

**Title:**

Mitochondrial metagenomics provides a reliable and scalable method for assessing eukaryotic freshwater biodiversity.

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**Abstract (300 words maximum): :**

Climate change and increasing pollution are accelerating the global biodiversity decline. This is especially true in freshwater habitats, where small shifts in seasonal mixing/stratification regimes can lead to significant changes in the local community composition. Species loss can undermine the stability and resilience of ecosystems, potentially triggering cascading effects that disrupt their ecological balance and reduce their ability to recover from disturbances. This could have severe consequences on the ecosystem services upon which we heavily rely on. Lake Zurich, for instance, serves as a vital drinking water source for its region, highlighting the critical need to safeguard its ecological integrity. Monitoring and understanding the changing biodiversity will be essential for fostering a sustainable relationship with our environment. To effectively track and verify community changes on a broader scale, new monitoring methods are needed that are both reliable and scalable.

We propose that implementing a mitochondrial-based monitoring method offers an efficient and straightforward approach to measuring microbial eukaryotic diversity. Compared to classic monitoring methods like microscopy or 18S rRNA metabarcoding, our mitochondrial approach shines by minimizing human bias and providing significantly greater species-level resolution. The fast-evolving mitochondrial genome provides high variability in recent ancestry, enhancing accuracy in identifying recent evolutionary events. This improved resolution reduces ambiguity in species-level taxonomic identification, enabling more precise and reliable monitoring studies. The core of this study lays in assessing the feasibility of recovering mitochondrial genomes from metagenomic samples. Using a suite of bioinformatic tools, we first assembled mitochondrial genomes and then classified them into taxonomic groups, drawing inspiration from approaches used in prokaryotic metagenomics. Finally, we show how mitochondrial genomes can be used to monitor changes in the abundance of diverse organisms during the high-resolution sampling of a spring bloom event. Our approach represents a first step towards establishing a standardized monitoring pipeline and paves the way for a better understanding of Lake Zurich's microbial eukaryotic diversity.