Fokus Life Sciences

WS 21/22

Methods in Behavioral Physiology (I)

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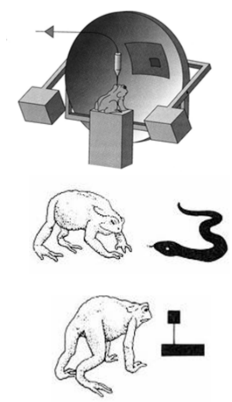
Behavioral <u>Physiology</u>:

what does it focus on?



on the <u>physiological</u> basis of behavior, i.e.
on the mechanisms underlying behavior

In more general terms….

Behavioral <u>Biology</u>:

…studies the <u>biological</u> basis of behavior, i.e., both the <u>underlying mechanisms</u> as well as the <u>consequences</u> (<u>adaptive value</u>) of behavior

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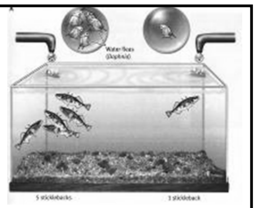
Methods:

How to study <u>animal behavior</u>?



but first, a definition: What is <u>animal behavior</u>?

Behavior includes all those processes by which an animal senses the external world and the internal state of its body, and responds accordingly (adaptively)

Many of these processes occur "inside" the animal, and may not directly be observable (but indirectly/directly detectable: → calcium imaging, electrophysiology, etc)

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… and from "outside" of the animal?

An animal may show marked activity or be completely at rest, and <u>both</u> are behavioral states.

That means, the range of phenomena called "behavior" presents us with two challenges (problems):

- how to observe
- how to measure

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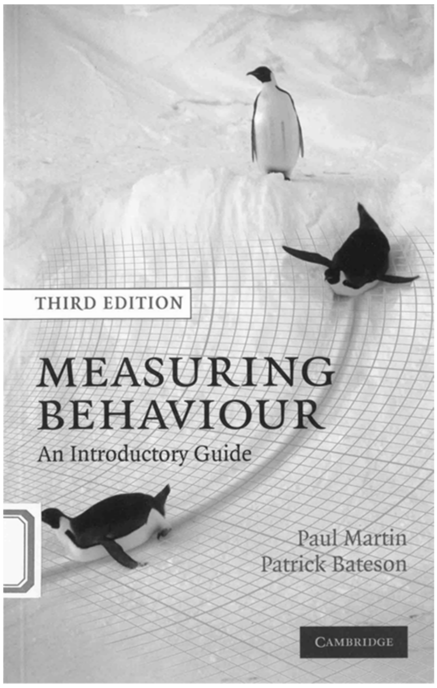

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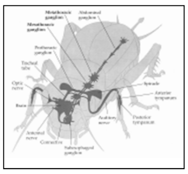
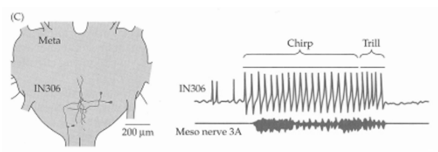
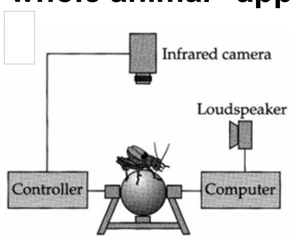
There are two main approaches to study animal behavior:

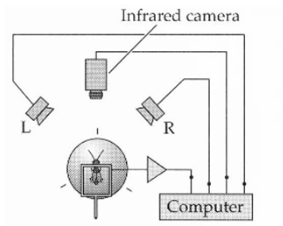
(1) The physiological approach





(2) The "whole animal" approach





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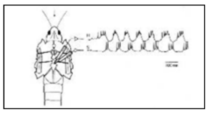
1) The physiological approach



Classically referred to as "behavioral physiology", it focuses on the study of the mechanisms underlying behavior

→ "how the machinery works" to produce complex behavior

2) The "whole animal" approach

It focuses on the study of behavior in intact animals and of the factors that influence it.

For instance:
What it is in the environment of a bird that prompts it to sing at a particular time? Or why does a bird sing at all?

When the approach focuses on the adaptive value of behavior, it is often referred to as "behavioral ecology"

Adaptive value of behavior → its fitness consequences

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VERY IMPORTANT !

1) The physiological approach
2) The "whole animal" approach

There is considerable overlap between the two approaches!!

Both are needed to understand behavior!!
→ integrative studies

For instance, a "whole animal" approach is often used to understand physiological mechanisms, and vice versa

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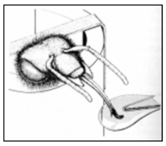
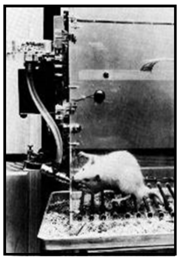
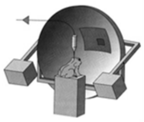
Examples:

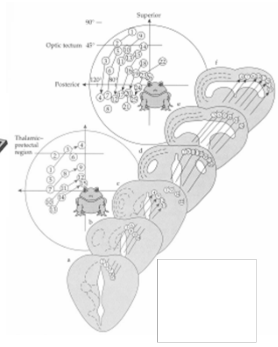




A "whole animal" approach used to understand cognitive abilities

A physiological approach used to understand "whole-animal" responses





Regardless of the approach used, what you need to understand animal behavior is:

"A feeling for the organism ….." → adaptive value / ecology of behavior!

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… *back to the methods*

1) The physiological approach

(not mutually-exclusive methods):

- molecular
- neural (cellular)
- hormonal

2) The "whole animal" approach

(not mutually-exclusive methods):

- field observations/descriptions (e.g. time budgets)
- "asking" animals under controlled, semi-natural conditions
- non-disturbing, contact-less monitoring of behavioral and/or physiological variables (e.g. thermography, respirometry, tagging/tracking devices)

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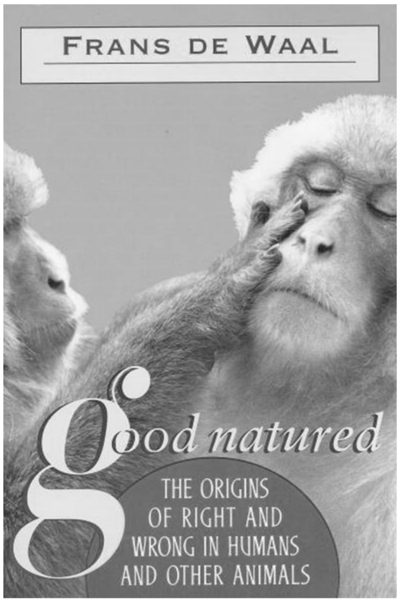
Do methods need to be complex or high-tech ?

Simple methods, large outcome …



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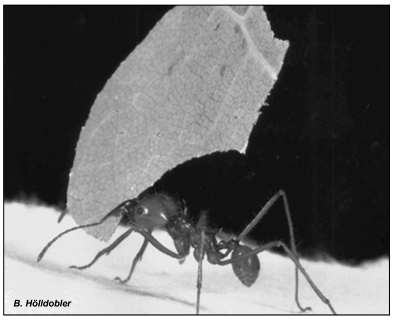
How to monitor behavioral states and/or physiological variables?

One approach is to quantify the energetic costs of behavioral responses (gain-costs analysis)

Most simple behaviors: activity vs. rest



foraging activity (locomotion / load transport)

rest

Method: high-resolution respirometry

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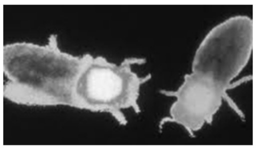
Respirometry:



measuring the "fire of life" (metabolic rates)

Metabolic rate is a measure of the "fire of life" (energy expenditure) burning *per unit time* within an organism

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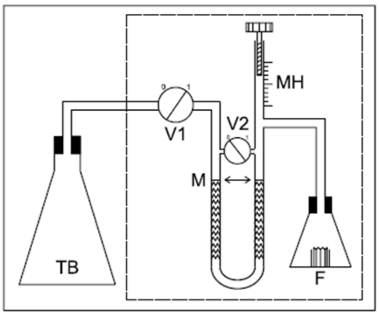
Respirometry measurements:

(1) The constant pressure respirometer (no gas sensors required)



F: flasks with the animal + CO_2-absorbent
TB: thermo-barometer (flask)
V: valves
M: manometer meniscus
MH: micrometer head

Procedure:

- **Put an animal within the flask**
- **Absorb any CO_2 produced (KOH)**
- **Measure the change in volume over time**
- **Periodically adjust the chamber's volume**

The change in the micrometer heads' reading over time is equivalent to the rate of O_2 consumed

Potential problem: organisms may be sensitive to decreasing O_2 levels (hypoxia)

The basic principle of the Warburg-respirometer is analogous

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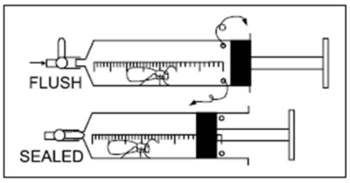
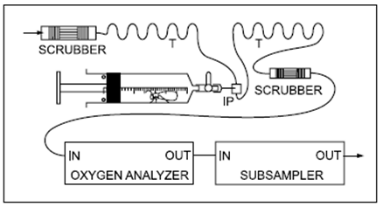
(Lighton JRB 2008, *Measuring Metabolic Rates*)

(2) Constant volume techniques with gas sensors

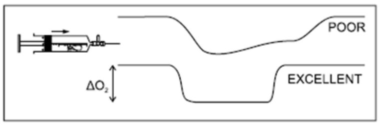


Experimental setting



Gas analysis after a time interval

Scrubber: water and CO_2-absorbtion
T: tubing
IP: injection port
Subsampler: pump



Output of the gas analyzer

(it depends on injection volume, injection rate, and baseline flow rate)

The area below the O_2-baseline (integral) corresponds to the total O_2-volume consumed

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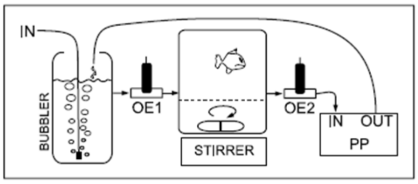
(Lighton JRB 2008, *Measuring Metabolic Rates*)

(3) Flow-through respirometers

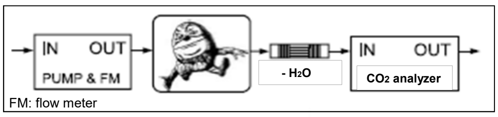
Basic principle: air (or water) circulates continuously through the respirometric chamber, and the rate of decrease in [O_2] or the rate of increase in [CO_2] are measured downstream

Example: aquatic flow-through respirometer (O_2)



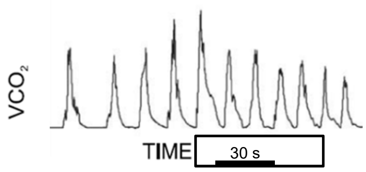
OE: oxygen electrodes
PP: peristaltic pump

Example: simple push-mode respirometer (CO_2)



FM: flow meter

Example: single insect



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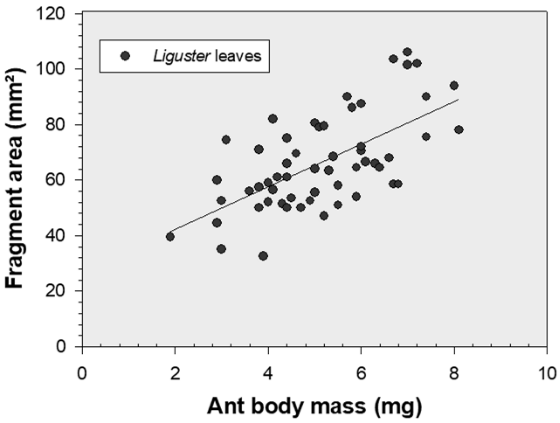
(Lighton JRB 2008, *Measuring Metabolic Rates*)

Energetics of behavior – costs of foraging

Leaf-cutting ants as a case study (I)

How do ants decide about the size of their loads?



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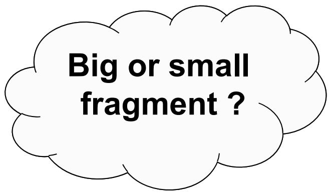
Foraging decisions: Loading and information transfer

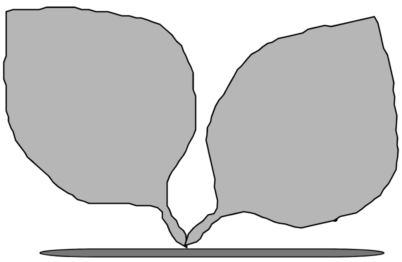


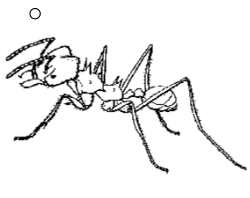
Larger fragments:

- **more material**

but….

- **higher cutting costs**
- **longer travel times**
- **information transfer?**





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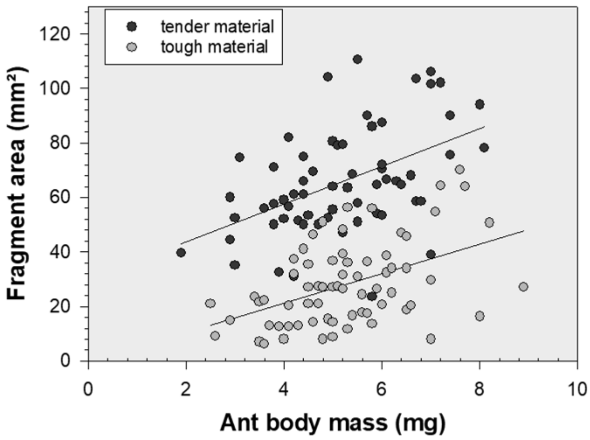
Flexible cutting rules:
leaf toughness and fragment size



The size of the harvested leaf fragments depends on the mechanical properties of the leaf
→ cutting costs important?

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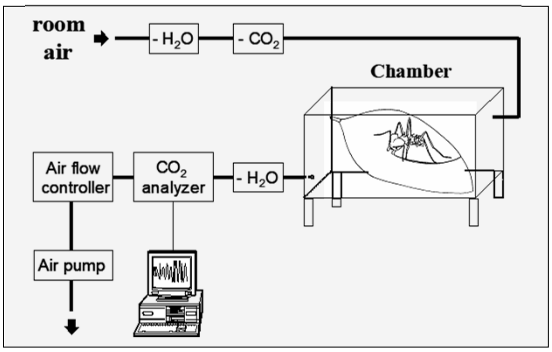
Cutting costs and fragment size



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Foraging costs: cutting and running

Treadmill running and locomotion energetics

Internal state: haemolymph sugar levels

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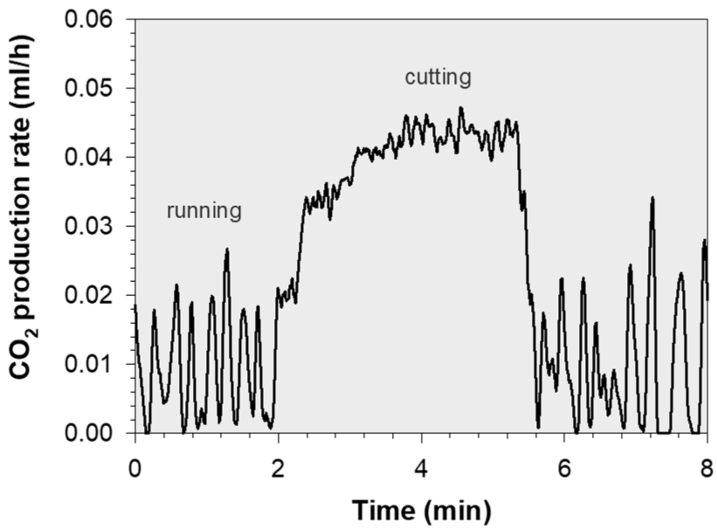
CO_2 production rate while cutting



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Factorial Metabolic Scope

(factorial increase in metabolic rate during activity, compared with resting metabolic rate)

Running (vertebrates & invertebrates)	≈ 12
Flying	20 - 100
Leaf-cutting	31

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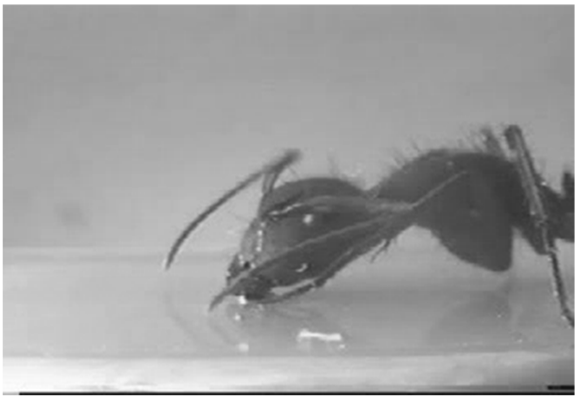
Extrafloral nectar is not provided *ad libitum*, but at variable flow rates ....

- while feeding, ants have to wait for the nectar to be produced

... time / energy budgets become important



Video: Oliver Geißler

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How *Camponotus rufipes* workers decide about the size of their nectar loads?

Does the energy investment during feeding influence the decision when to stop drinking and return to the nest?

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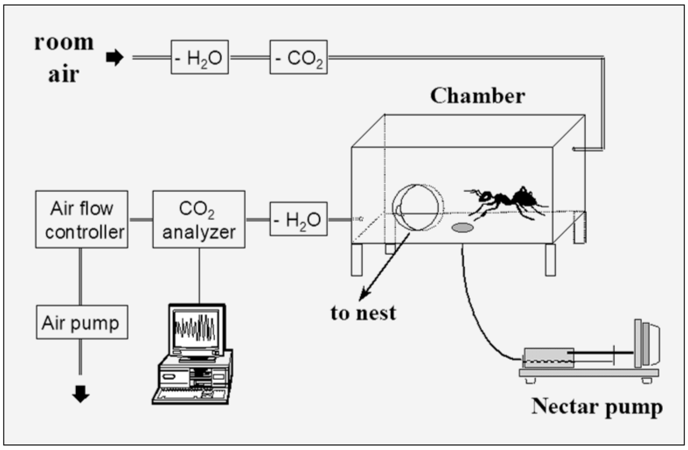
Experimental set-up



Energy INPUT: ingested nectar (at controlled flow rates)
Energy OUTPUT: CO_2 production rates (respirometry)

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Summary

1- Energy costs are important in determining behavior

2- Respirometry measurements allow behavior to be energetically characterized, with high temporal resolution

3- Precise gain-costs analysis of behavioral responses are possible

4- Energetics considerations are relevant to understand the evolution of behavioral traits

To be read:

Schilman PE, Roces F (2006)
Foraging energetics of a nectar-feeding ant: metabolic expenditure as a function of food-source profitability. Journal of Experimental Biology 209:4091-4101

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