DNA sequence data deposited on GenBank for polychaetes used in the project studying the evolution and mechanics of burrowing in La Jolla, CA during 2011 (Burrowing polychaete mechanics project)

Website: https://www.bco-dmo.org/dataset/555354
Version: 2015-04-03

Project
» Functional diversity of infaunal burrowers: Towards a mechanistic understanding of animal-sediment interactions (Burrowing polychaete mechanics)

Contributors	Affiliation	Role
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Dataset Description

Related References:

Law, C, KM Dorgan, GW Rouse. 2014. Relating divergence in musculature within Opheliidae (Annelida) with different burrowing behaviors. Journal of Morphology 275:548-571.

Law, CJ, KM Dorgan, GW Rouse. 2013. Validation of three sympatric Thoracophelia species (Annelida: Opheliidae) from Dillon Beach, California using mitochondrial and nuclear DNA sequence data. Zootaxa 3608:67–74.

Methods & Sampling

Specimens were collected by hand and sorted or sieved from sediments and preserved for DNA and morphology analysis. Armandia brevis (Moore, 1906) and Thoracophelia mucronata (Treadwell, 1914) were collected from Mission Bay, San Diego, California on June 9, 2011 and La Jolla Shores Beach, California on May 4, 2012, respectively.

Data Processing Description

DNA Amplification and Sequencing:
Forty one specimens of 33 species were used for phylogenetic analyses: 25 opheliids, four polygordiids, and 10 scalibregmatids. The two outgroup taxa, a capitellid Notomastus sp. and an arenicolid A. marina were chosen based on Struck et al.'s (2011) annelid phylogeny (Table 1) with Notomastus sp. being used as the root terminal. Newly collected specimens [from Beaufort, NC (Ophelina sp1.); Costa Rica (Ophelina sp3.);

Friday Harbor, WA (Notomastus sp., O. acuminata, Polygordius sp., and Scalibregma inflatum); Greenland (O. acuminata, O. cylindricaudata, and O. limacina); La Jolla, CA (A. brevis, Polyophthalmus sp., and T. mucronata); Lizard Island, Australia (Armandia sp1.); and off the Oregon coast (Ophelina sp2.)] were relaxed in 7.5% MgCl2 and fixed in 95% ethyl alcohol. Sequences for the remaining 26 species were accessed through GenBank (Table 1).

A Qiagen DNeasy tissue kit was used to extract genomic DNA from specimens according to the manufacturer's instructions. Approximately 500 base pairs of the mitochondrial small subunit ribosomal DNA (16S) were amplified using the primers 16SarL and 16SbrL (Palumbi, 1996) with temperature profiles of 95C for 3 min, followed by 40 cycles of 95C for 40 s, 48C for 40 s, 68C for 50 s, and final extension at 68C for 5 min (see Supporting Information, Table S1).

Three nuclear loci were also sequenced. The small subunit ribosomal DNA (18S) was amplified using three primer sets: 1) 1F and 5R; 2) 3F and bi; and 3) a2.0 and 9R (Giribet et al., 1996, 1999). Temperature profiles for the 1F/5R and a2.0/9R primer sets were 95C for 3 min, followed by 40 cycles of 95C for 30 s, 52C for 30 s, 72C for 90 s, and final extension at 72C for 8 min. The temperature profile for the 3F/bi primer set was 95C for 3 min, followed by 40 cycles of 95C for 30 s, 49C for 30 s, 72C for 90 s, and final extension at 72C for 8 min. Approximately 930 base pairs of the large subunit ribosomal DNA (28S) were amplified using the primers Po28F1 and Po28R4 (Struck et al., 2006), and ~360 base pairs of the nuclear protein coding gene Histone H3 were amplified using the primers H3aF and H3aR (Colgan et al., 1998). Both genes were amplified using the same temperature profiles of 94C for 2 min, followed by 35 cycles of 94C for 45 s, 48C for 60 s, 72C for 90 s, and final extension at 72C for 10 min.

Amplification reactions (25 ml) were conducted containing 2 ml of DNA template, 1 ml of forward and reverse primers, 12.5 ml GoTaq Green Master Mix (Promega), and 8.5 ml H2O. ExoSAP-IT (Affymetrix) was used to purify PCR products. Sequencing was done by either Retrogen (San Diego, CA) or Eurofins MWG Operon (Louisville, KY). Sequences were edited using Geneious 5.5.6 (www.geneious.com) and aligned with MAFFT 3.8 (Katoh and Kuma, 2002) under default settings with no manual alterations. The combined molecular dataset consisted of 3,955 total characters, 1,075 of which were parsimony informative and 436 were uninformative.

Phylogenetic Analysis:
Parsimony analyses on the combined genes (16S, 18S, 28S, and H3) were conducted in PAUP* 4.0b10 (Swofford, 2002) using a heuristic search with random stepwise addition of the terminals for 1,000 replicates, with tree bisection and reconnection. The character matrix was equally weighted, and gaps were treated as missing data. Clade support was assessed using jackknifing with 37% deletion of sites over 1,000 replicates with 10 random additions per iteration. Maximum likelihood analyses were performed in RAxML 7.2.8 (Stamatkis, 2006) as a four-gene partitioned dataset and under the General Time Reversible1Gamma (GTR1G) model. Bootstrap (thorough option) values were estimated using 100 pseudoreplicates under the same model.

BCO-DMO Processing:

- added conventional header with dataset name, PI name, version date, reference information
- renamed parameters to BCO-DMO standard
- replaced blanks with NA
- transformed gene columns to rows
- added html links to GenBank accession pages

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Data Files

File	
polych_accnums.csv	(Comma Separated Values (.csv), 24.70 KB) MD5:0d3fc761a3b5483da9b30eb12225bd3e
Primary data file for dataset ID 555354	

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Parameters

Parameter	Description	Units
taxon	polychaete family	unitless
species	taxonomic genus and species name with authority	unitless
specimen_origin	location of specimen collection	unitless
lat	latitude; north is postitive	decimal degrees
lon	longitude; east is postitive	decimal degrees
gene	Type of gene sequenced: voucher_SIO=SIO voucher number rRNA_16S=GenBank accession number for 16S ribosomal RNA gene rRNA_18S=GenBank accession number for 18S ribosomal RNA gene rRNA_28S=GenBank accession number for 28S ribosomal RNA gene Histone_H3=GenBank accession number for; histone 3 gene ITS=GenBank accession number for 18S ribosomal RNA gene; internal transcribed spacer mtCOI=GenBank accession number for cytochrome oxidase subunit 1 gene	unitless
accession_number	GenBank accession number	unitless

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Instruments

Dataset-specific Instrument Name	DNA Sequencer
Generic Instrument Name	Automated DNA Sequencer
Dataset-specific Description	Sequencing was done by either Retrogen (San Diego, CA) or Eurofins MWG Operon (Louisville, KY).
Generic Instrument Description	General term for a laboratory instrument used for deciphering the order of bases in a strand of DNA. Sanger sequencers detect fluorescence from different dyes that are used to identify the A, C, G, and T extension reactions. Contemporary or Pyrosequencer methods are based on detecting the activity of DNA polymerase (a DNA synthesizing enzyme) with another chemoluminescent enzyme. Essentially, the method allows sequencing of a single strand of DNA by synthesizing the complementary strand along it, one base pair at a time, and detecting which base was actually added at each step.

Dataset-specific Instrument Name	camera
Generic Instrument Name	Camera
Dataset-specific Description	Canon Powershot G9 camera attached to a Leica DMR microscope; Canon T1i camera attached to an Olympus CX41 microscope
Generic Instrument Description	All types of photographic equipment including stills, video, film and digital systems.

Dataset-specific Instrument Name	microscope
Generic Instrument Name	Microscope - Optical
Dataset-specific Description	Olympus CX41 compound microscope; Leica DMR microscope; Zeiss AxioObserver Z1 microscope with DIC filters and AxioVision software
Generic Instrument Description	Instruments that generate enlarged images of samples using the phenomena of reflection and absorption of visible light. Includes conventional and inverted instruments. Also called a "light microscope".

Dataset-specific Instrument Name	Thermal Cycler
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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Deployments

Rouse_2011

Website	https://www.bco-dmo.org/deployment/555360
Platform	SIO_Rouse
Start Date	2011-06-09
End Date	2012-05-04
Description	polychaete studies

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Project Information

Functional diversity of infaunal burrowers: Towards a mechanistic understanding of animal-sediment interactions (Burrowing polychaete mechanics)

Coverage: California

Description from NSF award abstract:

Benthic communities comprise diverse and abundant organisms with important ecological and biogeochemical roles. They convert organic carbon into biomass that is transferred to higher trophic levels, regenerate nutrients, and determine the fate of pollutants and organic carbon buried in sediments. In many coastal environments, anthropogenic stresses, including eutrophication and resulting hypoxia, trawling and disturbance from fisheries, and pollutants have negative and often dramatic affects on species diversity. Assessing the ecological and biogeochemical impacts of changes in species diversity is nearly impossible, however, without understanding the functional roles of the species. In sedimentary environments, determining functionality is especially important for organisms closely associated sediments, such as infaunal deposit feeders that ingest sediments while living in and moving through them.

Burrowing behaviors and morphologies have been examined for individual species, but decades have passed since even broad burrowing behaviors were compared across diverse taxa. Moreover, such comparisons largely ignored the mechanical response of sediments, an omission similar to studying swimming without considering fluid mechanics. Since that time, there have been several major advances in the physics of animal-sediment interactions. Muddy sediments are elastic solids through which burrows are extended by fracture. In contrast, sands are granular materials whose mechanics are governed by gravitational forces acting on individual grains, rather than by adhesion and cohesion of the mucopolymeric matrix dominating mud mechanics. Use of gelatin as a clear analog for muds has enabled visualization of burrowing and analyses of forces and kinematics. This research will combine structural and anatomical studies and kinematic analyses of burrowing in gelatin and sand analogs with mechanical testing and numerical modeling of real sediments. Linkages would be made among anatomies, morphologies, and behaviors to burrowing function in sands versus muds. Polychaetous annelids, a diverse and abundant component of benthic communities, will be the focal taxon.

Functional groupings of burrowing infauna have been based on morphologies and trophic roles but advances in sediment mechanics suggest that similar morphologies may have different functions in sands versus muds (e.g., expansible structures extend cracks in muds but are anchors in sands). In addition, seemingly different morphologies may have analogous functions (e.g., the pharynx of Nereis virens and the muscular anterior of the cirratulid Cirriformia moorei both exert dorso-ventral stress to extend burrows by fracture). Linking functions to morphologies and behaviors of burrowers is important in understanding functional roles of infauna and resulting functional diversity of benthic communities. The diversity of burrowing mechanisms revealed in this study will enable generalizations about burrowing mechanics in different environments. Important characteristics of burrowing locomotion will be identified as those shared by diverse burrowers. How the different physical constraints of sand and mud specify burrowing mechanics and affect morphologies and behaviors of burrowers will be contrasted for closely related taxa from different environments.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1029160

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