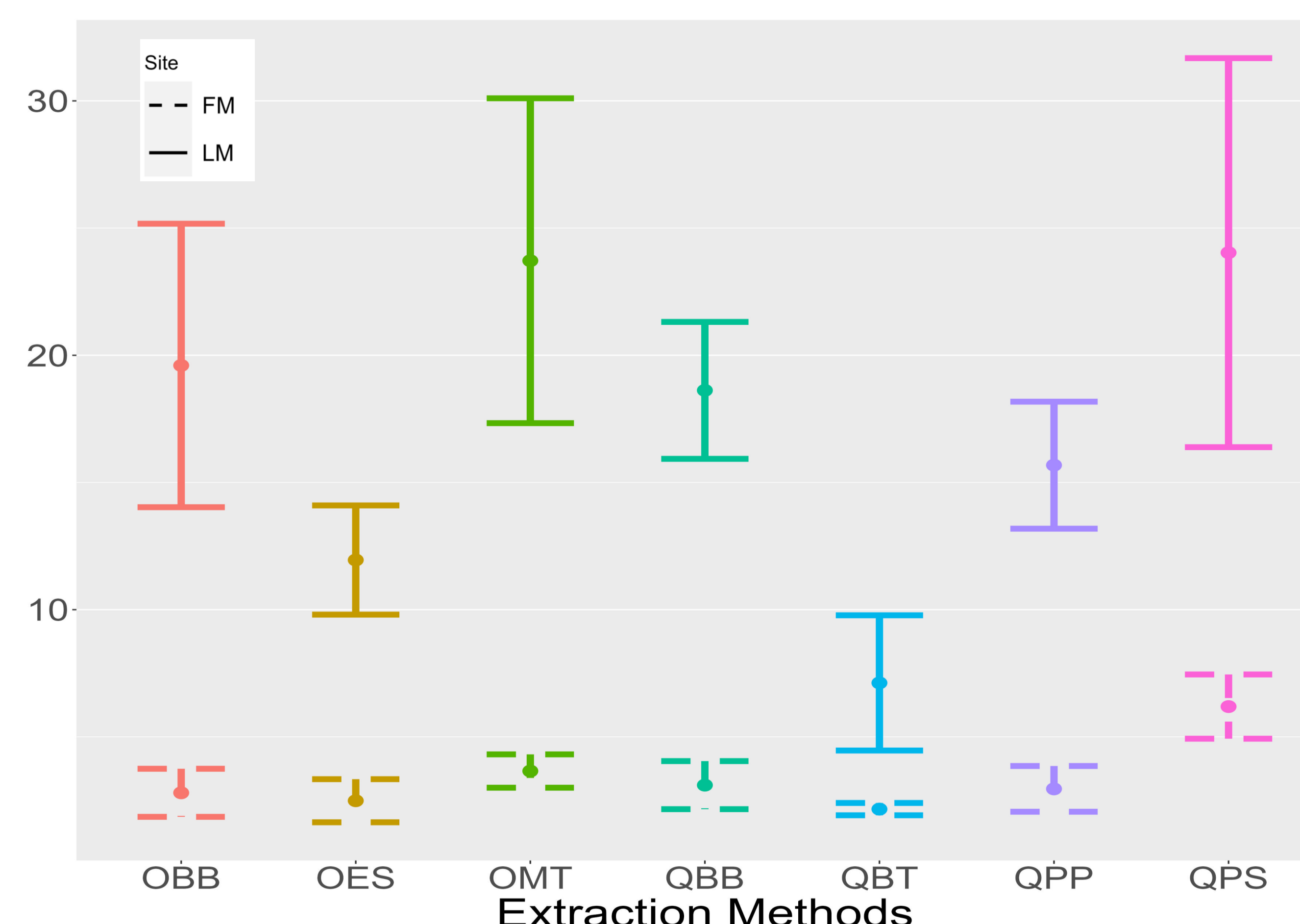
## Microscopy Taxa



### Other Microscopy Phyla

- Cyanophyta

## DNA CONCENTRATION (nG/μL)



## PRELIMINARY RESULTS

- Turbid water (LM site) produced greater mean DNA concentrations.
- The Qiagen Power Soil Kit resulted in a significantly higher ( $p < 0.05$ ; LM) mean DNA concentration than all kits in turbid water and 3 kits (OES, OMT, & QPP) in clear water.
- Metabarcoding detected 22 unique phyla and microscopy detected 1 unique phylum (a bacteria).
- A similar composition of phyla were resolved between both methods.
- Metabarcoding improved taxonomic resolution.

## NEXT STEPS

- Optimize the bioinformatics pipeline to generate ASVs for remaining samples and compare to multiple databases.
- Sequence using the 16S V3-V4 primer region to detect bacteria.
- Use this toolkit to sequence plankton from 10 sites across the GTM over the course of a year and synthesize these data with water quality.