



Ocean Acidification
International
Coordination Centre

OA-ICC



UNIVERSITY OF
GOTHENBURG



THE ROYAL SWEDISH ACADEMY OF SCIENCES

KUNGL.
VETENSKAPS-
AKADEMIEN

Basic training course on ocean acidification

EVT1804704

14-19 March 2022

Experimental design



Take home messages

Every experiment is an abstraction of reality



George E. P. Box

There is nothing like a perfect experiment !

“Essentially, all models are wrong,
but some are useful”

Essentially, all experiments are wrong,
but most are useful

Be aware and honest about your limitations

How to design your experiment



1. What is your question? Your hypothesis?
2. How can I test this?
 - What are my limitations?
 - What is the best model?
 - What are the best endpoints?
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 - What are my controls?
 - etc.

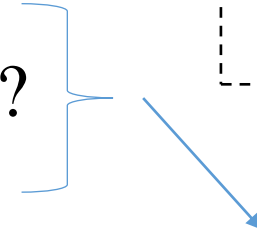
Can I REALLY answer my question with the collected data?

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Do the
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thing

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Trade-offs



Realism

[duration, tested parameter, environment, etc.]

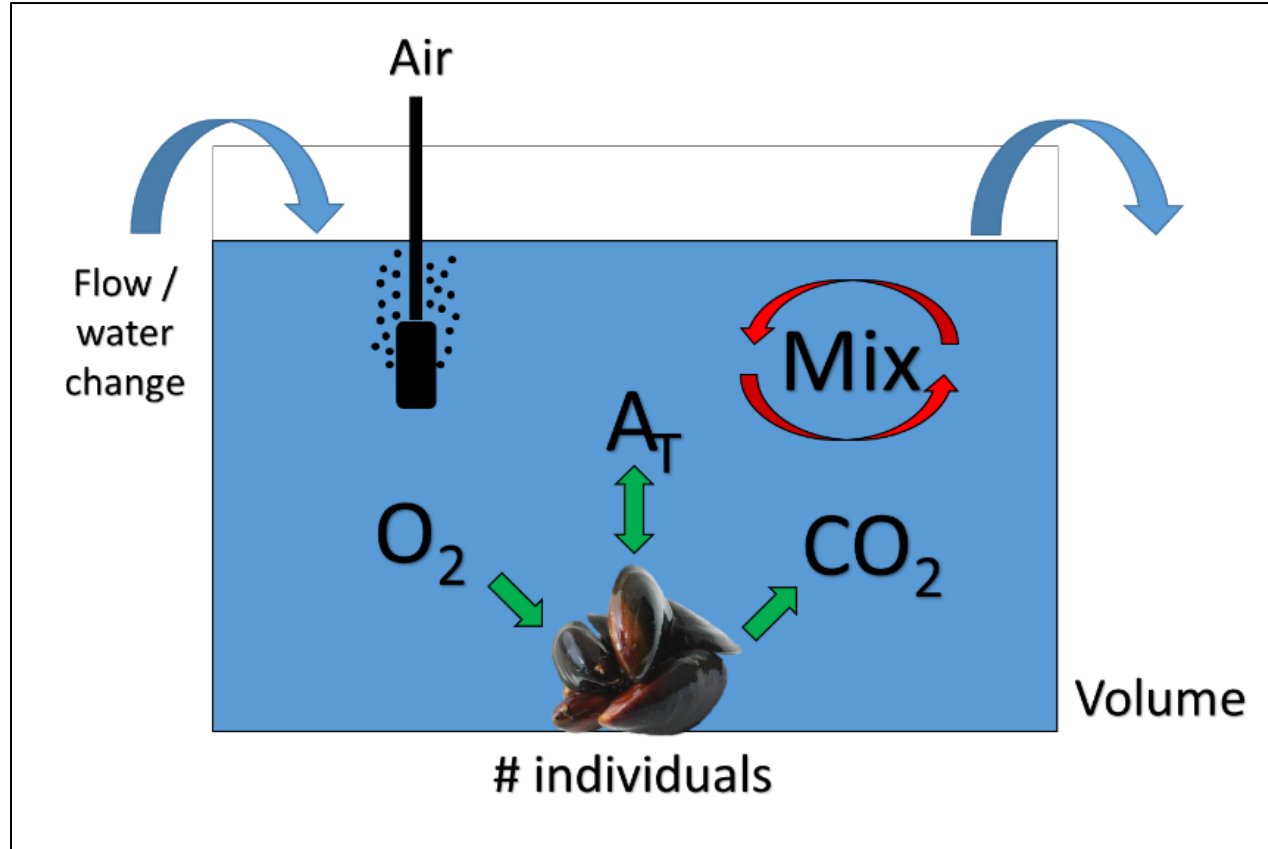
VS.

Feasibility

[manpower, money, space, time]

The aquarium system

Depend on the species and stage/size/density/species specificities



The aquarium system

Depend on the species



- ✓ Type and amount of food
- ✓ Physico-chemical conditions
- ✓ Behaviour (escape, cannibalism, etc.)
- ✓ Etc.

If not well designed, can lead to
confounding factors

Need pilot experiment

Duration

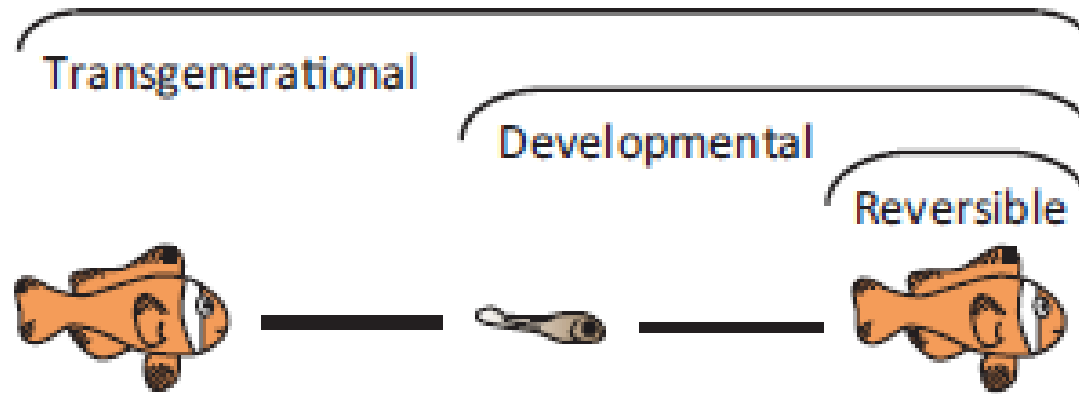
Depend on the species and question



Effect of ocean acidification
on fecundity

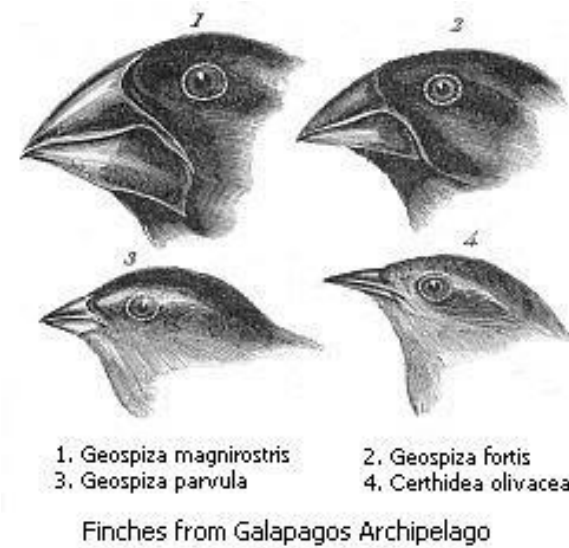
... but can be long

Duration - stages



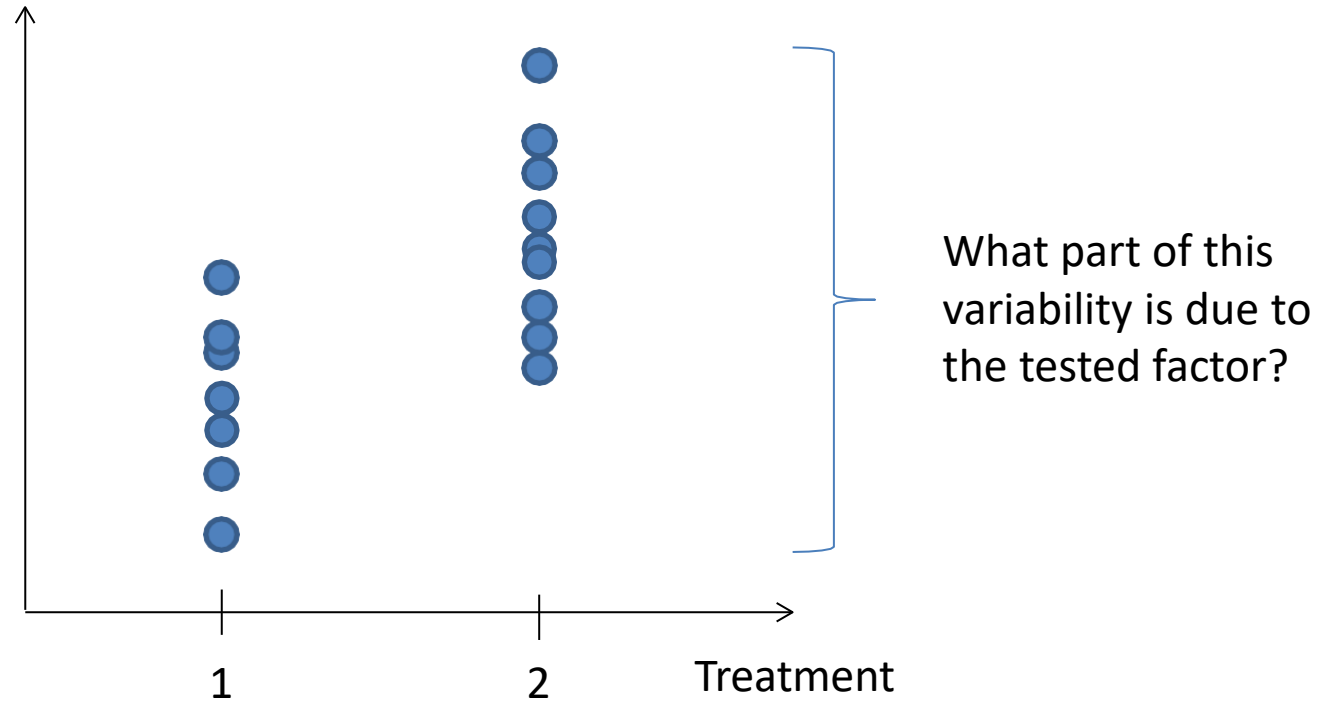
More resilient offsprings to OA

Variability



Explain variability

Sources of variability



”biologically relevant” vs.
”technical/confounding factors”

Confounding factors – need for replication



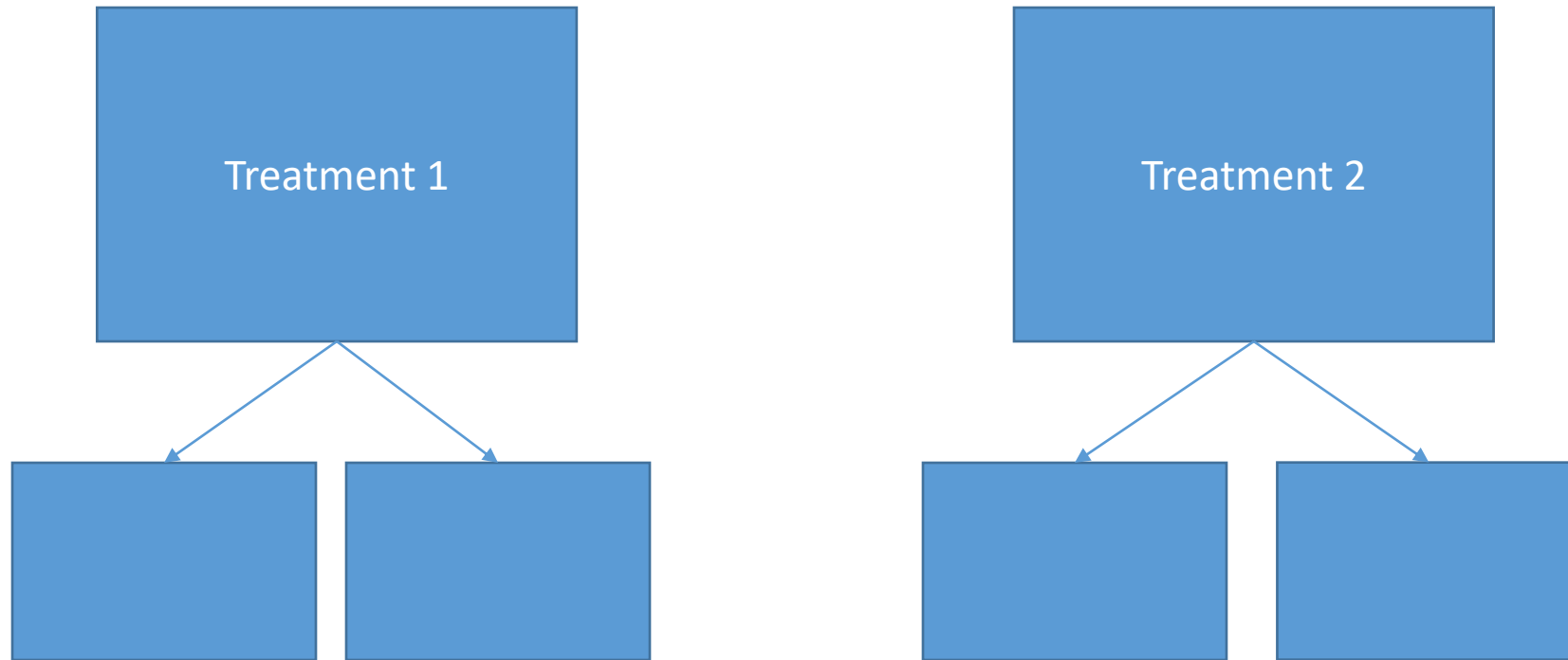
e.g. position in the room
dirt in aquarium
etc.

-> Observed effect not attributed to
the treatment

Solution: replication + randomization



Pseudo-replication



Sources of variability



Unwanted variability may "hide" real variability

Higher the variability, the more data you need to see effects

Variability depends on endpoints

-> Minimize unwanted variability

-> Pilot studies + power analyse
(number of replicates / samples)

Replication



Parameters

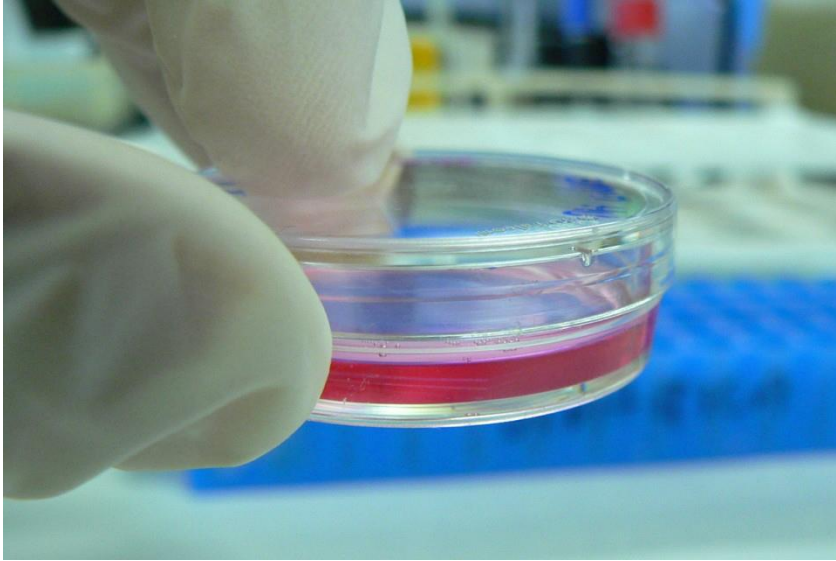
Treatments

replicates

tanks

1	x	2	x	2	=	4
1	x	2	x	4	=	8
1	x	4	x	4	=	16
2	x	4	x	4	=	32
3	x	4	x	4	=	48

Practical limitations



Practical aspects - Summary



- ✓ Aquarium system
(water quality, stability, etc.)
- ✓ Duration
- ✓ Source of variability / Frequency
- ✓ True replication
- ✓ Randomization

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August Krogh

VIII.

The Abnormal CO_2 -Percentage
in the Air in Greenland and the General Relations between
Atmospheric and Oceanic Carbonic Acid.

By

(Krogh 1904)

August Krogh.

Krogh's principle

“For such a large number of problems there will be some animal of choice, or a few such animals, on which it can be most conveniently studied”

The top model



- Biological feature (e.g. life cycle, generation time)
- Ecological / Economical importance
- Tools available (e.g. functional methods, genome)
- Charismatic species
- etc.

What fits your question?



- Size / Weight
- Age
- Life-history stages
- Etc.

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Endpoints?



Fitness (e.g. survival, growth, reproduction)

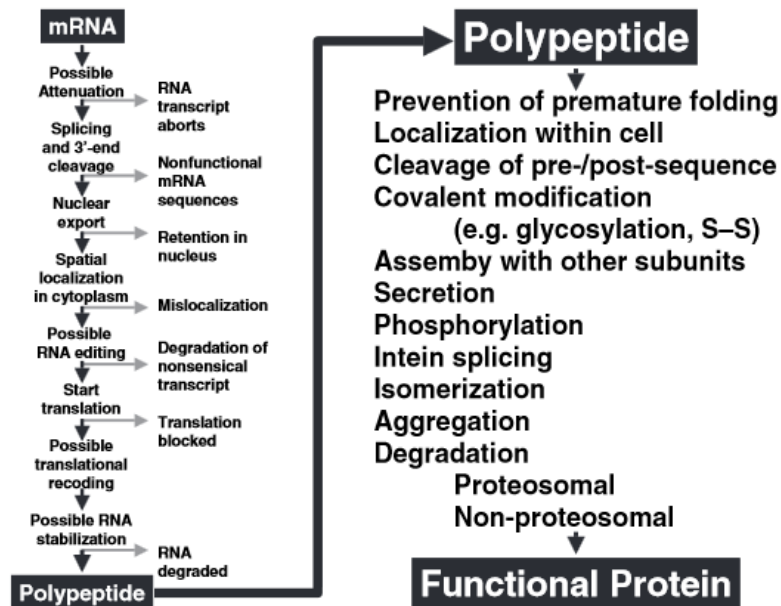
Physiology – energy budget
(e.g. respiration, feeding, excretion, calcification)

Etc. etc.

Best endpoints?

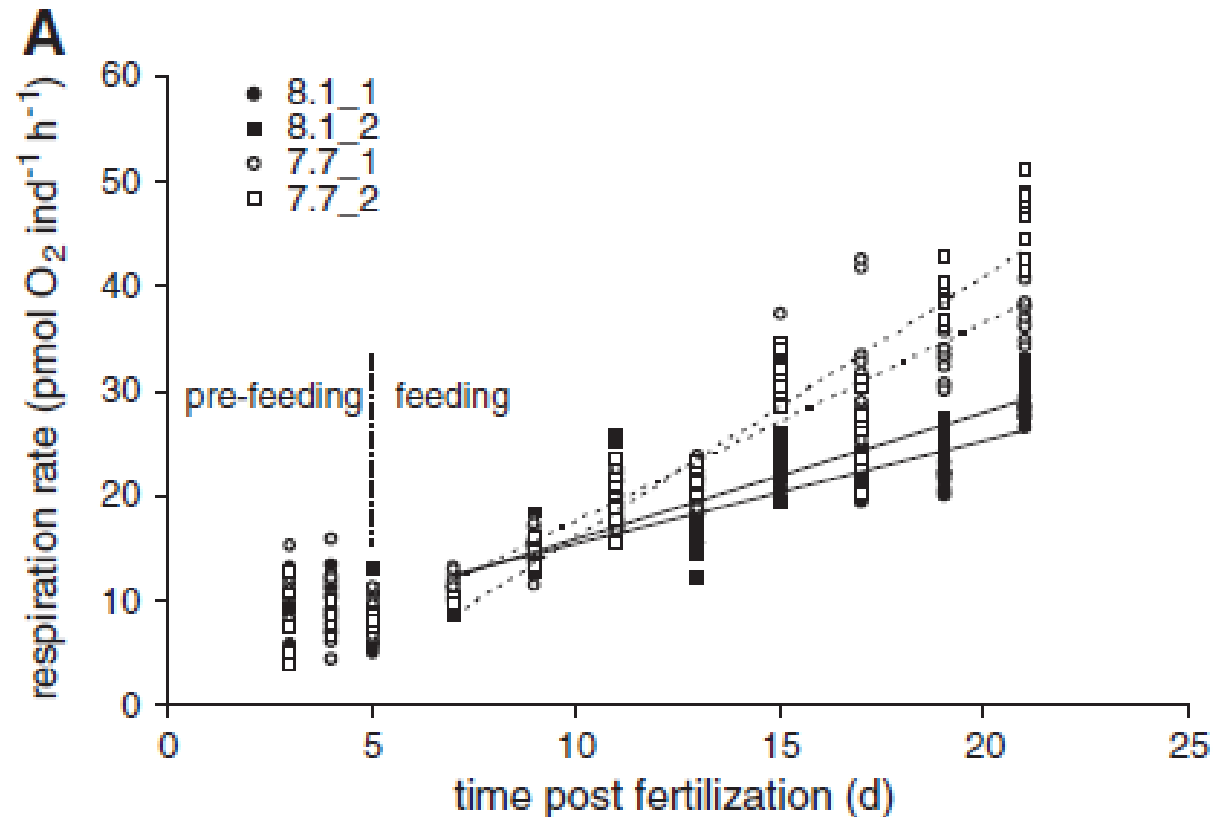


- Not the “coolest” method
- Not the most familiar method
- As close to function as possible (e.g. fitness)



How often?

Frequency (more = more chance to identify effects & interactions)



Changes \neq bad



We like bad news Negative effect: 9.8 citations / year
Positive/neutral effect: 6.2 citations / year

A change in your proxy \neq change in fitness

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How to design your experiment



“ANOVA” design

Little predictive power



$n=x$



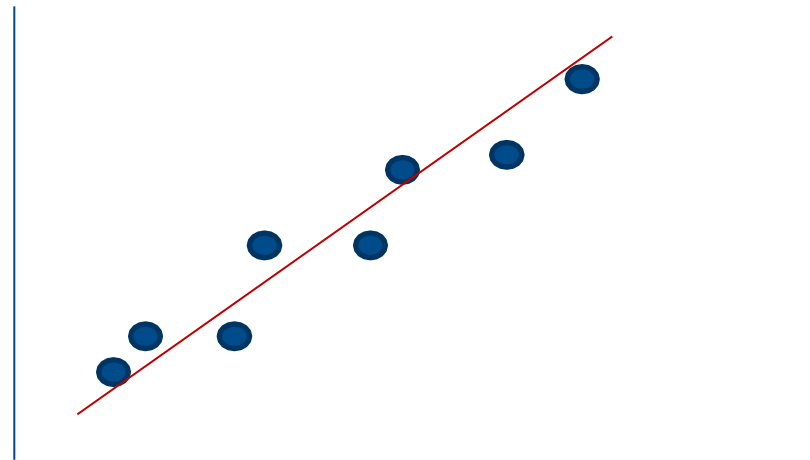
$n=x$



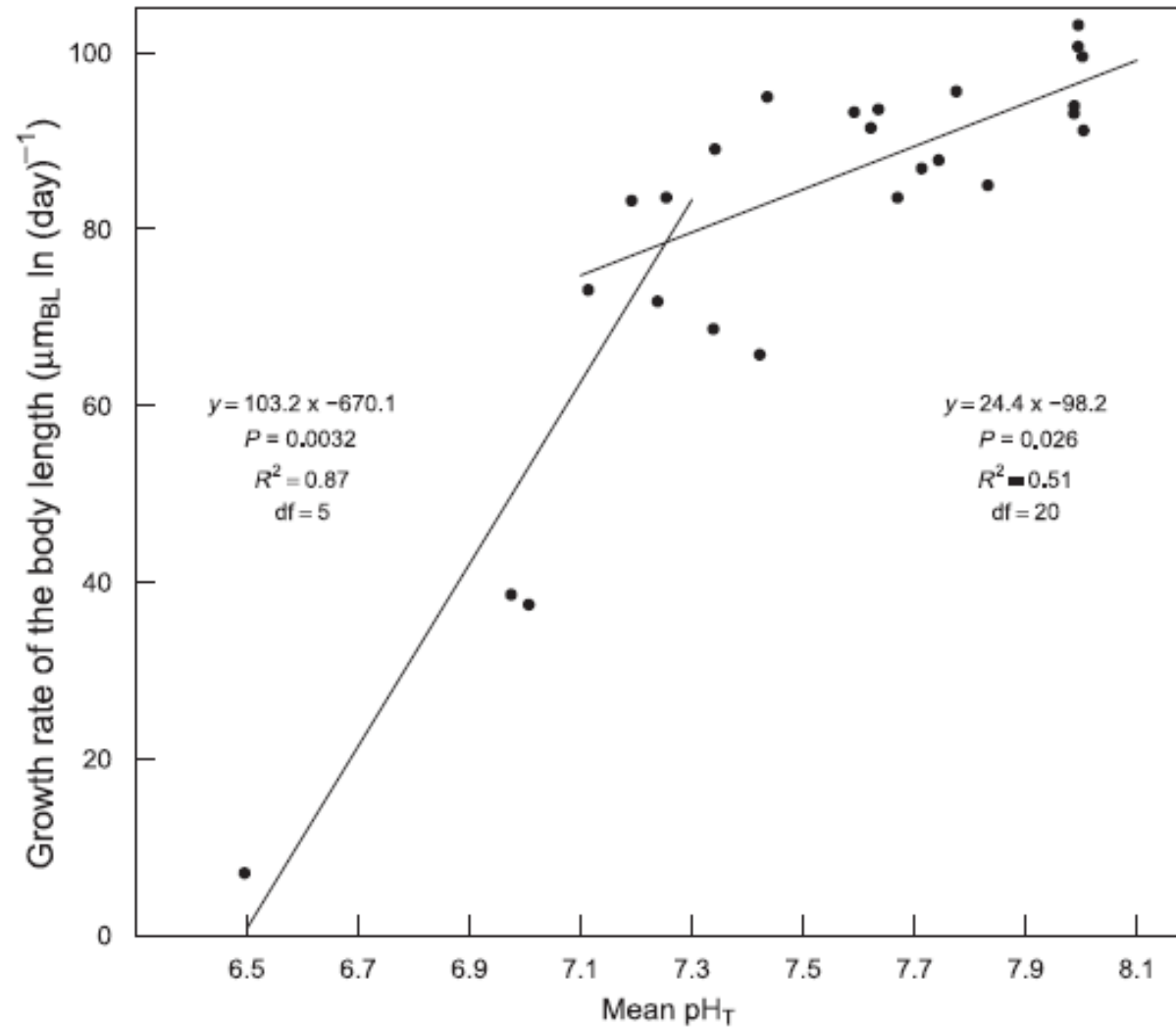
$n=x$

“Regression” design

Need to have a relationship



ANOVA vs. Regression



(Dorey et al. 2013)

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Test the right scenarios



Take species niche, behaviour & variability into your thinking

Example: impact of ocean acidification on chemical communication



Two temperature: CTL vs OA

Example: impact of ocean acidification on chemical communication



Emission



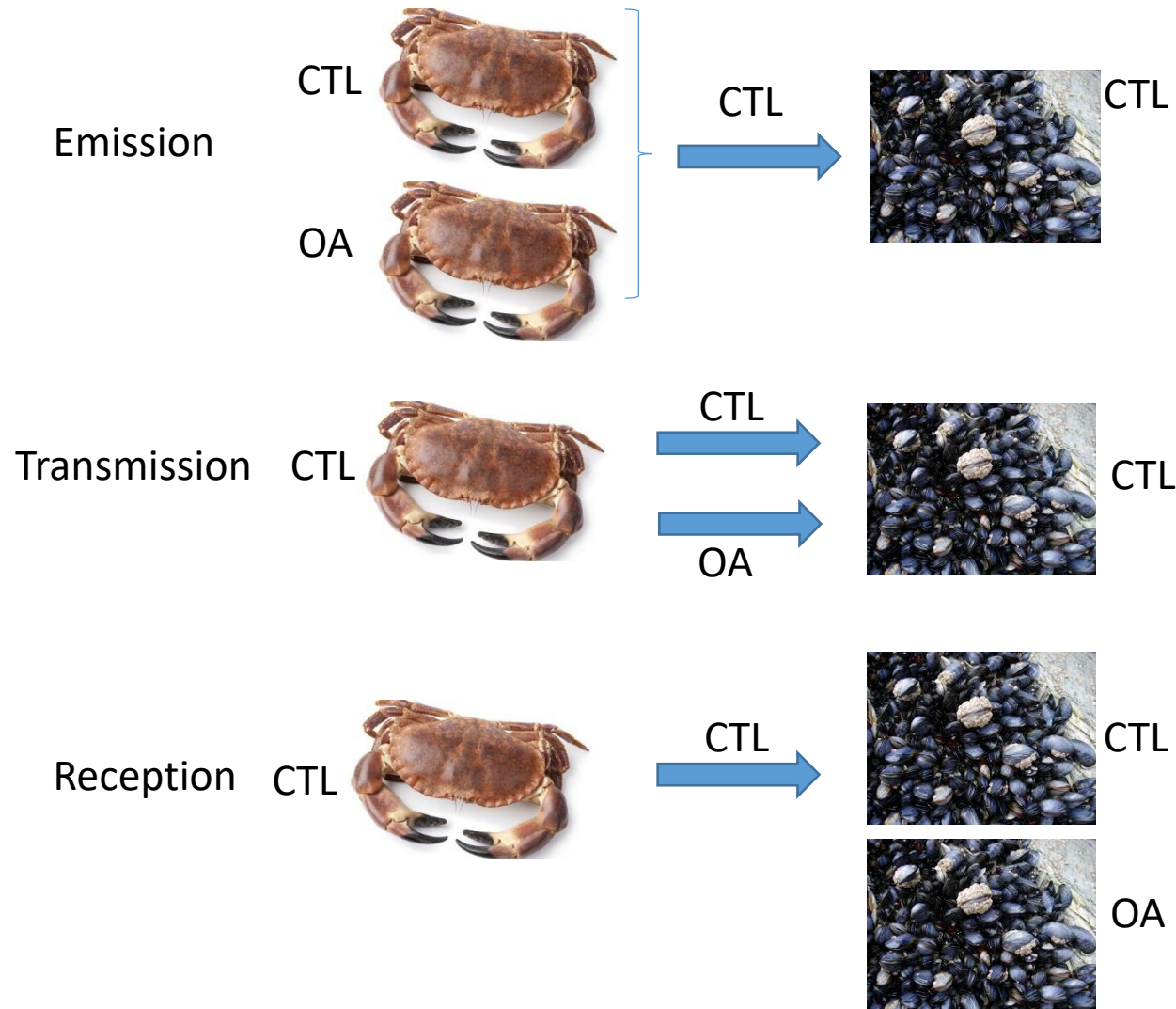
Transmission



Reception



Example: impact of ocean acidification on chemical communication



+ *interactions*

***2 emissions x
2 transmissions x
2 receptions
= 8 treatments x
replicates***

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