The power of zooplankton metabarcoding: Resolving changes in habitat use by zooplankton across the California Current Ecosystem

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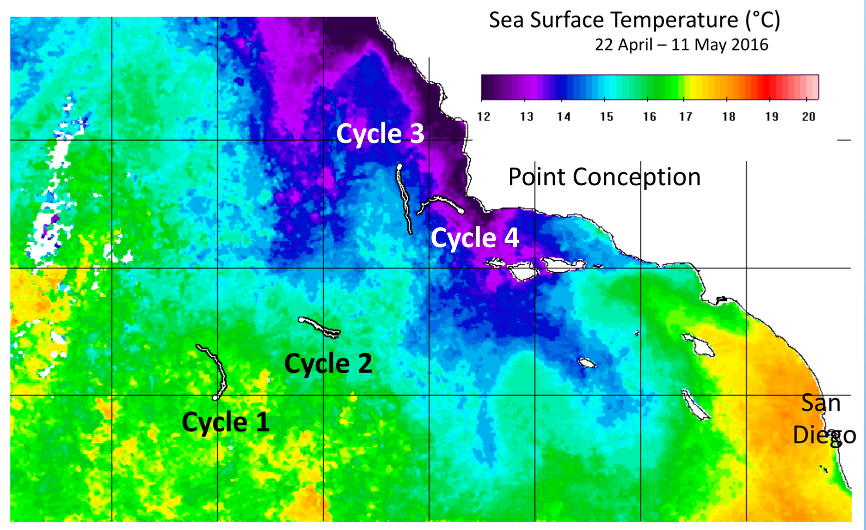
Questions

- Do zooplankton vertical habitats shift with major cross-shore environmental gradients?
- Does vertical habitat use covary with species' traits?
- Does feeding strategy shift within the zooplankton community in response to density stratification?

Introduction

- Vertical gradients in temperature, food availability, and predation risk tend to be compressed in upwelling regions and expanded in the stratified offshore.
- If zooplankton actively select their vertical habitat in response to such environmental cues, their vertical distributions should change with distance offshore.



Conclusions

- Clear vertical habitat shifts of DNA sequences as well as morphologically determined biomass
- OTU-specific vertical shifts best correlate with the depth of the 1% light level, a proxy for predation risk
- Stronger vertical shifts among carnivores than among herbivores
- Current feeding dominates, but feeding modes are more diverse offshore
- Offshore stratified waters, a spatial proxy for future oceans, indicate that warming may drive zooplankton deeper and shift the prey field available to both epipelagic and mesopelagic consumers

Cross-shore Shifts in Vertical Habitat by 2 Methods

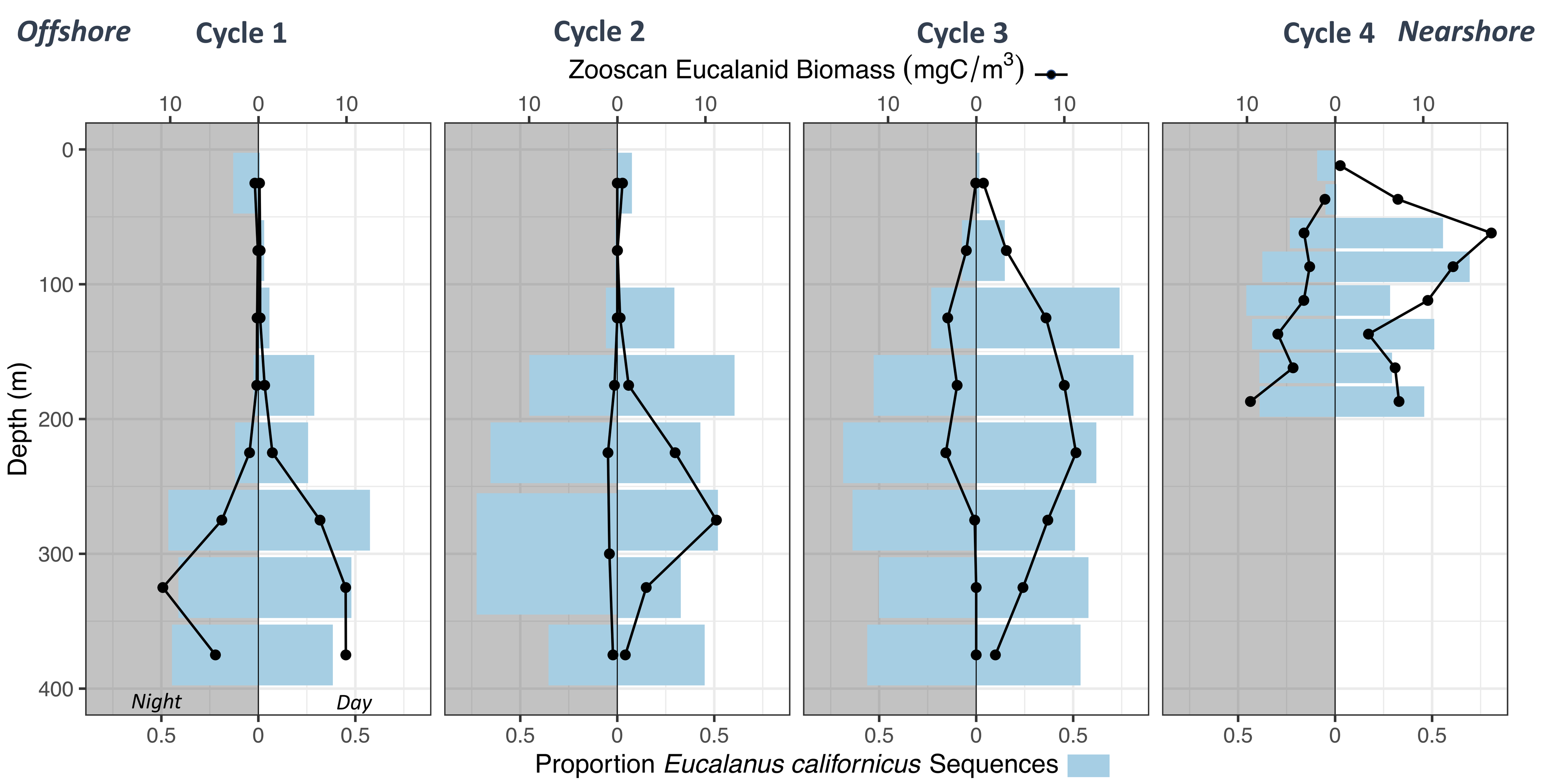


Fig 1. Both **ZooScan-assessed biomass** (circles and lines) and **mtCOI DNA sequences** (blue bars) reveal that the highest abundance of eucalanid copepods is found in subsurface waters, with the abundance peak deepening with distance offshore. There is good correspondence between the two methods, and sequencing appears to be more sensitive near the surface, where eggs and nauplii may be concentrated.

Habitat Shifts by Diet

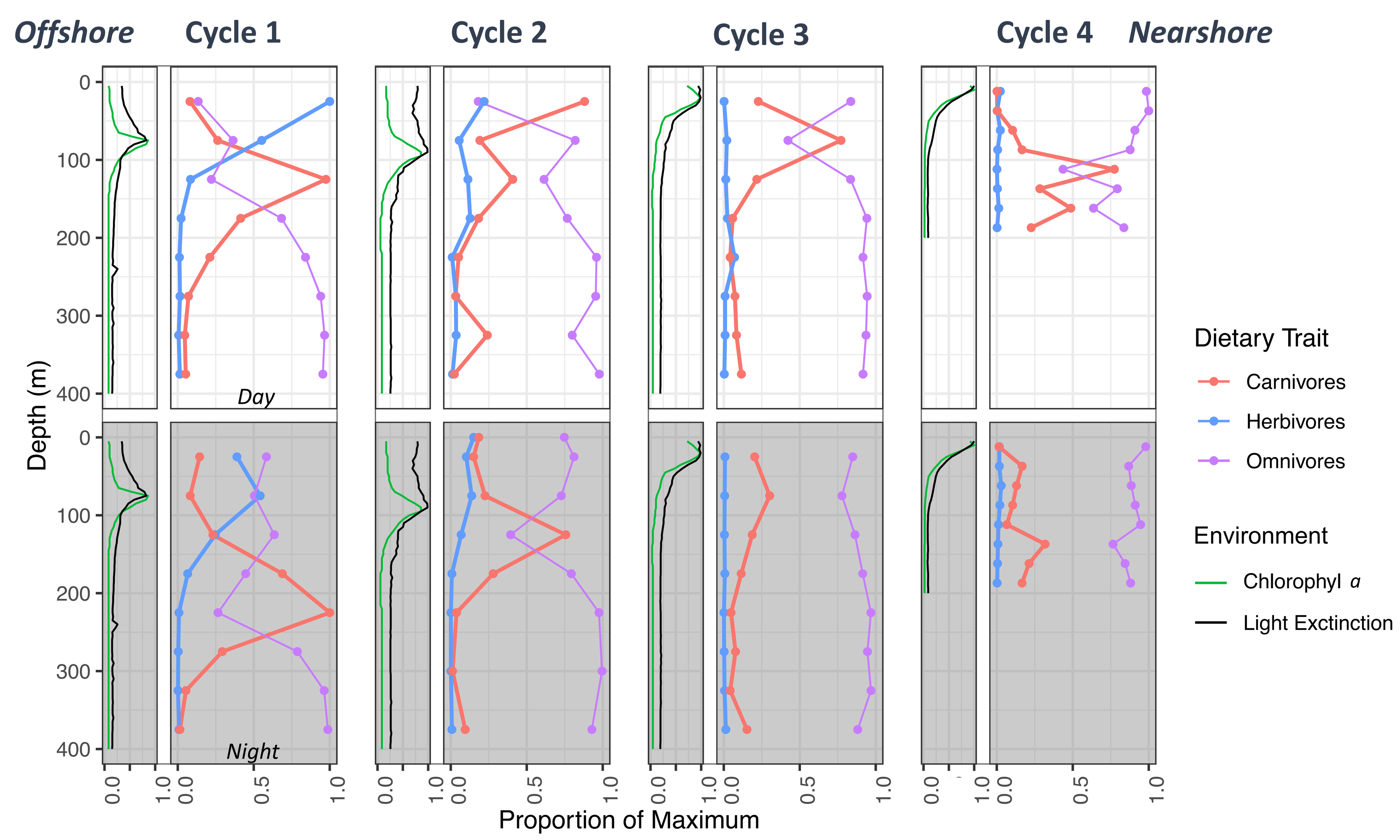


Fig 3. Carnivorous zooplankton shift to deeper depths in more stratified offshore waters. Offshore, herbivorous zooplankton vertical distributions become closely associated with the chlorophyll maximum. Traits were assigned to DNA sequences based on taxonomy.

Vertical Response to an Environmental Gradient

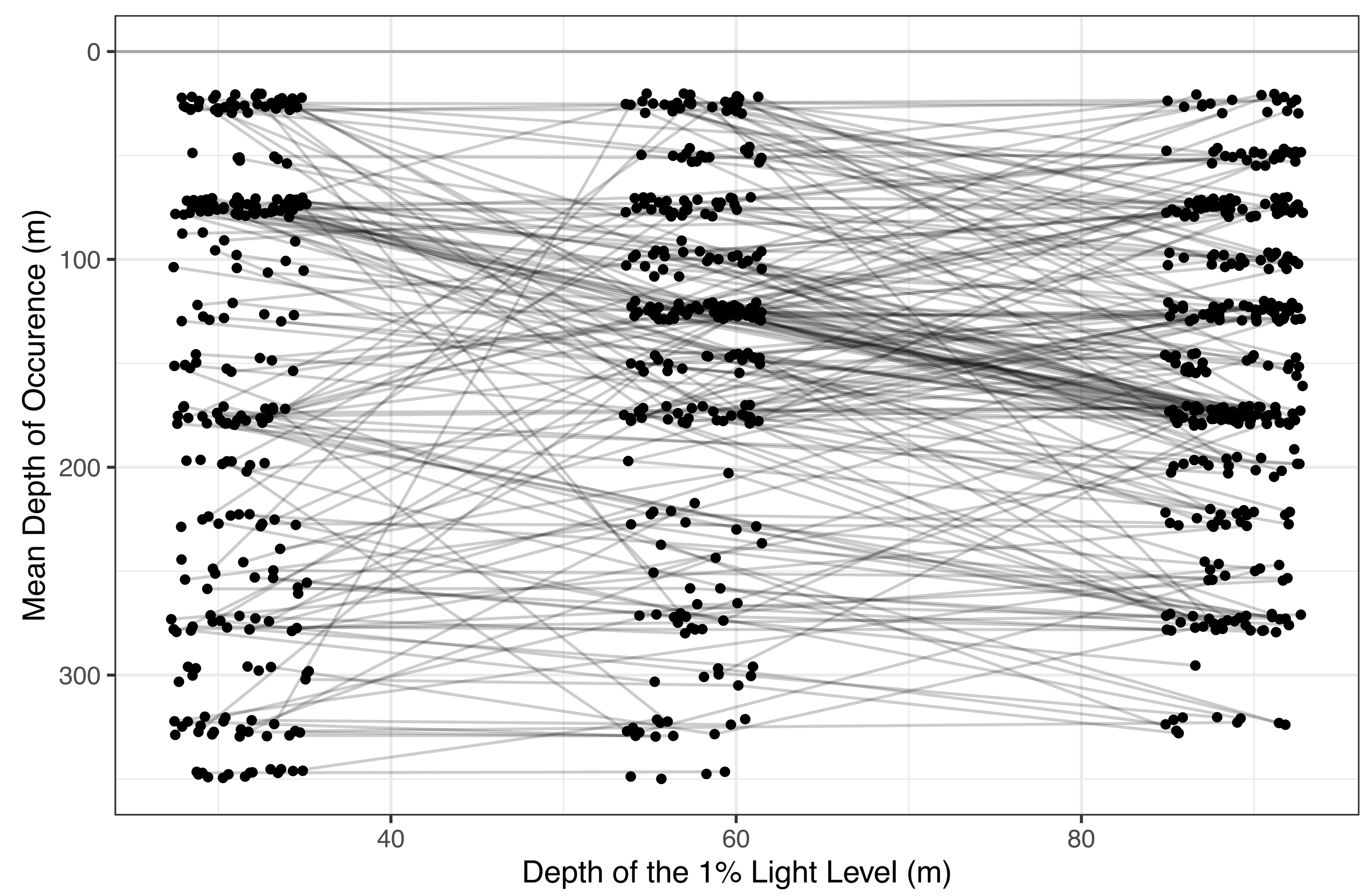


Fig 2. Many zooplankton OTUs have **deeper mean depth of occurrence with increasing light penetration**. The depth of the 1% light level is a better predictor than distance offshore or the depth of the chlorophyll maximum. Lines connect the same OTUs sampled in regions of different light penetration

Cross-shore Feeding Strategies

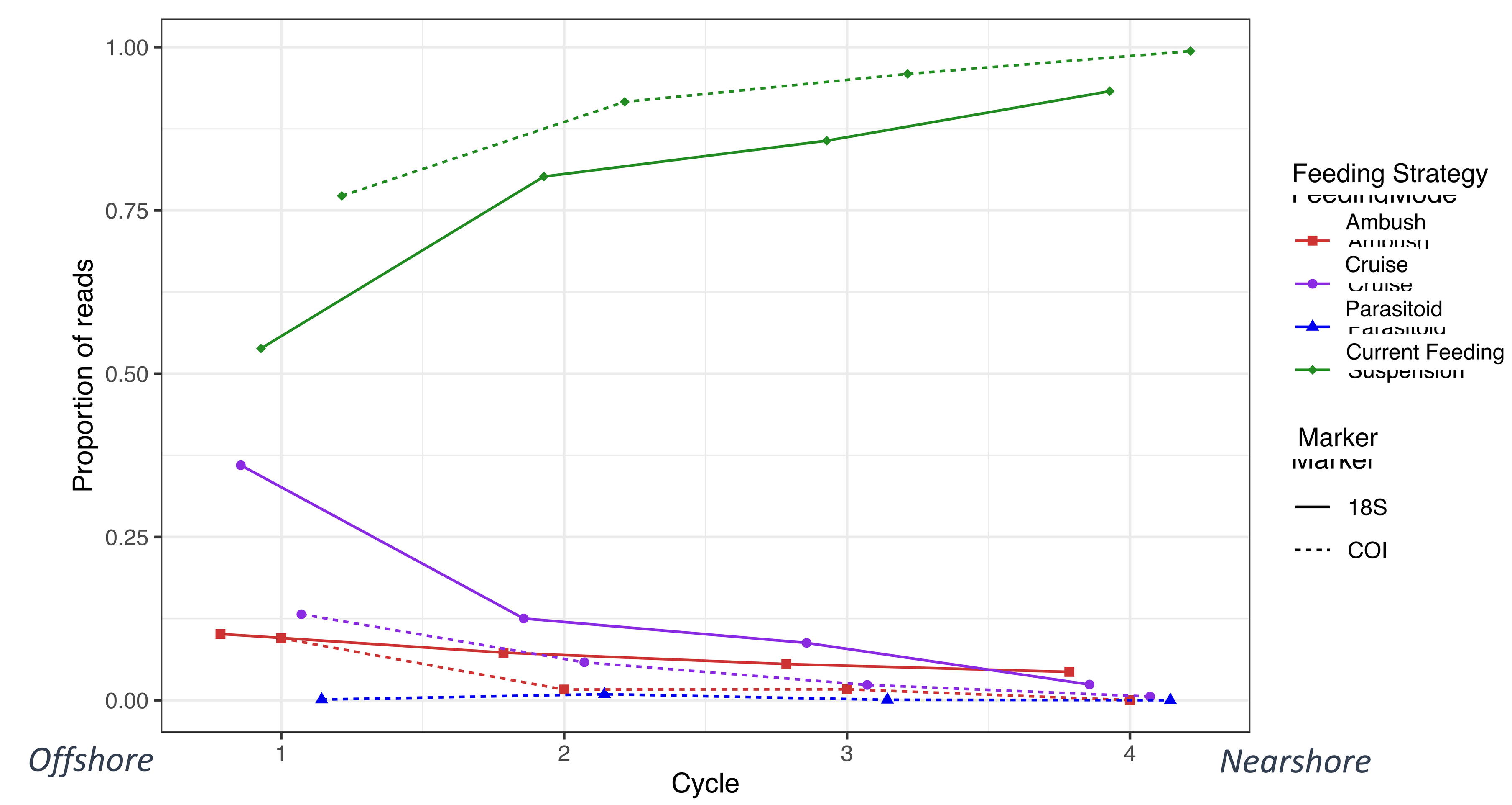


Fig 4. Current feeding dominates the community, but cruise, ambush, and parasitoid feeding becomes more common in the offshore waters. Cross-shore patterns detected by COI and 18S are similar. Traits were assigned to DNA sequences based on taxonomy, following Kiorboe (2011).

Methods

Sampling

Zooplankton were collected with vertically stratified paired day and night MOCNESS tows along an environmental gradient in the California Current Ecosystem (Cycles 1 – 4, above left) in April 2016, and split for preservation in 5% Formalin or 95% buffered EtOH (Matthews et al. 2021).

ZooScan Morphological Analysis

Zooplankton were imaged with a ZooScan, sorted into 27 taxonomic categories using machine learning, and all image classifications verified manually (Gorsky et al. 2010).

Traits

Traits were assigned to OTUs based on taxonomy. Traits included maximum size, diet, feeding behavior, spawning strategy, presence of asexual reproduction, DVM behavior, and carbon content

Metabarcoding Analysis

mtCOI (Leray et al. 2013) and 18S (v4-v5, Zhan et al. 2013) were amplified in triplicate with a 2-step PCR and library prep (Sommer et al. 2017). Bioinformatic analyses were conducted in QIIME2 (Bolyen et al. 2019). ASVs were inferred with DADA2 (Callahan et al. 2016), then clustered at 97% similarity to obtain COI OTUs. Taxonomy was assigned using the MetaZooGene database with an 80% confidence cutoff.

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