Non-invasive biodiversity assessment of estuarine wetlands using environmental DNA

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Statement of work

Community biodiversity can be difficult to monitor in aquatic environments such as wetlands. Traditional monitoring methods such as trawls and seining can be destructive, ineffective, costly, or time-consuming. A pioneering method called environmental DNA (eDNA) uses trace amounts of DNA to non-invasively survey species via shed skin, mucus, and waste in water. eDNA can track changes in biodiversity, which leads to improved conservation strategies and novel research questions[1]. In this study, I will use eDNA to monitor fish and invertebrate communities in Suisun Marsh, a tidal marsh in the San Francisco Estuary (SFE).

Suisun Marsh is the largest contiguous tidal wetland on the US west coast[2]. Monitoring surveys (otter trawl and beach seine) are used to determine the effectiveness of conservation, restoration, and management actions, including operation of the Suisun Marsh Salinity Control Gate. Traditional sampling under-samples diverse communities[3]. eDNA may be a more sensitive method for assessing biodiversity compared with traditional sampling. Traditional identification requires taxonomic expertise, potentially limiting sample size and power of a study. Particular groups of interest for Suisun Marsh include native and invasive fishes, invasive clams, and crustaceans such as amphipods. eDNA can also be used to detect new and potentially harmful invasive species that are present in low densities. A better understanding of species distributions in space and time could help guide future restoration to support native species. While eDNA studies have been used in a range of aquatic habitats, eDNA has not yet been used to characterize fish and macroinvertebrates in the SFE.

Although no work has yet been completed for this study, it builds on my previous experience using eDNA sampling and high-throughput genetic sequencing (HTS) in the SFE. At UC Davis I have conducted eDNA studies of endangered delta smelt (*Hypomesus transpacificus*) in the SFE. The proposed study will build on the results of this previous work which focused on development of technical aspects of eDNA detection. In the proposed study, eDNA will be paired with HTS for community assessment. I used HTS to identify plankton and plankton food web interactions for my MS thesis research at San Francisco State University[4,5 6]. I prepared HTS sample libraries following Illumina MiSeq guidelines and conducted bioinformatic analysis using Python and R programming languages. Sample preparation and bioinformatic analysis for the proposed study will closely parallel the methods used for my MS research.

The purpose of the proposed study is to evaluate the accuracy and effectiveness of eDNA monitoring for fishes and macroinvertebrates in Suisun Marsh. Results of eDNA monitoring will be compared to results of the UC Davis Suisun Marsh Survey[7] in an effort to determine concordance between the two monitoring methods. In this study I will characterize spatial and temporal variability of eDNA detections.

Methods

I will sample eDNA in Suisun Marsh at sites paired with the trawl and seine sites of the UC Davis Suisun Marsh Survey[7] . eDNA sampling for spatial comparisons (Objective 1) will take place over one week. Samples (n=35 including negative controls) will be collected as 3 replicates x 2 sites x 5 microhabitats: (1) water surface; (2) 1 m below the surface; (3) just above the benthos; (4) amoung aquatic vegetation; and (5) as a sediment sample. In the temporal study (Objective 2), eDNA samples (n=80 including negative controls) will be collected monthly as 3 replicates x 4 sites x 6 months. With additional funding, I hope to extend the study to one year.

Environmental data (water temperature, conductivity, turbidity, pH, dissolved oxygen, and microhabitat type) will be recorded with each sample.

Samples will be barcoded using universal 16S rRNA primers[8]. Sequencing will follow Illumina meta-barcoding protocols (Illumina, Inc.) and will use an Illumina MiSeq (UC Davis Genome Center). Sequence data will be analyzed in R[9] and an occupancy model[10] will be used to calculate detection probabilities for indicator species. Species richness and Shannon-Weiner diversity will be measured at each site and time. Non-parametric multidimensional scaling will be used to visualize differences between sampling sites and through time.

Benefits to coastal wetlands

Loss of aquatic species has been linked to decreased ecosystem function[11], decreased primary production, and re-provisioning of ecosystem services[12]. Socioeconomic and cultural losses caused by decreased biodiversity also provides practical incentives for conservation. Climate change, altered flows, and invasive species are just a few of the threats to wetlands in the near future[13]. Biodiversity assessments provide the data needed for conservation, restoration, and management decision-making. eDNA shows promise as a sensitive and accurate method for assessing biodiversity and determining species distributions, especially of rare or invasive species[1,8,14]. Despite promise as a monitoring approach, dynamics of eDNA detection are still poorly understood[15]. This research addresses gaps in knowledge of eDNA application and interpretation such as how microhabitat and season fluence concordance with traditional survey results. eDNA methods will be valuable for future studies in the region as well as in other aquatic and wetland ecosystems. Going forward, eDNA sampling is expected to be an important complement to existing survey methods and to aid in decision-making for conservation, restoration, and management[1,8,14].

Budget overview

The Coastal Wetlands Scholarship would fund 6 months of fieldwork. Funding requested in this proposal would provide a paid conservation biology internship to an undergraduate student and fund field supplies. Unpaid science internships are common in conservation fieldwork but may be a barrier to entry, contributing to a lack of diversity in conservation science[16,17]. I have experience successfully mentoring an undergraduate in a paid internship[18]. The intern will assist with sample collection in the field, DNA extraction, and HTS sample preparation and data analysis. The student will also be able to develop an independent research project. I will consult with campus resources that support educationally or economically disadvantaged students to recruit a student for this project. This funding will support at least 2 quarters of undergraduate work up to 10 hours per week (approximately $3000). The remainder of the funds will be used for motorboat operator training ($600 at UC Davis Bodega Marine Laboratory), 12 days of truck rental to tow the jon boat ($708 at UC Davis Fleet Services), 12 days of kayak rental ($516 at UC Davis), and filters for sample collection ($310 for 200). Other equipment (e.g. such as a water pump and jon boat) is already available at UC Davis.

Plans for sharing results

The results of this study will be of immediate interest to resource managers and researchers in the San Francisco Estuary and other coastal wetlands and will be submitted for publication. I will present the results at a regional conference (e.g. Bay Delta Science Conference 2020 (Sacramento, CA) where I have free registration by virtue of winning 2018 Best Student Presentation) and seek funding to present at a national conference (e.g. Ecological Society of America or Society of Wetland Scientists). In addition, the results of this study may be integrated into my ongoing science outreach work including STEM workshops for girls in underserved

communities that I am organizing this year with the support of 500 Women Scientists and IGNITE.

Literature Cited

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