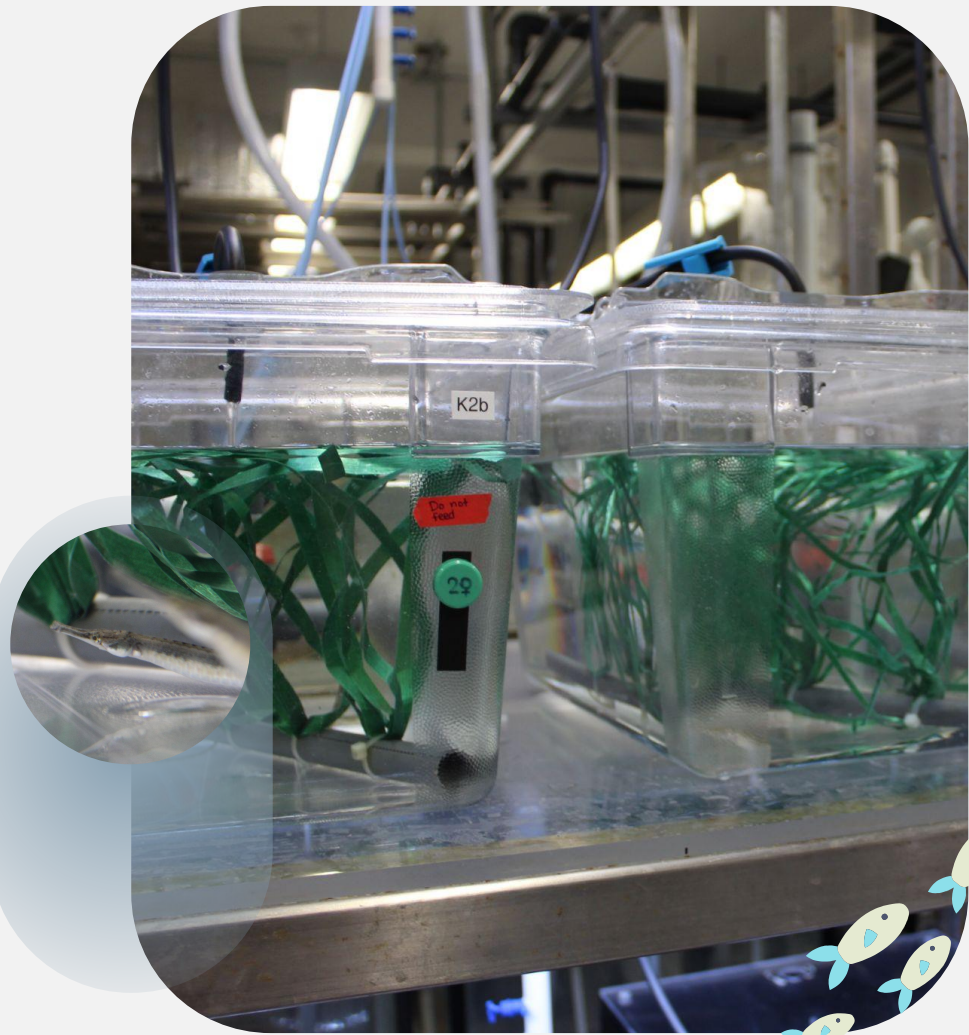


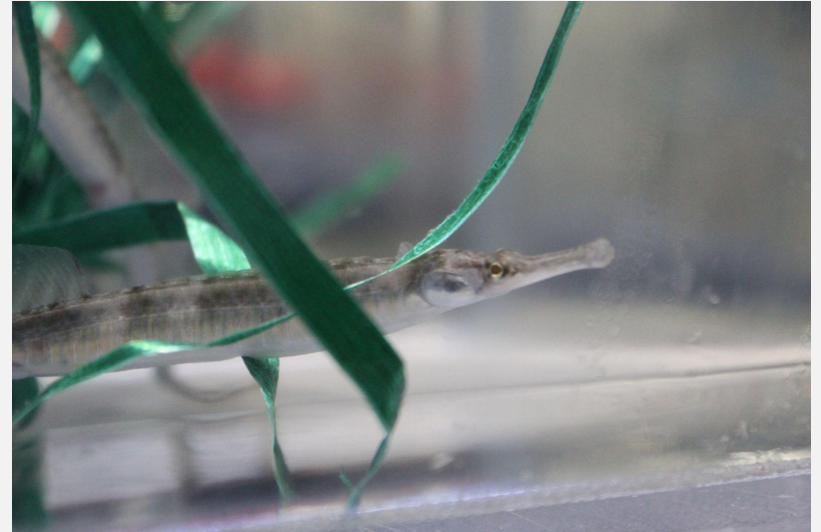
Investigating Parental Investment and Reproductive Behavior in Northern Pipefish (*Syngnathus fuscus*)

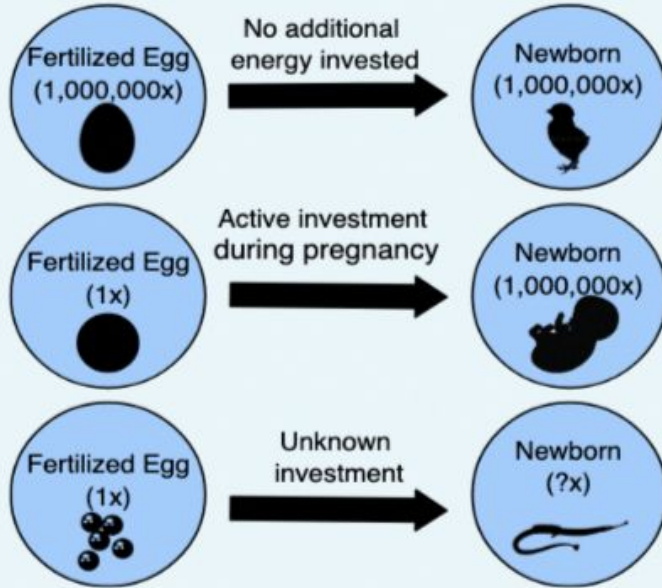
By Xylo and Evelyn



Background

- Northern pipefish belong to the family Syngnathidae
- Males are equipped with a brood pouch, allowing for male pregnancy
- Previous research shows males provide and absorb nutrients to/from embryos in the brood pouch (Ahnesjo et al. 2020)





Parental Investment

-How much energy parents invest into offspring during pregnancy

-Varies by sex and species

Other Important Terms

Fry

Newly born fish

Polygamy

A pattern of mating in which an animal has more than one mate

Monogamy

A pattern of mating in which an animal has only one mate at a time

Dry Weight

An organism's weight after all water content has been removed by drying



Previous Experiments

2019

- Found larger females produce larger eggs
- Found male energetic investment in *S. Fuscus* equates to that of female investment

2021

- Found only lipids in all samples (but higher lipid content in fry, contrary to past studies³ and high male mortality rate (90%) indicates subpar health in the population)
- Found female polygamy observed in 3 breeding pairs (female monogamous behavior in the wild may be due to low partner availability)

2022

- Found females mating with multiple males (polyandry is a physically possible mating strategy)
- Further evidence of monogamous mating due to low partner availability

What We Want to Do



- Measure energy investment by sex

- Determine if we can raise juveniles in the lab

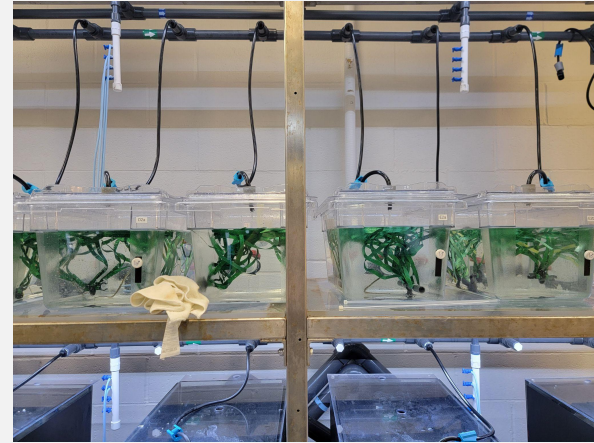
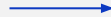
How Do We Do This?

- **Select females and males of similar* size, mass, and BMI**
- **Arrange mating pairs of 1 female to 2 males**
- **Measure carbohydrate, lipid, and protein concentrations of unfertilized eggs and fry**
- **Measure dry weight of unfertilized eggs and fry**
- **Repeat at least twice with same mating pair**
- **Set up nursery tank to raise fry not used for analysis**

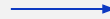
*relative to size distribution of the fish we already have



AREAC 127's Tank System



Connecting Top Tanks to the System



Selecting Female Pipefish

1)



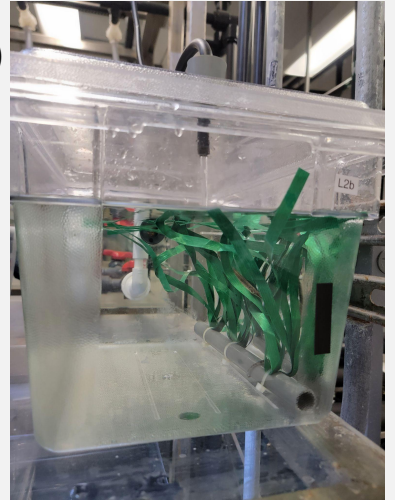
2)



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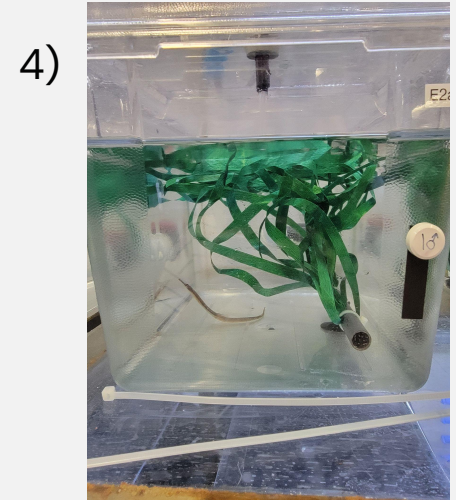
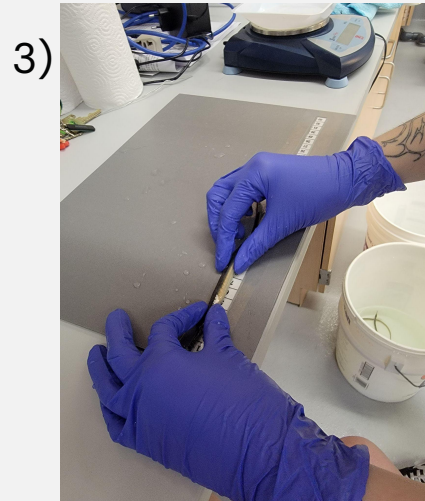
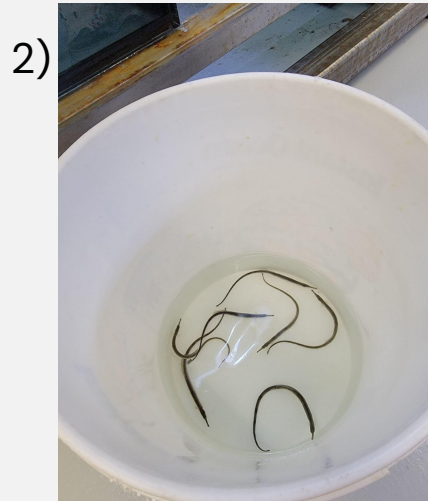
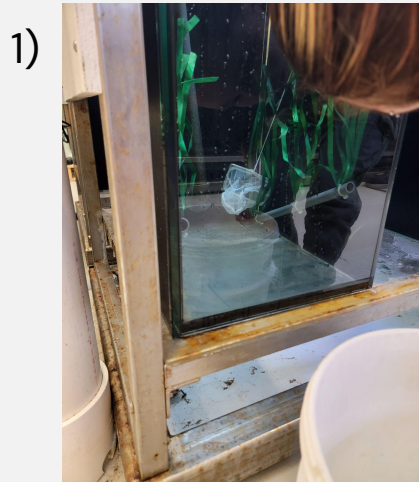


4)



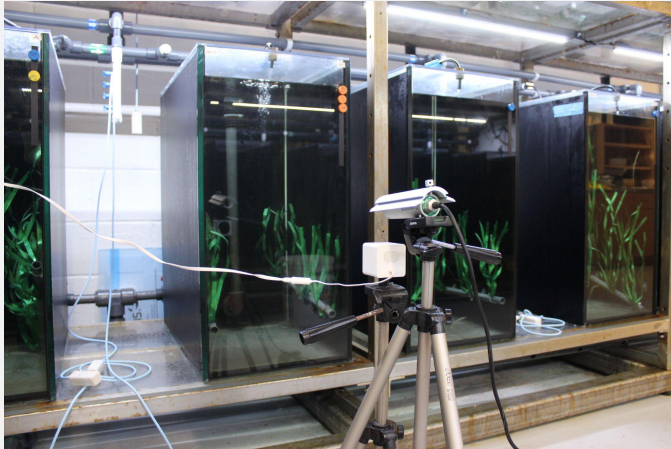
- We selected females between 16.5 and 18 cm in length with a rounded BMI of 0.009 to 0.01 at time of measurement
- Females who presented these measurements appear most healthy and sexually mature

Selecting Male Pipefish



- We selected males between 14 and 17 cm in length and with a rounded BMI 0.006 and 0.01
- Males who presented these measurements appear most healthy and sexually mature

Pilot Experiment



- Placed one female and two males together to see if they would mate (F: 15.2cm, M1: 14.9cm, M2: 15.1cm)
- Helps estimate the time needed for mating



Pilot Experiment Results



- Took about 4 hours for first mating
- Female polygamy is possible

Experimental Design: Mating Pairs

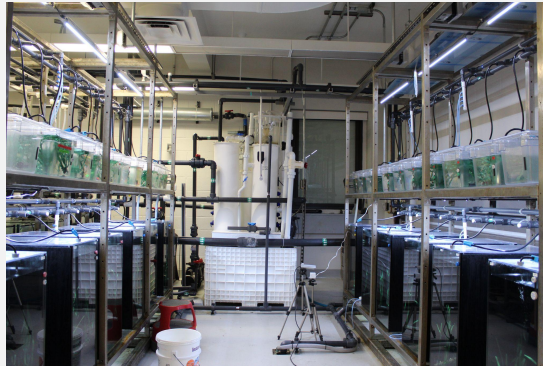
01

Measuring Fish



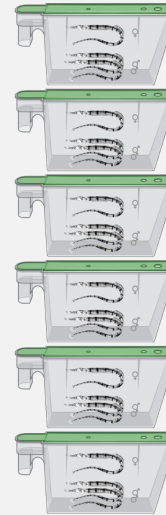
02

Selecting Fish



03

Running Experiment



Mating Pairs

- 6 females, 12 males
- Each female is paired with two males
- All 18 fish will be housed separately following mating

Repeat 2x with same pairs

Experimental Design: Nutrient Analysis

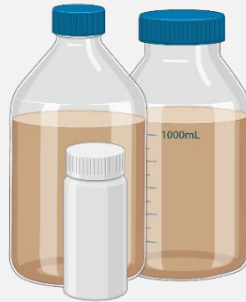
01

Gathering Protocol



02

Making Reagents



03

Performing Assays



Summary of Work

Complete:

- Performing fish care separately
- Measured and selected females and males
- Upgraded plumbing system for top tanks
- Prepared all tanks with enough fake plants to reduce stress
- Moved select females and males to top tanks
- Performed pilot experiment mating 1 female with 2 males

Working on:

- Collaborating w/ Dr. Musolf on behavioral experiments
- Selecting mating pairs
- Practicing nutrient analysis protocol

Future Applications + Relating to Urban Sustainability

Current Work

- Raising pipefish in captivity to avoid taking from natural habitat

Future Works

- Measuring the HSI of pipefish collected in different locations
- Determining if HSI is primarily influenced by environmental or genetic factors

Hepatosomatic Index (HSI)

- Ratio between liver weight and body weight
- Evaluates energy status of fish, good indicator of general health



Thank You! Any Questions?



Background Literature

- Braga Goncalves, Ines, et al. "Evolutionary Ecology of Pipefish Brooding Structures: Embryo Survival and Growth Do Not Improve with a Pouch." *Ecology and Evolution*, 24 Apr. 2016, www.ncbi.nlm.nih.gov/pmc/articles/PMC4864203/.
- Mobley, Kenyon B, et al. "No Evidence for Size-Assortative Mating in the Wild Despite Mutual Mate Choice in Sex-Role-Reversed Pipefishes." *Ecology and Evolution*, Jan. 2014, www.ncbi.nlm.nih.gov/pmc/articles/PMC3894889/.
- Paczolt, K., Jones, A. Post-copulatory sexual selection and sexual conflict in the evolution of male pregnancy. *Nature* 464, 401–404 (2010).
<https://doi.org/10.1038/nature08861>
- Paczolt, K.A., et al. *A Low Rate of Multiple Maternity for Pregnant Male Northern Pipefish Syngnathus Fuscus*, 2015, onlinelibrary.wiley.com/doi/abs/10.1111/jfb.12905.