# Common garden experimental data on temperature effects in offspring of four Odontesthes regia populations collected along the Chilean coast from Sep to Oct 2023

Website: https://www.bco-dmo.org/dataset/956677

Data Type: Other Field Results

Version: 1

Version Date: 2025-04-07

## **Project**

» Expanding the silverside system to quantify how climate gradients determine co- and countergradient adaptation strength in the ocean (Chilean silversides)

Contributors	Affiliation	Role
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#### Abstract

To study patterns of local adaptation to latitudinal temperature gradients in the ocean, we conducted the first common garden experiment on the Chilean silverside (Odontesthes regia). Wild adult spawners from four locations (20 - 42°S) along the Chilean coast were used to produce offspring that was then reared in triplicates from fertilization to approximately 35 mm total lengths (TL) under a set of four common temperatures in the laboratory (11-23°C) and ad libitum feeding conditions. Larval and juvenile TL were measured via calibrated digital pictures (intermediate samples) or via calipers (final samples). Subsequently, the data will allow calculating population, temperature, and life-stage-specific growth capacities (i.e., TL growth rates at excess feeding conditions). This will reveal whether higher latitude populations have evolved genetically faster growth capacities than lower latitude populations - as has so far been shown only for northern hemisphere fishes. In addition, adults collected from each location were x-rayed to count the vertebrae of each specimen and subsequently test, whether lower latitude populations have fewer vertebrae on average than their higher latitude conspecifics - a phenomenon known as Jordan's Rule. This dataset includes measurements of total length (TL) and wet weight (wW), developmental stage (Sample), individual ID, and key dates including fertilization, main hatching, and sampling. It also contains population information (Species, Population, PopLabel), collection site metadata (Longitude, Latitude), and specific conditions of the common garden experiment, such as temperature treatment (Temp), replicate number, and bucket ID. The supplemental tables provide additional data including adult total length and collection metadata (Table 1), vertebral counts from xray images and associated metadata (Table 2), and high-resolution temperature records from each experimental treatment tank throughout the common garden experiment (Table 3).

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**Location**: Chilean coastal waters between Iquique

**Spatial Extent**: N:-20.27333333 **E**:-70.14638889 **S**:-41.50444444 W:-72.9375

**Temporal Extent**: 2023-09-23 - 2023-12-25

## Methods & Sampling

## Field sampling of spawners, offspring production, transport

At each of four locations along the Chilean coast (north to south: Iquique [IQ, 20°S], Caleta Sierra [CS, 31°S], Dichato [DI, 37°S], Puerto Montt [PM, 42°S]), flowing-ripe adults of the Chilean silverside (*Odontesthes regia*) (urn:lsid:marinespecies.org:taxname:281830) were obtained during the species' spawning season in austral spring 2023 (September-October). With the help of local artisanal fishermen, adults were caught during night or early morning hours with gill nets of various lengths (2 cm stretched mesh). Fish were separated by sex and kept alive for up to one hour, before they were strip-spawned into plastic dishes filled with clean seawater. Our target was to use at least 20 spawners per sex and population, but the realized number of spawners varied. The northernmost site of Iquique was sampled twice; we denoted this in the data as IQ1 and IQ2. Fertilized embryos were transported in cooler boxes or thermos containers to the rearing facility in Dichato via car (PM, DI, CS) or airplane (IQ). Upon arrival at the rearing facility no more than 24h post-fertilization, they were randomly distributed among temperature treatments and replicate rearing containers.

# Common garden experiment

Offspring were reared at the INCAR (Centro Interdisciplinario para la Investigación Acuícola) facility of the Dichato Marine Research Station of the Universidad de Concepción. We used four 1200L recirculating tanks representing the four temperature treatments (11, 14, 18, 23°C). Each tank housed 12 rearing containers that represented the 3 replicates for each of the 4 populations (4 temperatures  $\times$  4 populations  $\times$  3 replicates = 48 containers). The container design was identical to previous silverside experiments (e.g., Baumann & Conover 2011, Murray et al. 2014), i.e., we used round, white 20L containers equipped with individual airlines and mesh-screened (150  $\mu$ m) holes, which guaranteed  $\sim$ 100% oxygenation and water exchange with the surrounding tank while retaining food. Tank water of  $\sim$  30 psu was drawn from Coliumo Bay, filtered/UV-sterilized, and controlled for temperature via thermostats connected to commercial aquarium heaters or chillers. Each tank was equipped with a HOBO® Pendant MX temperature logger (Onset) that recorded conditions every 30 minutes. For biofiltration we used 4 large FX4-250 (Fluval) canister filters. The photoperiod was 15h light: 9h dark throughout the entire experiment.

Starting at hatch, silverside larvae were fed *ad libitum* rations of newly hatched brine shrimp nauplii ( $Artemia\ salina$ , San Francisco strain, from Brine Shrimp Direct) produced daily on site.  $Ad\ libitum$  rations were ensured by never allowing rearing containers to become nauplii depleted. A few days after successful first feeding, the number of fish in each rearing container was equalized to 130 avoid density-dependent growth effects. Another standardization to n=80 per container occurred during when larvae reached approximately 15 mm total length (TL), and a final standardization to n=40 occurred when larvae reach approximately 25 mm TL. When larvae reached approximately 35 mm TL, the rearing concluded and all survivors were sampled.

## **Samples**

All collected adults – both spawners and non-spawners - were measured for TL and then preserved frozen (-20°C) for later vertebral number analysis. Experimental samples were first taken on the temperature- and population-specific day of hatch, when up to 200 hatchlings were preserved in 95% ethanol (0-hatch). The first and second larval samples were taken at approximately 15 mm TL (1-15mm) and 25 mm TL (2-25mm) and preserved in 95% ethanol as well. All larvae from these intermediate samples were measured for TL using the open-source software ImageJ (version 1.53a) and digital pictures taken with an I-Phone 11 in front of a gridded (5mm grid size) white background. The 95% ethanol was replaced once, at 24h after initial fixation. The final sample was taken at approximately 35 mm TL (3-35mm). All survivors were measured for TL using calipers (nearest 0.1 mm) and blotted wet weight (Mettler-Toledo; nearest mg) and then preserved frozen (-20°C) in individual plastic bags.

#### Vertebral number

The number of vertebrae was determined for all adult fish, regardless of whether they were used to produce offspring or not. Fish were X-rayed at the Oceanside Animal Hospital in Sandwich, MA, using a Vet Ray veterinarian X-ray system (SEDECAL, model A6504-25) and settings that were slightly modified from those recommended for small exotic pets (lizards). Specifically, we used a voltage of 60 kVp, power of 320 mA, and a shutter speed of 16 milliseconds. Digital x-ray pictures were analyzed using the multipoint tool in ImageJ to

mark each vertebra between but excluding the basiocciptal and the urostyle.

## **Data Processing Description**

We removed stunted or malformed fish from the dataset via low-outlier analysis. For each population (IQ, CS, DI, PM), temperature (11,14,18,23°C), replicate (1, 2, 3), and sample type (hatch, 1-15mm, 2-25mm, 3-35mm), we calculated the mean and standard deviation, then removed all specimens that were smaller than the mean –  $2 \times SD$  (outlier < mean TL – 2\*SD TL).

## **BCO-DMO Processing Description**

- Imported NSF\_OCE 2313288 BCO-DMO source data.xlsx, sheet indexes 5, 4, 3, 2, and 1, into the BCO-DMO system
- Converted all dates to YYYY-MM-DD ISO format
- Converted lat and lon to decimal degrees format
- Rounded lat and lon values to 4 decimal places
- Renamed fields to remove spaces and units from parameters to comply with BCO-DMO system and style requirements
- Removed Month, Day, and Year from sheet 4, to avoid duplication
- Exported files as 956677\_v1\_common\_garden\_exp.csv (main file), adult\_total\_length.csv (supplemental file), adult\_vertebral\_number.csv (supplemental file), and experimental\_temperatures.csv (supplemental file)

Accepted species identifier confirmed on 2025-04-05.

## **Problem Description**

Poor fertilization success for PM and insufficient spawners for CS required reduced replication

The malfunction of chillers on 8 November 2023 and 12 December 2023 required the premature termination of the 11 and 14C treatments, respectively. Hence, final samples are not available for all populations for these temperatures.

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# **Data Files**

**File** 

956677\_v1\_common\_garden\_exp.csv(Comma Separated Values (.csv), 298.38 KB)
MD5:5b7ab00ccbebd8782f13f297668cdfd5

Primary data file for dataset ID 956677, version 1

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# **Supplemental Files**

#### File

# adult\_total\_length.csv

(Comma Separated Values (.csv), 17.88 KB) MD5:9a8a6cf02defc1c2d7bdd105ff6f9c14

Primary data file for dataset ID 956677, version 1

Table 1: includes Adult Total Length and Collection metadata

Species: All data in this table are for the Chilean silverside Odontesthes regia

Population: Sampling location, or population (Iquique, Caleta Sierra, Dichato, or Puerto Montt)

Longitude: Longitude of the sampling location, degrees decimal, West is negative Latitude: Latitude of the sampling location, degrees decimal, South is negative

Sampling date: Date of the field sampling of the adults, YYYY-MM-DD

Pop fish nr: Adult ID number by population

Sex: Sex of the fish: male, female, or not determined

Total length (cm): Total length of each fish determined via calibrated pictures

## adult vertebral number.csv

(Comma Separated Values (.csv), 16.89 KB) MD5:13e8122f2b3f597284b3ac1bd7bee505

Primary data file for dataset ID 956677, version 1

Table 2: includes Adult vertebral number and Collection metadata

Species: All data in this table are for the Chilean silverside Odontesthes regia

Population: Sampling location, or population (Iquique, Caleta Sierra, Dichato, or Puerto Montt)

Longitude: Longitude of the sampling location, degrees decimal, West is negative Latitude: Latitude of the sampling location, degrees decimal, South is negative

Sampling date: Date of the field sampling of the adults, YYYY-MM-DD Pop fish ID: Adult ID number by population (different from Table 1)

Sex: Sex of the fish: male, female, or not determined

Vertebral number: Number of vertebrae determined from digitial x-ray pictures

## experimental\_temperatures.csv

(Comma Separated Values (.csv), 346.51 KB) MD5:a86ca8778361f0c651adeb478cbe89e6

Primary data file for dataset ID 956677, version 1

Table 3: includes Experimental Temperatures

Treatment: Target temperature treatment of the experiment: 11, 14, 18, or 23C Temp: Actual temperature measured by Onset HOBO loggers in each of 4 tanks

ISO DateTime Local: Date and time of measurement, 30 minute intervals, using local Chile Summer Time CLST (UTC -3h)

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## **Related Publications**

Baumann, H., & Conover, D. O. (2011). Adaptation to climate change: contrasting patterns of thermal-reaction-norm evolution in Pacific versus Atlantic silversides. Proceedings of the Royal Society B: Biological Sciences, 278(1716), 2265–2273. https://doi.org/10.1098/rspb.2010.2479

Methods

Murray, C., Malvezzi, A., Gobler, C., & Baumann, H. (2014). Offspring sensitivity to ocean acidification changes seasonally in a coastal marine fish. Marine Ecology Progress Series, 504, 1–11. doi: 10.3354/meps10791

Methods

Schneider, C. A., Rasband, W. S., & Eliceiri, K. W. (2012). NIH Image to ImageJ: 25 years of image analysis. Nature Methods, 9(7), 671–675. https://doi.org/10.1038/nmeth.2089
Software

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## **Parameters**

Parameter	Description	Units
Species	All data in this table are for the Chilean silverside Odontesthes regia	unitless
Population	Sampling location or 'population' along the Chilean coast: Iquique, Caleta Sierra, Dichato, Puerto Montt	unitless
PopLabel	Abbreviated population label (IQ1 and IQ2 are used to describe two sampling events at the same location, see methods)	unitless
Longitude	Longitude of the sampling location, negative is West	decimal degrees
Latitude	Latitude of the sampling location, negative is South	decimal degrees
Temp	Target temperature treatment of the experiment: 11, 14, 18, or 23C	degrees Celsius
Replicate	ID of each replicate rearing container per population and temperature treatment	unitless
BucketID	Running ID of each rearing container	unitless
Fertilization_date	Date of offspring fertilization in the wild (adult Sampling date in supplemental tables "adult_total_length" and "adult_vertebral_number")	unitless
mainhatchday	Date when most offspring hatched	unitless
sampleday	Date when offspring where sampled and measured for total length	unitless
Sample	Sample stage: 0 - hatch, 1 -larvae 15 mm, 2 - juvenile 25 mm, 3 - final sample 35 mm	unitless
ID	Offspring ID per population, temperature, replicate, and sample	unitless
TL	Individual total length measured via digital, calibrated pictures	millimeter (mm)
wW	Individual blotted wet weight (final samples only)	milligram (mg)

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# Instruments

Dataset- specific Instrument Name	calipers
Generic Instrument Name	calipers
Dataset- specific Description	All survivors were measured for TL using calipers (nearest 0.1 mm) and blotted wet weight (Mettler-Toledo; nearest mg) and then preserved frozen (-20°C) in individual plastic bags.
Generic Instrument Description	A caliper (or "pair of calipers") is a device used to measure the distance between two opposite sides of an object. Many types of calipers permit reading out a measurement on a ruled scale, a dial, or a digital display.

Dataset- specific Instrument Name	iPhone 11
Generic Instrument Name	Camera
Dataset- specific Description	All larvae from these intermediate samples were measured for TL using the open-source software ImageJ and digital pictures taken with an I-Phone 11 in front of a gridded (5mm grid size) white background.
Generic Instrument Description	All types of photographic equipment including stills, video, film and digital systems.

Dataset- specific Instrument Name	1200L recirculating tank
Generic Instrument Name	circulating water bath
Dataset- specific Description	We used four 1200L recirculating tanks representing the four temperature treatments (11, 14, 18, 23°C). Each tank housed 12 rearing containers that represented the 3 replicates for each of the 4 populations (4 temperatures $\times$ 4 populations $\times$ 3 replicates = 48 containers). The container design was identical to previous silverside experiments (e.g., Baumann & Conover 2011, Murray et al. 2014), i.e., we used round, white 20L containers equipped with individual airlines and mesh-screened (150 $\mu$ m) holes, which guaranteed $\sim$ 100% oxygenation and water exchange with the surrounding tank while retaining food. Tank water of $\sim$ 30 psu was drawn from Coliumo Bay, filtered/UV-sterilized, and controlled for temperature via thermostats connected to commercial aquarium heaters or chillers.
Generic Instrument Description	A device designed to regulate the temperature of a vessel by bathing it in water held at the desired temperature. [Definition Source: NCI]

Dataset- specific Instrument Name	gill nets
Generic Instrument Name	Gillnet
Dataset- specific Description	With the help of local artisanal fishermen, adults were caught during night or early morning hours with gill nets of various lengths (2 cm stretched mesh).
Generic Instrument Description	Gillnetting uses curtains of netting that are suspended by a system of floats and weights; they can be anchored to the sea floor or allowed to float at the surface. A gillnet catches fish by their gills because the twine of the netting is very thin, and either the fish does not see the net or the net is set so that it traps the fish.

Dataset- specific Instrument Name	HOBO® Pendant MX temperature logger
Generic Instrument Name	Onset HOBO Pendant MX2201 temperature logger
Dataset- specific Description	Each tank was equipped with a HOBO® Pendant MX temperature logger (Onset) that recorded conditions every 30 minutes.
Generic Instrument Description	The Onset HOBO MX2201 is an in-situ instrument for wet or underwater applications. It supports soil temperature, temperature, and water temperature. A one-channel logger that records up to approximately 96,000 measurements or internal logger events with 8K bytes memory. It has a polypropylene housing case. Uses Bluetooth to transmit data. Can be used with a solar radiation shield. Measurement range: -20 deg C to 70 deg C. Accuracy: +/- 0.50 deg C from 0 deg C to 50 deg C. Water depth rating: 30.5 m

Dataset-specific Instrument Name	Mettler-Toledo
<b>Generic Instrument Name</b>	scale or balance
Dataset-specific Description	All survivors were measured for blotted wet weight (nearest mg) using a Mettler-Toledo scale.
Generic Instrument Description	Devices that determine the mass or weight of a sample.

Dataset- specific Instrument Name	SEDECAL, model A6504-25
Generic Instrument Name	X-Ray Imaging
Dataset- specific Description	Fish were X-rayed at the Oceanside Animal Hospital in Sandwich, MA, using a Vet Ray veterinarian X-ray system (SEDECAL, model A6504-25) and settings that were slightly modified from those recommended for small exotic pets (lizards).
Generic Instrument Description	A radiographic procedure using the emission of x-rays to form an image of the structure penetrated by the radiation.

# **Project Information**

Expanding the silverside system to quantify how climate gradients determine co- and countergradient adaptation strength in the ocean (Chilean silversides)

Website: https://befel.marinesciences.uconn.edu/expanding-the-silverside-system

Coverage: laboratory-based

#### NSF Award Abstract:

Many species have evolved adaptations to latitudinal climate gradients and studying these sheds light on how species will evolve in response to global climate change. To investigate adaptation in Chilean silversides, offspring of wild fish from four locations along the Chilean coast are being reared at four common temperatures. Differences in growth rates, vertebral number, and mercury uptake among populations from different latitudes indicate genetic differences due to local adaptation. The research team is integrating these Chilean silverside data with prior data from northern hemisphere silverside species to better understand the relationship between climate gradient strength and adaptation strength. This project provides training for two graduate students through a US-Chile graduate cross-cultural exchange. The research is being integrated into graduate and undergraduate courses taught in Chile and the research team is sharing the results with the public through a website and magazine articles.

This project advances the understanding of two forms of local adaptation, co-gradient variation (CoGV) and counter-gradient variation (CnGV), which underlie adaptation to large-scale, latitudinal climate gradients. Using a common garden experiment, the team is examining the relationship between temperature and genetic variation in Chilean silversides. Newly-fertilized offspring obtained from wild founders at four locations along the Chilean coast are being reared at four common temperatures to a common juvenile size. Differences among populations in trait measurements (growth capacity, vertebral number, and total mercury concentration) to test the hypotheses that 1) growth capacity increases with temperature and latitude (CnGV) and 2) vertebral number increases with latitude (CoGV). These data are being integrated with existing evidence from northern hemisphere silverside species to determine if there is a relationship between CnGV/CoGV strength and latitudinal gradient strength.

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# **Funding**

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-2313288

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