Module M2 "Concepts et méthodes en Biologie"

Part 2: Methods

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Recap:

- Observation is a prerequisite
- Framing the hypothesis
- Experimental design

Where do you do the experiment?









Dead samples (Fossils, scat samples, etc)

Live samples







Dead samples (museum collections, hospital donations, etc)

Live samples





In both





Ethics in Animal Experimentation – RRR concept



Choosing the right study system?

• Depends on the type of questions you want to ask!



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Choosing the right study system?

• Depends on the type of questions you want to ask!



Choosing the right model system in the lab

	2D cell culture	C.elegans	D. melanogaster	D. rerio	M. musculus	PDX	Human organoids
Ease of establishing system	✓/X	1	1	1	1	 Image: A second s	1
Ease of maintenance	1	\checkmark	\checkmark	1	1	 Image: A second s	1
Recapitulation of developmental biology	×	√	\checkmark	1	1	×	1
Duration of experiments	1	1	1	1	1	1	1
Genetic manipulation	1	1	1	1	1	×	1
Genome-wide screening	1	1	1	1	×	×	1
Physiological complexity	×	1	\checkmark	1	1	1	1
Relative cost	1	1	1	1	1	1	1
Recapitulation of human physiology	1	1	1	 Image: A start of the start of	1	1	√
	✓ Best	t 🗸 Good	🗸 Partly suitab	le 🛛 🗡 Not suita	ble		

Controlled environment in the lab



Controlled environment in the lab



Specific experimental paradigms to study evolution

Phylogenetic analysis



Dird Represe
 Micro-oyot
 Normal-eyed - Eyed

1.0

© S. hutsensis b S. plubelensis

Specific experimental paradigms to study evolution

Phylogenetic analysis



Anaeini leafwing butterfly lineages







Example: Drosophila Learning and memory



Example: Drosophila Learning and memory



Example: Drosophila Learning and memory





Generations

Cost and trade-offs



Cost and trade-offs



Experimental evolution



Traits evolved			
Improved survival on poor diet			
Smaller body size			
Faster development			
Reduced critical weight			
Reduced cell number in wings			
Increased larval competitive ability			
Reduced foraging behaviour			
Increased propensity for cannibalism			
Reduced prepupal tunneling			
Increased susceptibility to gut pathogens			
Modified nutrient allocation			

Line	Origin			
P ₁	Control line			
P ₂	Selected line			
F1	$P_1 \times P_2$			
F _{1R}	$P_2 \times P_1$			
F ₂	$F_1 \times F_1$			
F _{2R}	$F_{1R} \times F_{1R}$			
B _{1a}	$P_1 \times (P_1 \times P_2)$			
В1ь	$P_1 \times (P_2 \times P_1)$			
B _{1Ra}	$(P_1 \times P_2) \times P_1$			
B _{1Rb}	$(P_2 \times P_1) \times P_1$			
B _{2a}	$P_2 \times (P_1 \times P_2)$			
B _{2b}	$P_2 \times (P_2 \times P_1)$			
B _{2Ra}	$(P_1 \times P_2) \times P_2$			
B _{2Rb}	$(P_2 \times P_1) \times P_2$			



The Genomic Architecture of Adaptation to Larval Malnutrition Points to a Trade-off with Adult Starvation Resistance in *Drosophila*

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Hypothesis testing



The developmental instability- sexual selection hypothesis

Møller and Pomiankowski (1993)

Individuals could use fluctuating asymmetry as cues to assess the genetic quality of potential mates and sexual rivals.



Major assumption: Females can perceive & assess male-asymmetry to discriminate against asymmetric males during mate choice Since 1990's, several studies: diverse species and traits Yet results are largely inconclusive.

Major constrain • Natural systems have very little variation for FA in bilateral traits

Alternative approaches

not natural

- Artificially manipulating bilateral traits
- Environmental stress induced FA
- Genetic mutants and inbred populations that show FA

Suitable controls ; reproducibility and DI associated confounding effects





Random positioning machine







Random positioning machine







Altered gravity (Ag)

Uniform gravity (Ug)







FA index 6 = variance 2(R-L)/(R+L)



Vijendravarma et al. PNAS (2022)

Effect of AG on other bilateral traits



Development and life-history traits in AG



Uniform gravity
- Altered gravity

Vijendravarma et al. PNAS (2022)

Mate choice paradigm



Mate choice paradigm





Vijendravarma et al. PNAS (2022)

Sensory basis of this FA-based discrimination?



Drosophila courtship

McKinney 2019



Vijendravarma et al. PNAS (2022)









parameter





Vijendravarma et al. PNAS (2022)

FA in song parameters



Inter pulse interval

Pulse frequency

Vijendravarma et al. PNAS (2022)

FA in song parameters



Pulse amplitude

Pulse amount

Vijendravarma et al. PNAS (2022)



Morley et al 2018 (The Journal of the Acoustical Society of America)

"The Rational"

 \rightarrow



Larval imaginal discs



Adult bilateral structures

"The Rational"



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Open circulation of hemolymph

"The Rational"



Oxygen supply through bilateral tracheal network

"The Rational"



Oxygen supply through bilateral tracheal network

Role of trachea in maintaining bilateral precision?







Trachea in embryo

Preliminary result

Role of trachea in maintaining bilateral precision?



Preliminary result





Still- reared larvae



Rotated larvae

Preliminary observation





Preliminary observation



- We looked at HIF- dependent and HIF-independent gene expression for genes known to respond under hypoxia
- Three treatments: Still, Uniform gravity and Altered gravity
- Two developmental stages: mid-L3 and wandering stage larvae
- Method: qRT-PCR

List of HIF-dependent and HIF-independent genes

HIF-dependent genes

- 1. Fatiga
- 2. Spermineoxidase
- 3. Sequoia
- 4. Branchless
- 5. Breathless
- 6. CG31769

HIF-independent genes

- 1. peroxiredoxin 2540-2 (Prx2540)
- 2. LDH
- 3. Pfk
- 4. NMNAT
- 5. Alas
- 6. Cyp9b1
- 7. Cyp6a17
- 8. Cyp6a22

HIF-dependent genes



HIF-independent genes



Working model



Working model



Working model

