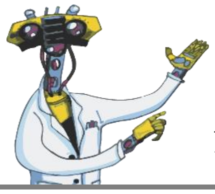

Video tutorials to support the

Best Practice Guide
for Multiple Drivers Marine Research

Experimental Evolution

Tutorial: The Experimental Evolution video tutorial can be found on the MEDDLE for Multiple Drivers Research YouTube channel.

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Transcripts: Rebecca Zitoun, University of Otago, New Zealand

Resources: The complete resources for the *Best Practice Guide for Multiple Drivers Marine Research* are available on the MEDDLE website.

0:00 – *Introduction*

When we study responses to global change we are often concerned with drivers that are going to change every season, decades or even centuries, which opens up the possibility that organisms evolve in response to this. Now because of this we might want to know what these evolutionary responses are and one way to study that is to watch evolution in the lab. So both of us work on marine microbes but evolution can occur in any organism as long as that organism has enough time for its genotypic composition to change. In the extreme that can be from one generation to the next generation.

0:40 – *Plastic responses*

So when we expose organisms to a new environment right away and just see what they do in the very short term we are often measuring a plastic response and a plastic response is what an organisms can do by changing the expression of its genes.

Text (0:51): Plasticity: a change in phenotype without an underlying change in genotype.

It is just using what it has, it is today's organism in tomorrow's world. And we all understand plasticity, this is what you do if you sweat or shiver due to changes in temperature, it is what the hulk does when he hulks out. You see big changes in traits but no underlying change in genetic material.

Text (1:09): Plastic response: you see changes in traits, but no underlying change in genetic material

1:14 – *Evolution: The change in the genetic composition of a population over multiple generations.*

So in order to study tomorrow's organisms in tomorrow's world we need to take evolution into account. One way to conceptionalize what evolution actually is, is to think about antibiotic resistance. So, what we have here (see graphic below) is a population made largely of bacteria that are going to die if you feed them an antibiotic. These are the yellow bacteria. We have in the population also a couple of bacteria that are already resistant to that antibiotic, they already carry a mutation that makes them resistant. These are displayed in red. So, if you hit this population with strong antibiotic, most of the bacteria that don't carry the resistant making mutations are going to die. So you are left with a population of mainly the red bacteria.

Text (1:20): Let's consider antibiotic resistance. Yellow bacteria: susceptible to antibiotics. Red bacteria: resistant to antibiotics.

2:00 – *What is experimental evolution?*

So what is experimental evolution? Really, it is just a method where you take populations and you evolve them in the lab under conditions that you care about and under the drivers you are interested in for long enough for evolution to happen.

Text (2:04): Experimental evolution - Evolve populations in the laboratory under conditions you are interested in.

And ‘long enough’ will depend on a lot of thing s. It will depend on how strong selection is for that population in that environment, it will depend on how much standing genetic variation is, how fast your population can generate more variation and whether or not your population has sex.

Text (2:14): Experimental evolution - “Long enough” will depend on selection, genetic variation, and sex.

So these are all things that have to be considered when you are planning an evolution experiment. And this means that some multigenerational experiments will involve evolution and some won’t.

Text (2:32): Experimental evolution - Some multigenerational experiments will involve evolution. Some won’t.

2:36 – *Fitness*

One thing that is really nice about experimental evolution is that it is meant to be general and one way in which we keep things general is by framing what is happening in terms of fitness. And fitness is quite easy to define but notoriously difficult to measure. Fitness is the contribution of a genotype to the next generation. So it is how many of your genes you put forward into the next generation.

Text (2:52): Fitness: the contribution of a genotype to the next generation.

And that means, when you are planning an evolution experiment, you need to have a pretty good idea of what constitutes fitness in your particular experiment. And that is something that, to some degree, you can manipulate. For example, if there are no grazers in your experiment then resistance to grazing is not part of your fitness in your experiment.

One example that we were able to find this out, we looked at whether evolution is different in stable vs. fluctuating environments and whether or not it matters if you go into in environment and being already plastic or not having a lot of plasticity.

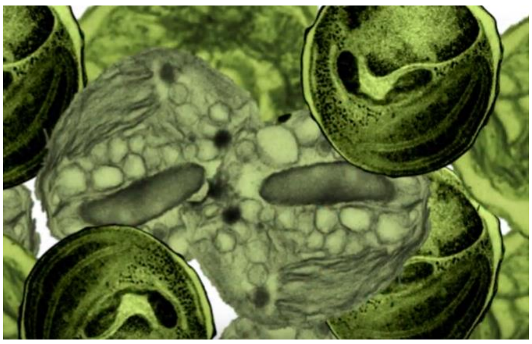
Text (3:20): Is evolution different in stable vs. fluctuating environments? An example from the lab.

3:35 – *Designing the experiment: Is evolution different in stable vs. fluctuating environments?*

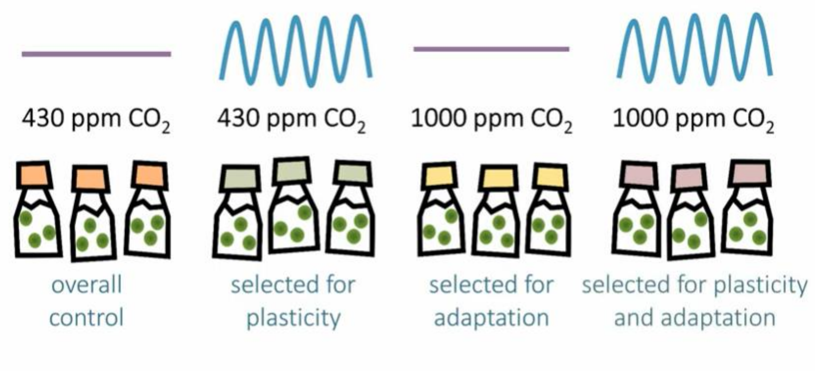
In order to answer our questions about how fluctuations in the environment have an impact on evolution and specifically on the evolution of plasticity we needed a driver of choice which is fairly easy to manipulate in the laboratory an also it needed to be a driver which we knew will change in the future and one we knew would be different in different regions of the ocean today.

Text (3:41): Question: how do fluctuations in the environment impct the evolution of plasticity?

So that was a pretty tall order, so we ended up choosing CO_2. We also needed an organism that was really widely distributed and easy to grow in the lab, because you are going to discover that we grew a lot of cultures of this organisms and that is how we fell madly in love with *Austria Cocus.* We grew these three replicates at each population in four different environments for 400 generations. And by we, I mean Elisa. You can see that there are two levels of CO_2, ambient and high, and fluctuating and stable levels of CO_2 for each case. The stable ambient was the overall control and this is a standard lab environment for *Austria Cocus.* The fluctuating ambient was selected for plasticity only, so no high CO_2, just experiencing environmental fluctuations. The stable high CO_2 environment was selection for adaptation to high CO_2 only and the fluctuating high CO_2 environment was selected on both plasticity and the ability to grow at high CO_2. So at the end of the experiment we used reciprocal transplants to measure how much each population had evolved.



Text (5:13): Reciprocal transplants were used to measure how much each population had evolved.



We measured some traits that were important for how *Austria Cocus* fits into the marine ecosystem. So this could be size, photosynthesis rates and cellular composition but to understand what was going on with the natural selection we measured two different fitness proxies, that being growth rate and competitive ability.

Text (5:19): Size, photosynthesis rate and cellular composition were uses to understand food webs an biogeochemical cycles. Two fitness proxies were used to understand natural selection: growth rate and competitive ability.

We found that populations with more plastic ancestors evolve more and this agreed, thank goodness, with all of the theory on the topic, which gets me to the next bit, which is that experimental evolution is founded on a large body of theory. Which means we often can make really strong hypotheses for our experiments an often run models and simulations beforehand, which is good because the experiments take a long time.

Text (5:40): Schaum, C. Elisa, and Sinéad Collins. "Plasticity predicts evolution in a marine alga." Proceedings of the Royal Society B: Biological Sciences 281, no. 1793 (2014): 20141486.

6:02 – *Recommendations*

Text (6:02): Tip 1: simulate your experiment

We have a few tips for you and the first one, probably the most important one, is to simulate your experiment and analyse your simulated data before you start. MEDDLE is a great place to start with this but simulating your experiments can be done with a wide variety of platforms.

Text (6:19): Tip 2: know your power

When you end up designing your experiment it is really important that you know your statistical power and you can get to that statistical power from your simulated data set. Also, it is really really important to carry out pilot studies. Tonnes of pilot studies. Tonnes and tonnes of pilot studies.

Text (6:35): Tip 3: have an idea of the expected response

Third tip is to have an idea of how large you expect your effect size to be and how noisy you expect your data to be. This is going to influence not only the amount of replication you use but how you chose to detect your signal and then methods that you use. It is also going to effect the number of times you have to measure the same population over and over if that is one of the ways you are detecting evolution.

Text (6:59): Tip 4: have sufficient replication

Related to power, make sure you have enough statistical replication, because otherwise you won't be able to link cause and effect.

Text (7:04): Tip 5: collaborate, collaborate, collaborate

Experimental evolution or evolution is sort of the Zen of science in that it is one of those words that everyone throws around a lot but not too many people are sure what it actually means. Evolution is a well-developed science with quantitative theory inside of it. So we strongly suggest collaborating. Especially with marine biologist, because marine biology an evolutionary biology often ask questions that will actually benefit from collaborating with each other. Sadly marine biology and evolutionary biology in the past have kind of grown up separately from each other, but we strongly suggest that you collaborate.