DNA microsatellite alleles from flowering shoots and seeds collected Curlew Beach in Nahant, MA and Niles Beach in Gloucester, MA in 2014

Website: https://www.bco-dmo.org/dataset/851773
Data Type: Other Field Results
Version: 1
Version Date: 2021-05-11

Project
» RUI: Collaborative Research: Trait differentiation and local adaptation to depth within meadows of the foundation seagrass Zostera marina (ZosMarLA)

Contributors	Affiliation	Role
Hays, Cynthia	Keene State College (Keene)	Principal Investigator, Contact
Hanley, Torrance C.	Northeastern University	Co-Principal Investigator
Hughes, A. Randall	Northeastern University	Co-Principal Investigator
Rauch, Shannon	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

Abstract
This dataset includes information on DNA microsatellite alleles from flowering shoots and seeds collected by SCUBA at Curlew Beach in Nahant, Massachusetts and Niles Beach in Gloucester, Massachusetts in 2014.

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Coverage

Spatial Extent: **N**:42.597 **E**:-70.655 **S**:42.42 **W**:-70.915
Temporal Extent: 2014-07-14 - 2014-08-03

Methods & Sampling

SCUBA was used to sample *Zostera marina* in late summer 2014 from two coastal eelgrass meadows in the Gulf of Maine, USA, separated by approximately 48 km: Curlew Beach in Nahant, MA (hereafter CB) and Niles Beach in Gloucester, MA (hereafter NI). Samples were collected from three depths at each site: the center of the meadow (mid), and approximately 5 m from the inshore and offshore edges (shallow and deep, respectively); depth of our shallow, mid and deep samples was approximately 1, 3 and 5 m MLLW, respectively. At each depth, 8-10 flowering shoots. separated by at least 2 meters, were collected; from each of these focal flowering shoots, a leaf tissue sample was harvested along with ~8-15 developing seeds from within a single spathe on the highest and youngest rhipidia, or cluster of inflorescences (n = ~250 seeds per site). Leaf tissue was preserved in silica or frozen until DNA extraction; seeds were frozen at -80C.

Molecular methods:
DNA was extracted from leaf tissue by grinding each sample with a Retsch mixer mill MM400 and using the Omega Bio-Tek E.Z.N.A.® Plant DNA Kit. DNA from seeds was extracted with Chelex 100 (Bio-Rad). Individual seeds were ground by hand in microfuge tubes with micropestles after removing the seed coat; samples were then incubated at 55°C for 8-24 h, gently mixed, heated to 98°C for 10 min, vortexed, centrifuged at 14,000 rpm for 5 min, and the supernatant stored at -20°C until PCR.

Each leaf sample was genotyped using 12 microsatellite loci developed for *Zostera marina*, multiplexed in three 11 ul PCR reactions. Each reaction consisted of 1 ul DNA template, 5 µl 2X Type-It multiplex master mix (Qiagen), and 0.25 ul of each 10 uM primer. PCR cycling conditions included initial activation/denaturation at 95°C for 5 min, followed by 28 cycles of 95°C for 30 sec, 60°C for 90 sec, and 72°C for 30 sec, and final extension at 60°C for 30 min. PCR products were separated on a 3730xl Genetic Analyzer (Applied Biosystems) at the Yale University DNA Analysis Facility, and fragment analysis was performed using GeneMarker version 2.6 (SoftGenetics). Nine of the 12 markers used for adult shoots proved useful for analyzing seed diversity. ZMC-12075 was eliminated due to a pattern of stutter around peaks that made scoring paternal alleles unreliable; two other markers (GA-3, CT-12) showed low allelic diversity in the adult populations and thus low power to discriminate among fathers.

Data Processing Description

BCO-DMO Processing:
- changed date format to YYYY-MM-DD.

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Data Files

File
seeds_DNA_microsatellites.csv (Comma Separated Values (.csv), 62.39 KB) MD5:57bdedd8cd723e32356ab2e83c56d142 Primary data file for dataset ID 851773

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Related Publications

Hays, C. G., Hanley, T. C., Graves, R. M., Schenck, F. R., & Hughes, A. R. (2020). Linking Spatial Patterns of Adult and Seed Diversity Across the Depth Gradient in the Seagrass Zostera marina L. Estuaries and Coasts, 44(2), 383–395. doi:10.1007/s12237-020-00813-1
Results

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Parameters

Parameter	Description	Units
date	Sampling date; format: YYYY-MM-DD	unitless
lat	Latitude of sampling site	degrees North
lon	Longitude of sampling site	degrees East

site	Site: CB = Curlew Beach; NI = Niles Beach.	unitless
type	mom or seed	unitless
family	unique identifier for the maternal family	unitless
quad	Quadrat number	unitless
sample_code	Unique identifier for the genetic sample	unitless
CT3_a	allele 1 for locus ZosmarCT-3	bp (base pairs)
CT3_b	allele 2 for locus ZosmarCT-3	bp (base pairs)
GA2_a	allele 1 for locus ZosmarGA-2	bp (base pairs)
GA2_b	allele 2 for locus ZosmarGA-2	bp (base pairs)
CT19_a	allele 1 for locus ZosmarCT-19	bp (base pairs)
CT19_b	allele 2 for locus ZosmarCT-19	bp (base pairs)
CL412_a	allele 1 for locus CL412Contig1	bp (base pairs)
CL412_b	allele 2 for locus CL412Contig1	bp (base pairs)
ZMC13053_a	allele 1 for locus ZMC13053	bp (base pairs)
ZMC13053_b	allele 2 for locus ZMC13053	bp (base pairs)
CL32_a	allele 1 for locus CL32Contig2	bp (base pairs)
CL32_b	allele 2 for locus CL32Contig2	bp (base pairs)
ZMC19017_a	allele 1 for locus ZMC19017	bp (base pairs)
ZMC19017_b	allele 2 for locus ZMC19017	bp (base pairs)

CL172_a	allele 1 for locus CL172Contig1	bp (base pairs)
CL172_b	allele 2 for locus CL172Contig1	bp (base pairs)
GA35_a	allele 1 for locus ZosmarGA-35	bp (base pairs)
GA35_b	allele 2 for locus ZosmarGA-35	bp (base pairs)

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Instruments

Dataset-specific Instrument Name	3730xl Genetic Analyzer (Applied Biosystems)
Generic Instrument Name	Automated DNA Sequencer
Generic Instrument Description	A DNA sequencer is an instrument that determines the order of deoxynucleotides in deoxyribonucleic acid sequences.

Dataset-specific Instrument Name	SCUBA
Generic Instrument Name	Self-Contained Underwater Breathing Apparatus
Generic Instrument Description	The self-contained underwater breathing apparatus or scuba diving system is the result of technological developments and innovations that began almost 300 years ago. Scuba diving is the most extensively used system for breathing underwater by recreational divers throughout the world and in various forms is also widely used to perform underwater work for military, scientific, and commercial purposes. Reference: https://oceanexplorer.noaa.gov/technology/technical/technical.html

Dataset-specific Instrument Name	
Generic Instrument Name	Thermal Cycler
Generic Instrument Description	A thermal cycler or "thermocycler" is a general term for a type of laboratory apparatus, commonly used for performing polymerase chain reaction (PCR), that is capable of repeatedly altering and maintaining specific temperatures for defined periods of time. The device has a thermal block with holes where tubes with the PCR reaction mixtures can be inserted. The cycler then raises and lowers the temperature of the block in discrete, pre-programmed steps. They can also be used to facilitate other temperature-sensitive reactions, including restriction enzyme digestion or rapid diagnostics. (adapted from http://serc.carleton.edu/microbelife/research_methods/genomics/pcr.html)

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Project Information

RUI: Collaborative Research: Trait differentiation and local adaptation to depth within meadows of the foundation seagrass Zostera marina (ZosMarLA)

Coverage: Massachusetts, USA

NSF Award Abstract:
Understanding how species cope with spatial variation in their environment (e.g. gradients in light and temperature) is necessary for informed management as well as for predicting how they may respond to change. This project will examine how key traits vary with depth in common eelgrass (Zostera marina), one of the most important foundation species in temperate nearshore ecosystems worldwide. The investigators will use a combination of experiments in the field and lab, paired with fine-scale molecular analyses, to determine the genetic and environmental components of seagrass trait variation. This work will provide important information on the microevolutionary mechanisms that allow a foundation species to persist in a variable environment, and thus to drive the ecological function of whole nearshore communities. The Northeastern University graduate and Keene State College (KSC) undergraduate students supported by this project will receive training in state-of-the-art molecular techniques, as well as mentorship and experience in scientific communication and outreach. A significant portion of KSC students are from groups under-represented in science. Key findings of the research will be incorporated into undergraduate courses and outreach programs for high school students from under-represented groups, and presented at local and national meetings of scientists and stakeholders.

Local adaptation, the superior performance of "home" versus "foreign" genotypes in a local environment, is a powerful demonstration of how natural selection can overcome gene flow and drift to shape phenotypes to match their environment. The classic test for local adaptation is a reciprocal transplant. However, such experiments often fail to capture critical aspects of the immigration process that may mediate realized gene flow in natural systems. For example, reciprocal transplant experiments typically test local and non-local phenotypes at the same (often adult) life history stage, and at the same abundance or density, which does not mirror how dispersal actually occurs for most species. In real populations, migrants (non-local) often arrive at low numbers compared to residents (local), and relative frequency itself can impact fitness. In particular, rare phenotypes may experience reduced competition for resources, or relative release from specialized pathogens. Such negative frequency dependent selection can reduce fitness differences between migrants and residents due to local adaptation, and magnify effective gene flow, thus maintaining greater within-population genetic diversity. The investigators will combine spatially paired sampling and fine-scale molecular analyses to link seed/seedling trait variation across the depth gradient at six meadows to key factors that may drive these patterns: local environmental conditions, population demography, and gene flow across depths. The team will then experimentally test the outcome of cross-gradient dispersal in an ecologically relevant context, by reciprocally out-planting seeds from different depths and manipulating relative frequency in relation to both adults and other seedling lineages. The possible interaction between local adaptation and frequency-dependence is particularly relevant for Zostera marina, which represents one of the best documented examples of the ecological effects of genetic diversity and identity. Further, a better understanding of seagrass trait differentiation is not simply a matter of academic interest, but critical to successful seagrass restoration and conservation.

This award reflects NSF's statutory mission and has been deemed worthy of support through evaluation using the Foundation's intellectual merit and broader impacts review criteria.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1851432
NSF Division of Ocean Sciences (NSF OCE)	OCE-1851043

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