Title
Assessing Aquatic Invertebrate Establishment in a Restored Wetland Over Time

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Introduction

The Devereux slough spans two reserves: North Campus Open Space (NCOS) and Coal Oil Point Reserve (COPR). COPR has been a protected reserve for decades, while NCOS was restored from a golf course beginning in 2017. Ecological restoration at NCOS created over 40 acres of wetlands and improved over 60 acres of upland habitat. The slough is unique because it is intermittently connected to the ocean, and has varying levels of salinity throughout the year due to precipitation and breaching of the mouth. COPR, being closer to the ocean, tends to have higher salinity than NCOS.

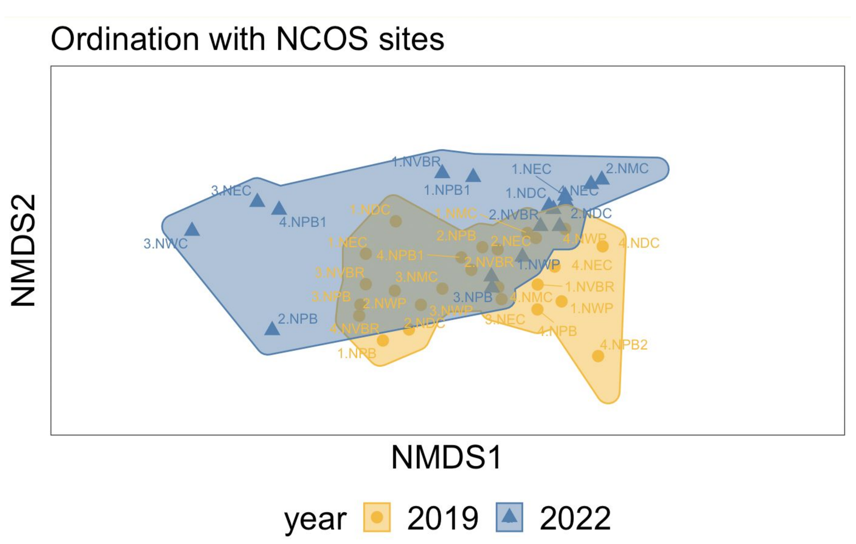
The Devereux Slough is home to endangered birds and fish such as the Snowy Plover and Tidewater Goby which rely on aquatic macroinvertebrates as a food source. Aquatic macroinvertebrates are often used as bioindicators of water and ecosystem quality due to their sensitivity to pollution. For these reasons, the Santa Barbara Audubon Society developed a Citizen Science program in collaboration with UCSB to collect long-term data on aquatic macroinvertebrate assemblages and water quality in order to understand the changing health of the slough. This project continues to receive funding from the Santa Barbara Audubon Society, and is now run by the Cheadle Center.

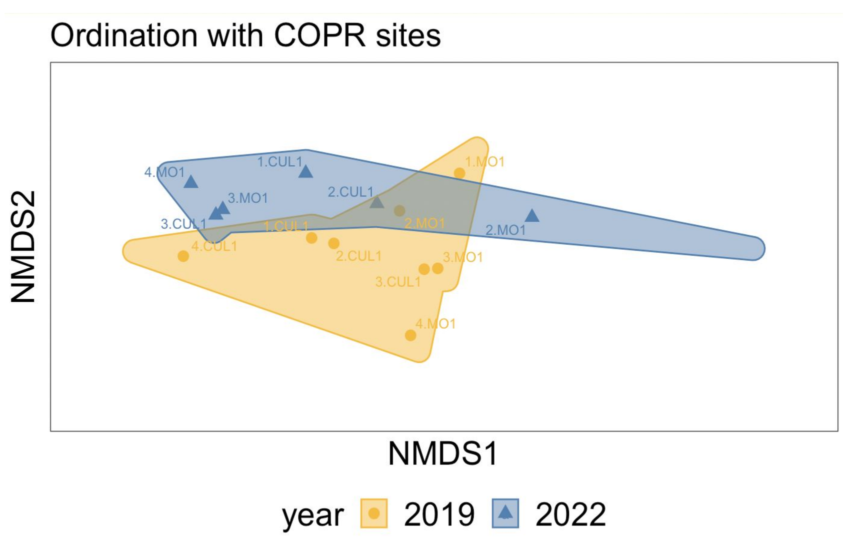
Methods

Groups of 2-6 students collected macroinvertebrates at sites in the Devereux Slough. A volume of 7.5 liters of water in 2018 and 2019 and 70 liters of water in 2021 and 2022 was filtered through a 0.25mm mesh. The filtered macroinvertebrates were then flushed into a vial using a 70 percent isopropyl alcohol. Two samples were taken at each site, one primary and one replica. At the same time, pH was measured using a pH meter. Dissolved oxygen and conductivity were measured using a YSI 2030 meter, at both 10 cm below the surface of the water and 5cm above the sediment. At each sampling site, aquatic eDNA (environmental DNA) samples were taken from just under the water surface with an eDNA kit from commercial provider Jonah Ventures. Samples were mailed to their lab where the DNA was extracted, amplified with the ArthCOI primer, and sequenced. Sequences were matched to taxa using NCBI BLAST.

At the lab, macroinvertebrates were counted and identified by student volunteers using a stereo microscope, macroinvertebrate key, and tally counter. Samples were verified and inputted to a data sheet by student leaders with more experience. Aquatic macroinvertebrate abundance data, water quality data, and eDNA data were cleaned and combined using R.

Results





Figures 1 and 2.
Invertebrate assemblage dimension reduction, using non-metric multidimensional scaling (nMDS) in the R vegan package. Assemblages from manual sampling in 2019 and 2022 are compared. Data from COPR and NCOS are separated. Organism counts were multiplied by 1000 in order to run the nMDS. Each point represents a sampling site and season. The numbers associated with each point represent the quarter of the year during which the sampling took place. The letters next to each point correspond to sampling sites at the slough. Points with a greater distance between them are less similar in terms of community composition while those closer together are more similar. Both ordinations have a stress of 0.17.

2019 rainfall (mm)	2022 rainfall (mm)
683.8	232.8

Table 1. Total rainfall in 2019 and 2022

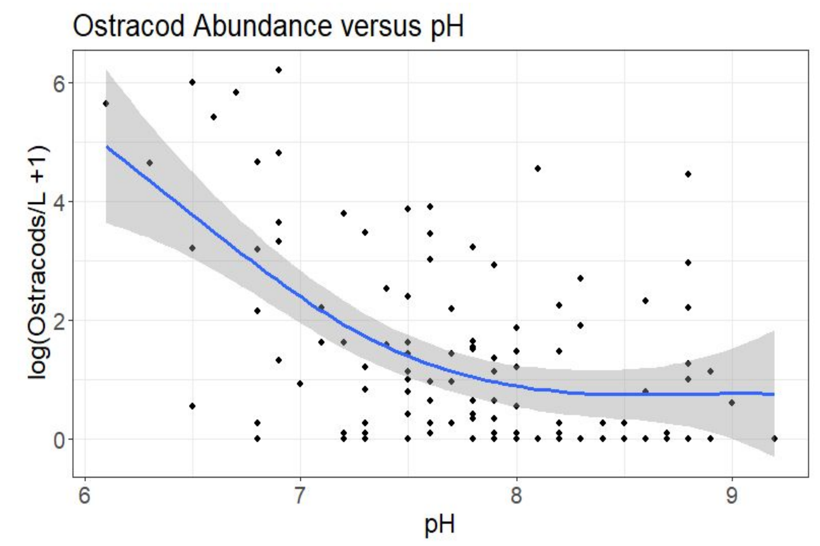


Figure 5. Logarithm of ostracods per liter compared to pH at the time and place of sampling. From manual sampling data.
Generalized additive model.
log(ostracods/L+1) ~ s(pH, bs = "cs")
R-sq = 0.312, p < 0.001.

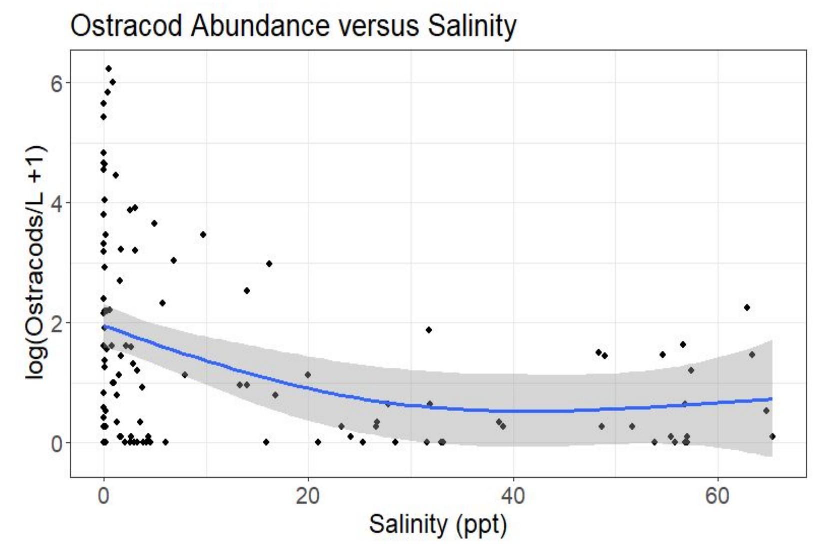


Figure 6. Logarithm of ostracods per liter compared to salinity (in ppt) at the time and place of sampling. From manual sampling data.
Generalized additive model.
log(ostracods/L+1) ~ s(salinity, bs = "cs")
R-sq = 0.134, p < 0.001.

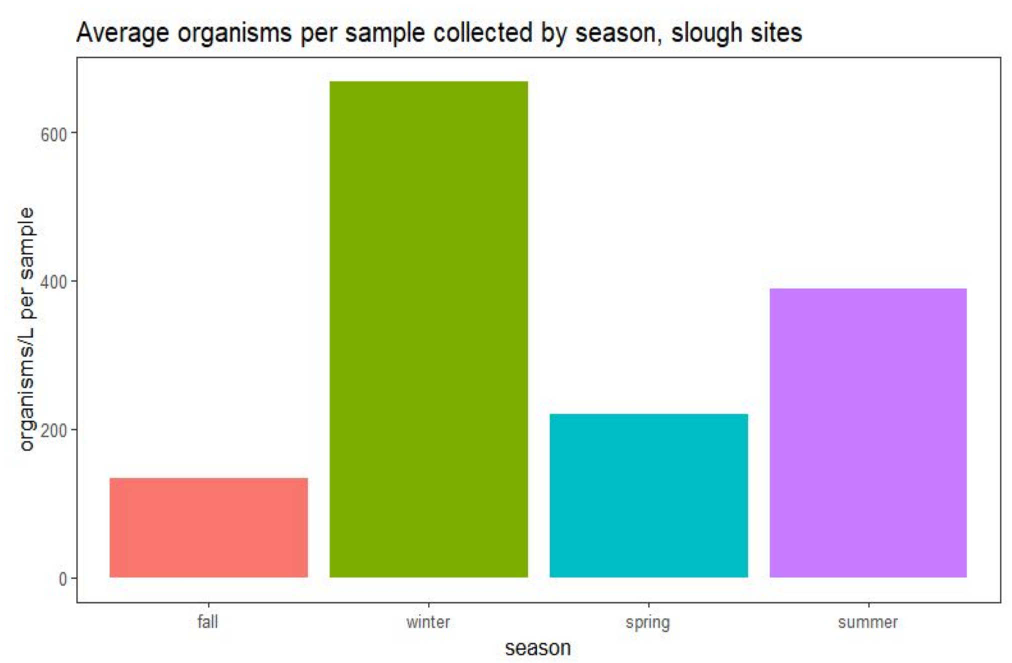


Figure 3 (left).
The average number of organisms/liter in each sample per season, from manual sampling at slough sites. The average concentration of organisms is highest in winter and lowest in fall.

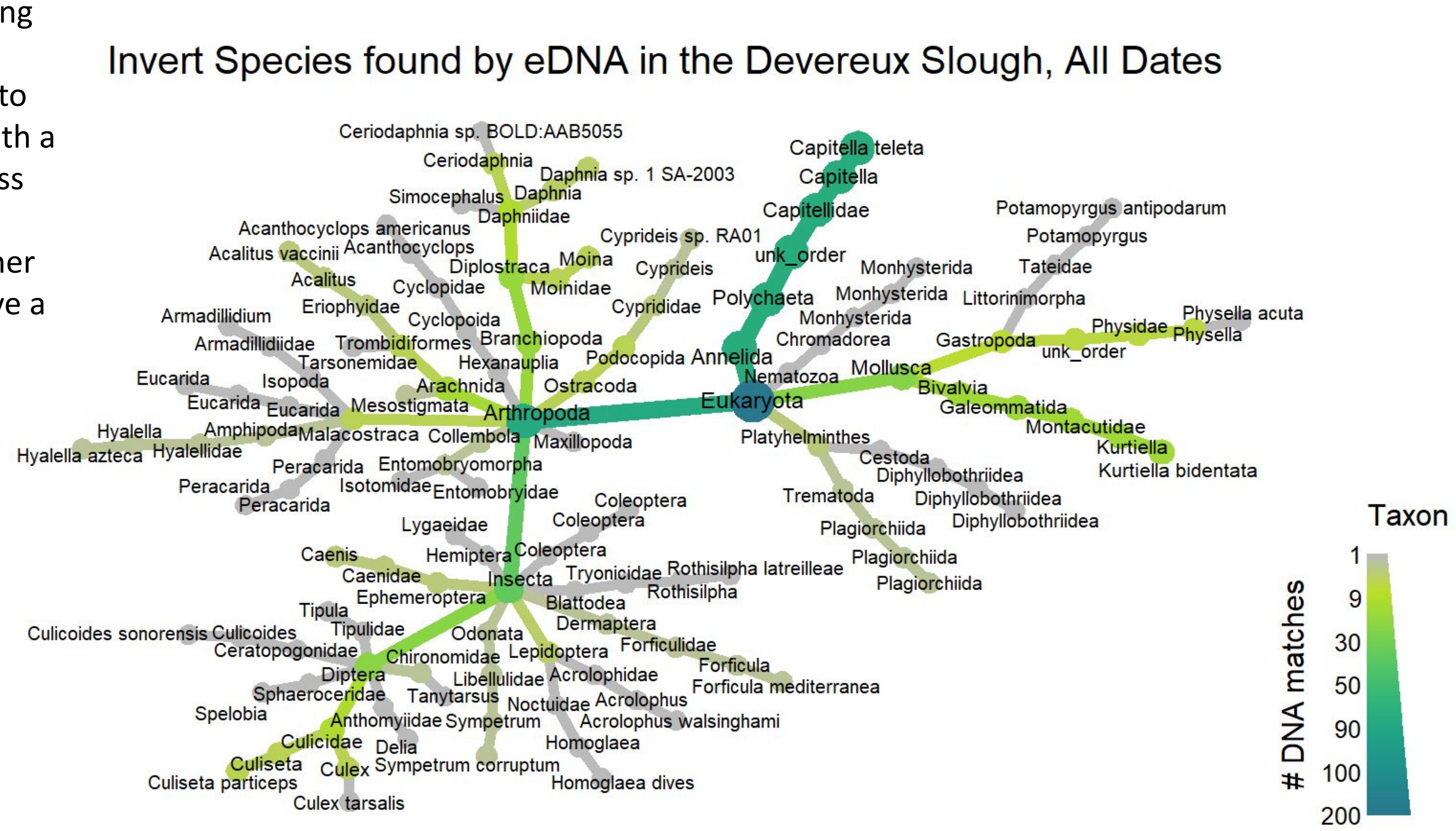


Figure 4. Phylogenetic tree of taxa detected in the Devereux Slough using aquatic environmental DNA.

Discussion

Figures 1 and 2 suggest that invertebrate assemblages differ between 2019 and 2022 in both NCOS and COPR. Assemblages from NCOS and COPR sites were separated into two different plots to account for the strong salinity difference between the reserves. Restoration at NCOS took place from 2017-2023; this is one factor that may account for yearly differences. Other factors that differ both between and during years include rainfall, salinity, and tidal influence. The distance between matching sites sampled during different seasons suggest that aquatic macroinvertebrate assemblages differ throughout the year. Further research is required to determine the role of restoration and/or seasonality on aquatic macroinvertebrate communities at NCOS and COPR.

Figure 3 shows that the number of invertebrates in the slough is not constant. The spike in organisms/liter observed in the winter is likely due to seasonal rains in December through February. The rains cause the slough to spill into the ocean at the mouth, and also increase the volume of input from Phelps and Devereux creeks. This decreases the salinity in most of the slough. Because many organisms (including ostracods as shown in figure 6) thrive in lower salinity, the spike in the number of organisms during the winter is likely connected to the more favorable winter water quality.

eDNA sampling identified many of the same organisms as manual invertebrate sampling, but did not provide consistent results over multiple rounds of sampling. eDNA was useful for increasing the taxonomic resolution of identified organisms.

A major limitation of our data is that 7.5 liters of water was used for samples in 2018/2019 while 70 liters was used for samples in 2021/2022. Organism counts were normalized by liters sampled, however the samples with less water volume may have detected fewer of the rarer species. In addition, there are some sampling event gaps in our data.

References

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Acknowledgements

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