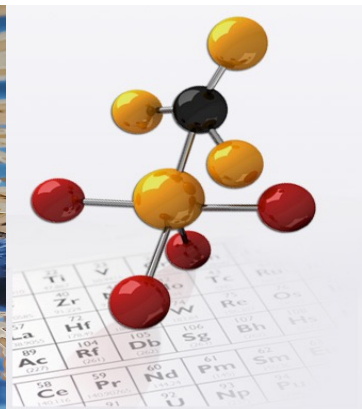


# How Do We Study What Animals Eat?

- **Direct Observation**
  - Pros: Rich data, can be collected at the individual level
  - Cons: Time- and cost-intensive, predation is difficult to observe
- **Scat or Gut/Stomach Content Analysis**
  - Pros: Captures diversity of diet
  - Cons: Snapshot, biased towards hard parts, invasive (stomach/gut)
- **Chemical Proxies (Stable Isotope Analysis)**
  - Pros: Integrates over a variety of timescales, non-invasive
  - Cons: Low resolution (does not capture prey taxonomic diversity)

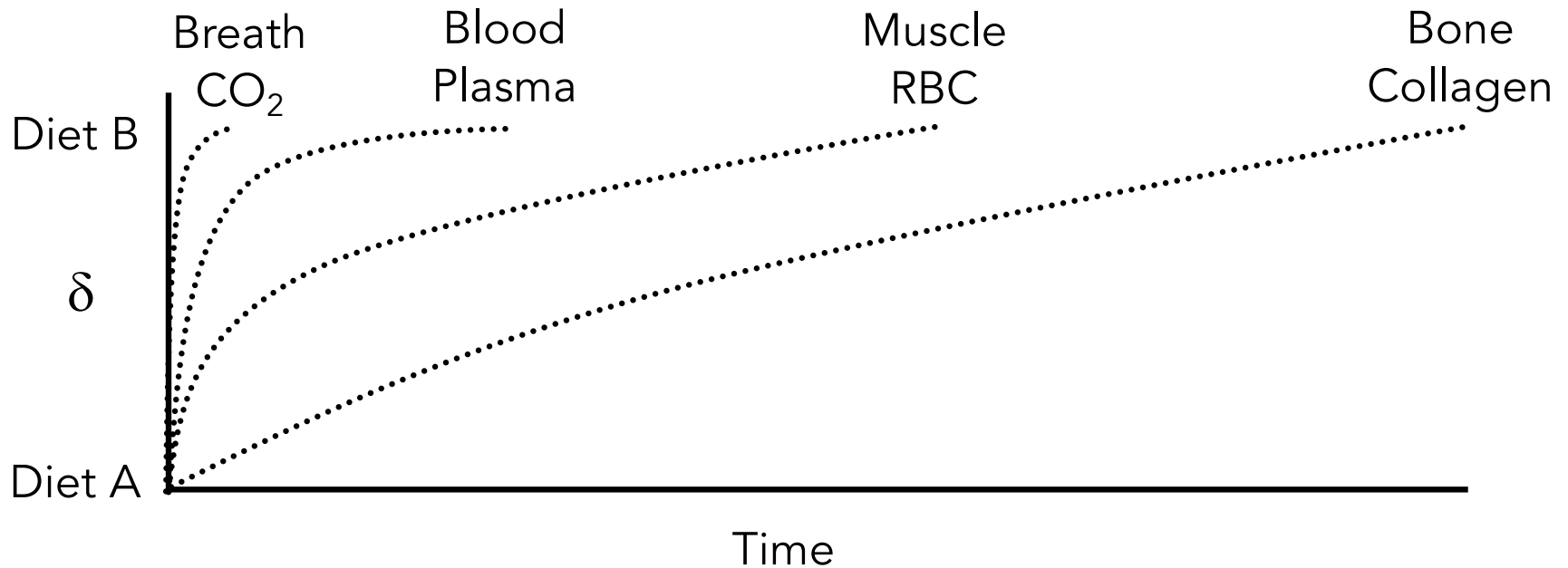


# Isotopic Incorporation (aka Turnover)

Metabolically Active  
(Continuous Turnover)

Breath (CO<sub>2</sub>)  
Blood Plasma & Liver  
Skin  
Red Blood Cells  
Muscle  
Bone Collagen

Fast  
Slow

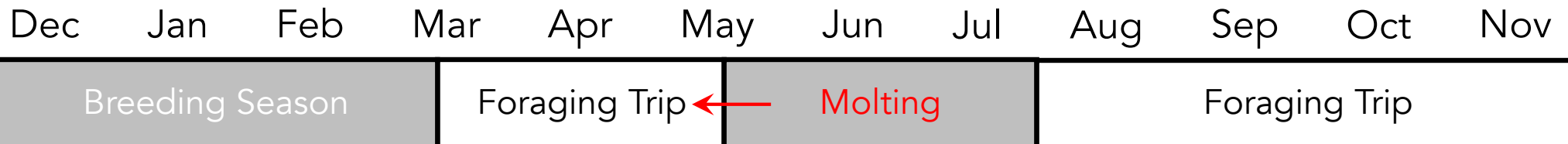
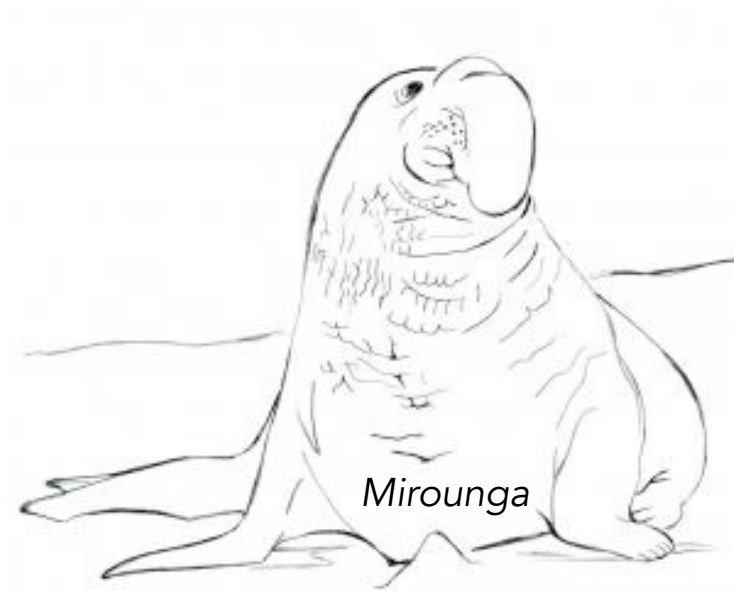


# Isotopic Incorporation

Metabolically Active  
(Continuous Turnover)  
Breath (CO<sub>2</sub>)  
Blood Plasma & Liver  
Skin  
Red Blood Cells  
Muscle  
Bone Collagen

Metabolically Inert  
(Short Time Periods)

Fur  
Feathers



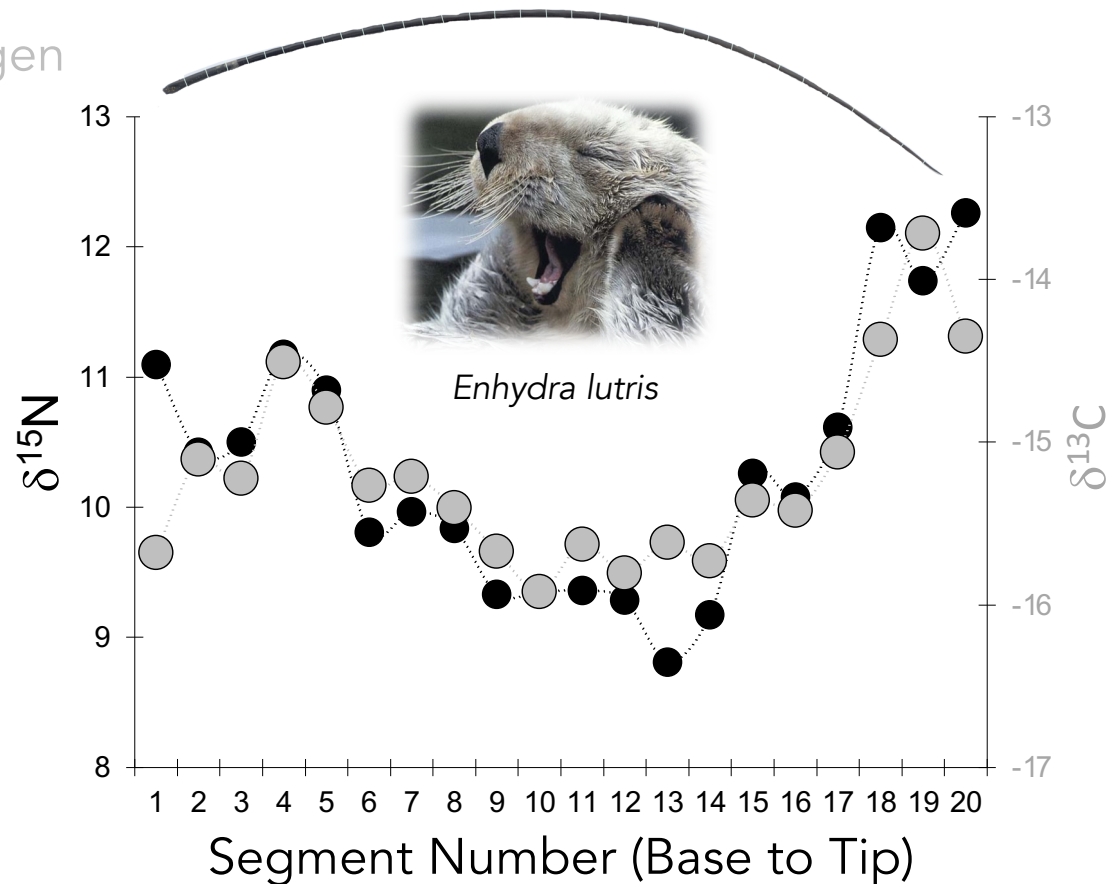
DO NOT EAT

# Isotopic Incorporation

Metabolically Active  
(Continuous Turnover)  
Breath (CO<sub>2</sub>)  
Blood Plasma & Liver  
Skin  
Red Blood Cells  
Muscle  
Bone Collagen

Metabolically Inert  
(Short Time Periods)  
Fur  
Feathers

Metabolically Inert  
(Accretionary)  
Tooth Dentin  
Vibrissae  
Baleen  
Claws/Nails



## Carefully Chose Which Tissue You Sample

### Metabolically Active (Continuous Turnover)

Blood Plasma  
Liver  
Red Blood Cells  
Muscle  
Bone Collagen  
Breath (CO<sub>2</sub>)

### Metabolically Inert (Short Time Periods)

Fur  
Feathers

### Metabolically Inert (Accretionary)

Tooth Dentin  
Vibrissae  
Baleen  
Claws/Nails

What do you want to know?

Must think about life history, especially annual life cycle!



# What About Plants?





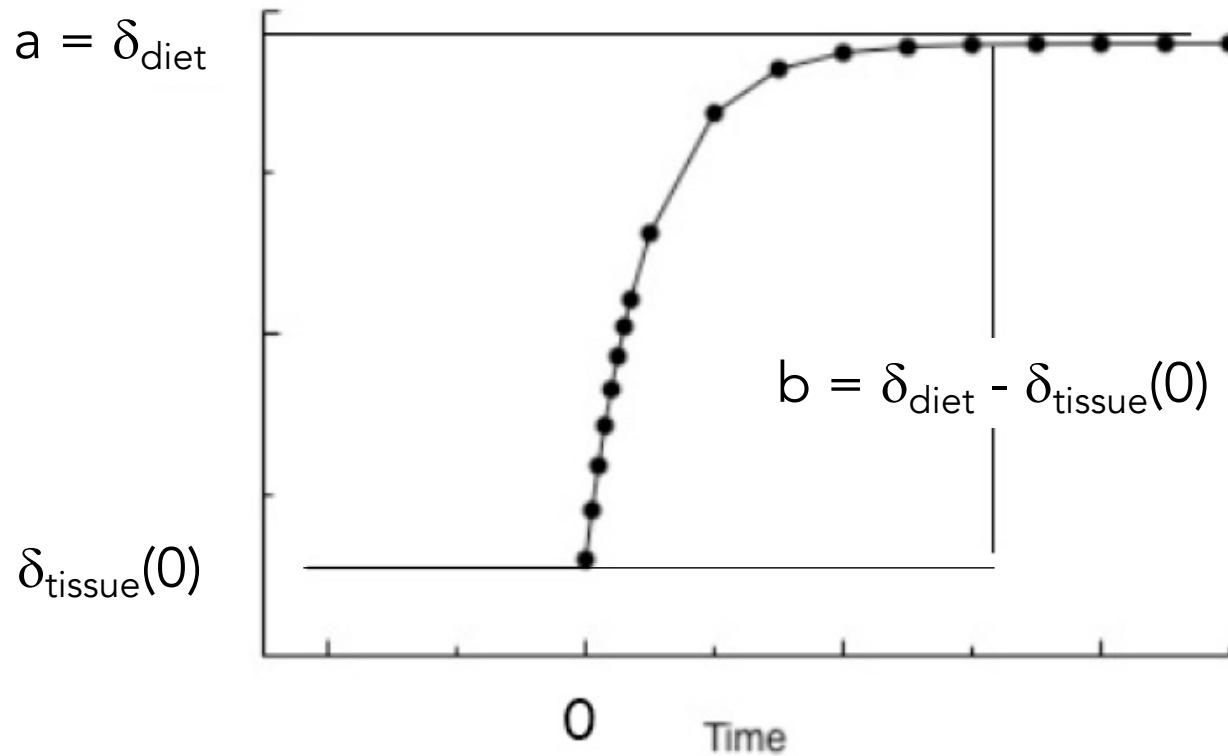
What About Bacteria?



# Kinetics of Isotopic Incorporation

$$\delta_{\text{tissue}}(t) = \delta_{\text{diet}} - (\delta_{\text{diet}} - \delta_{\text{tissue}}(0))e^{-rt}$$

$$\delta_{\text{tissue}}(t) = a - be^{-rt}$$



Diet change at time zero from  $\delta_{\text{tissue}}(0)$  to  $\delta_{\text{diet}}$

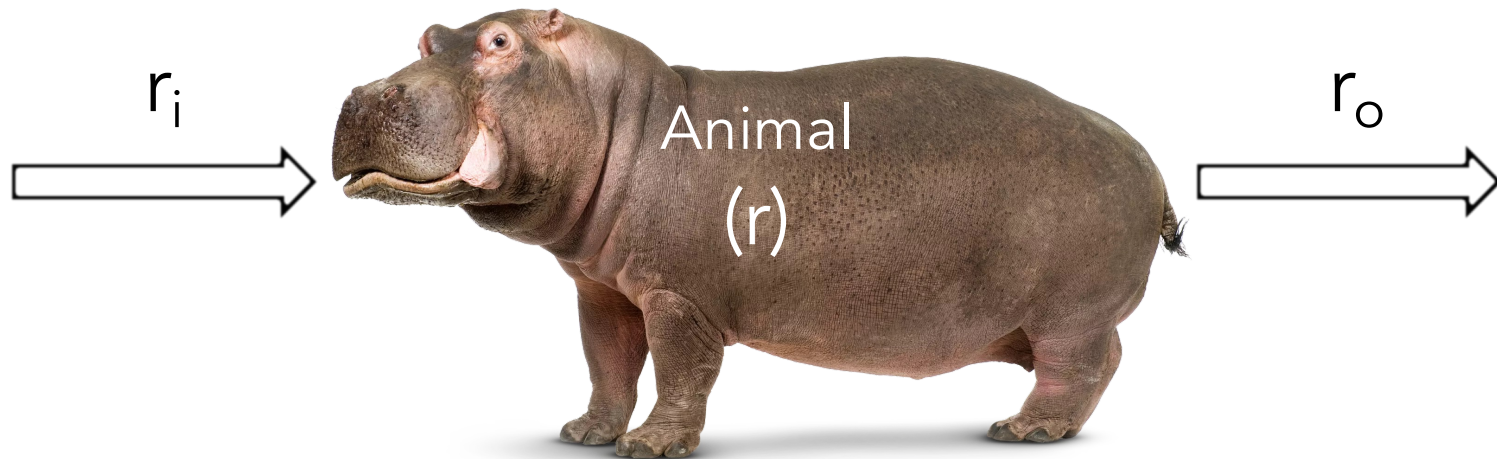


# Kinetics of Isotopic Incorporation

$$\delta_{\text{tissue}}(t) = a - be^{-rt}$$

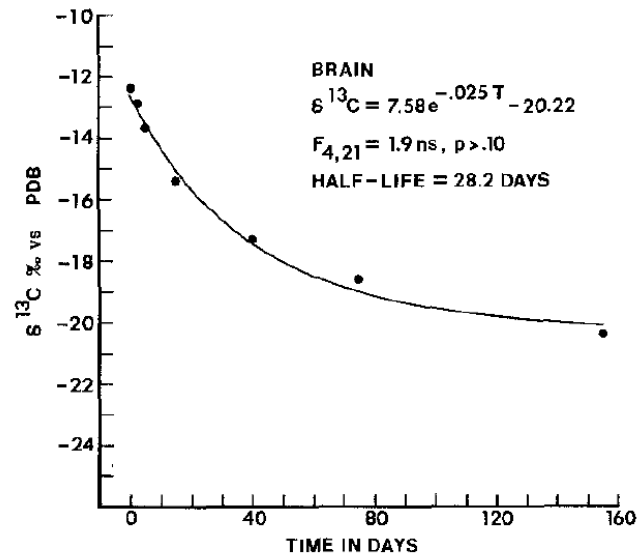
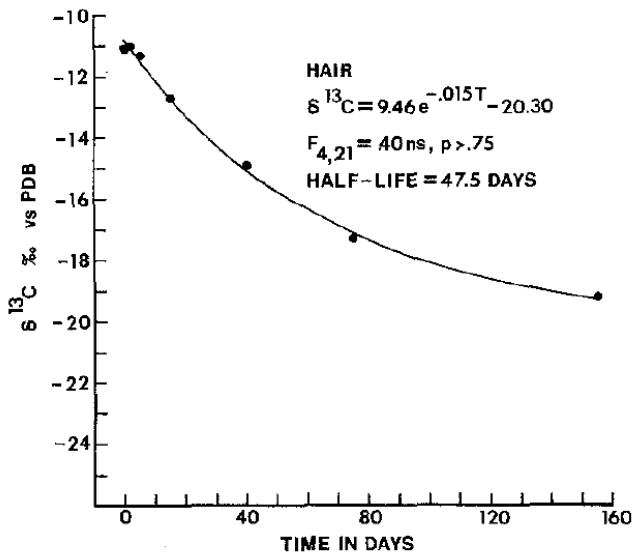
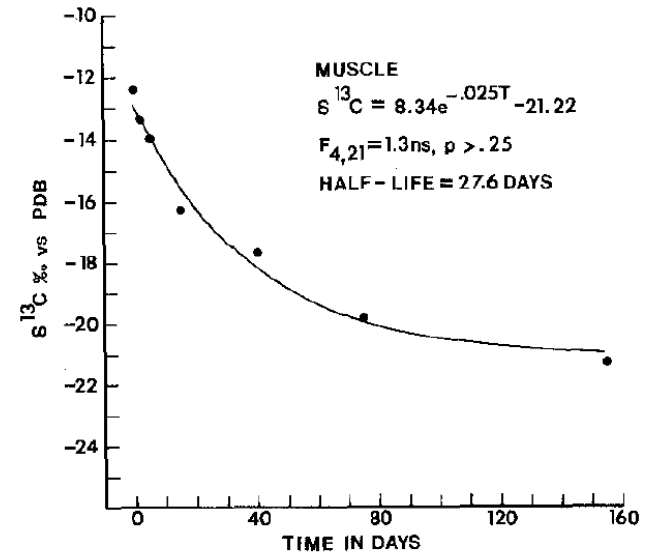
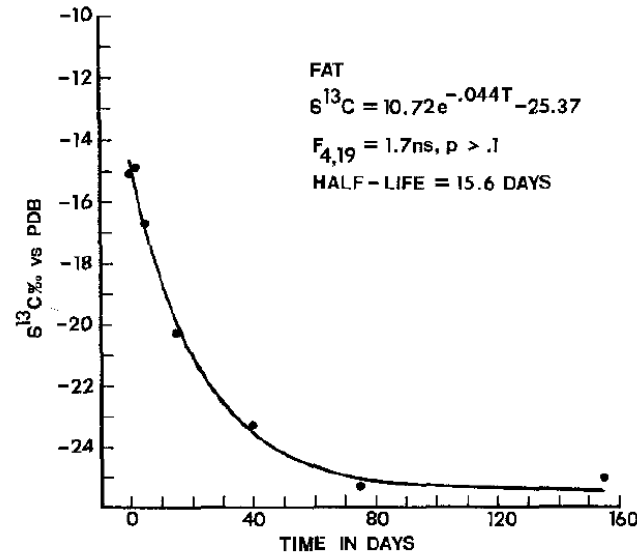
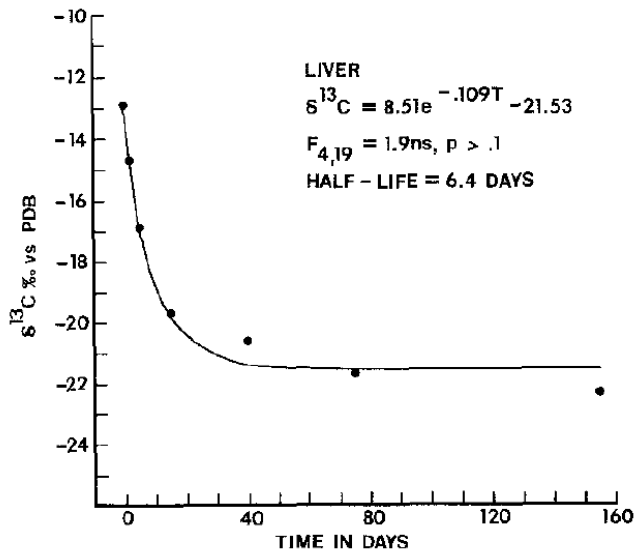
$r = r_i$  = fractional rate of isotopic incorporation  
flux into the pool / size of the pool

If animals are at steady state, then  $r = r_i = r_o$   
If animals are growing, then  $r = r_{\text{growth}} + r_{\text{metabolism}}$



"Half-Life"  
 $t_{1/2} = \text{Ln}(2)/r$

# Isotopic Incorporation Origins



*Meriones unguiculatus*

# Different Tissues Have Different Incorporation Rates



*Cortunix japonicus*

	1/r	Half-Life (days)
Plasma or Liver	3.7	2.5
Whole Blood	15.9	11.1
Muscle	17.6	12.2
Bone Collagen	250	173.3

The isotopic composition of liver tells us what the bird ate over the past ~10 days; muscle over the past ~60 days.

It's useful to analyze more than one tissue!  
(plasma proteins versus red blood cells or muscle versus bone collagen)



# Isotopic Incorporation Scales with Body Size

If  $r = \text{flux}/\text{pool}$ , allows us to make an allometric prediction:

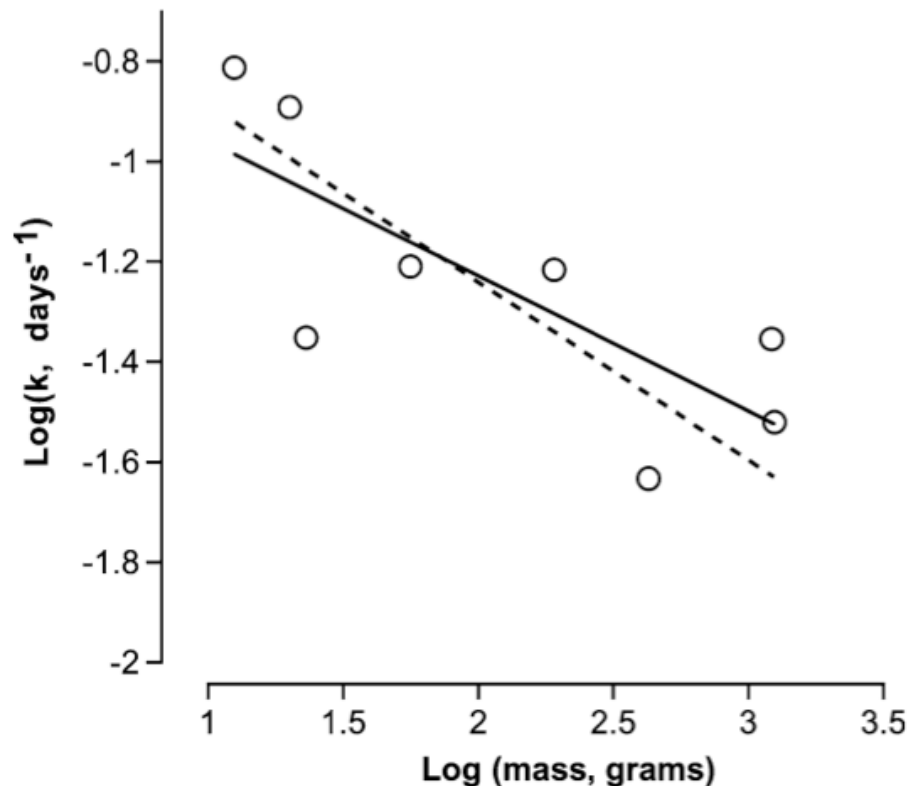
Pool (A) = amount of element in an animal is a function of its mass

If we know A, then we can estimate F (flux)

$$F = \text{flux} \propto \text{Mass}^{0.75}$$

$$\text{Thus } r = F/A \propto \text{Mass}^{0.75} / \text{Mass}$$

$$r = F/A \propto \text{Mass}^{-0.25}$$



From warblers (10g) to ducks (1100g):  
exponent of relationship does not differ  
from -0.25 for whole blood in birds.

Isotopic Incorporation is faster in  
smaller animals than in large beasts.  
**(BUT WE NEED MORE DATA!)**

## Isotopic Half Life Scales with Body Mass<sup>-0.25</sup>

In big animals, even tissues that turnover relatively fast (e.g., blood) integrate ecological inputs over a long time periods.

For example, the half life of carbon in the blood plasma of a  
~10g warbler should be ~10 days  
~100kg ostrich should be ~90 days.



VS



# Does Isotopic Incorporation Scale with Metabolic Rate?

...*"turnover rates of carbon isotopes are related to metabolic rate."*  
(Voigt et al. 2003)



*"These turnover rates were low compared to the information available for birds and eutherian mammals, and probably relate to the typically low metabolic rates of marsupials."* (Klassen et al. 2004)





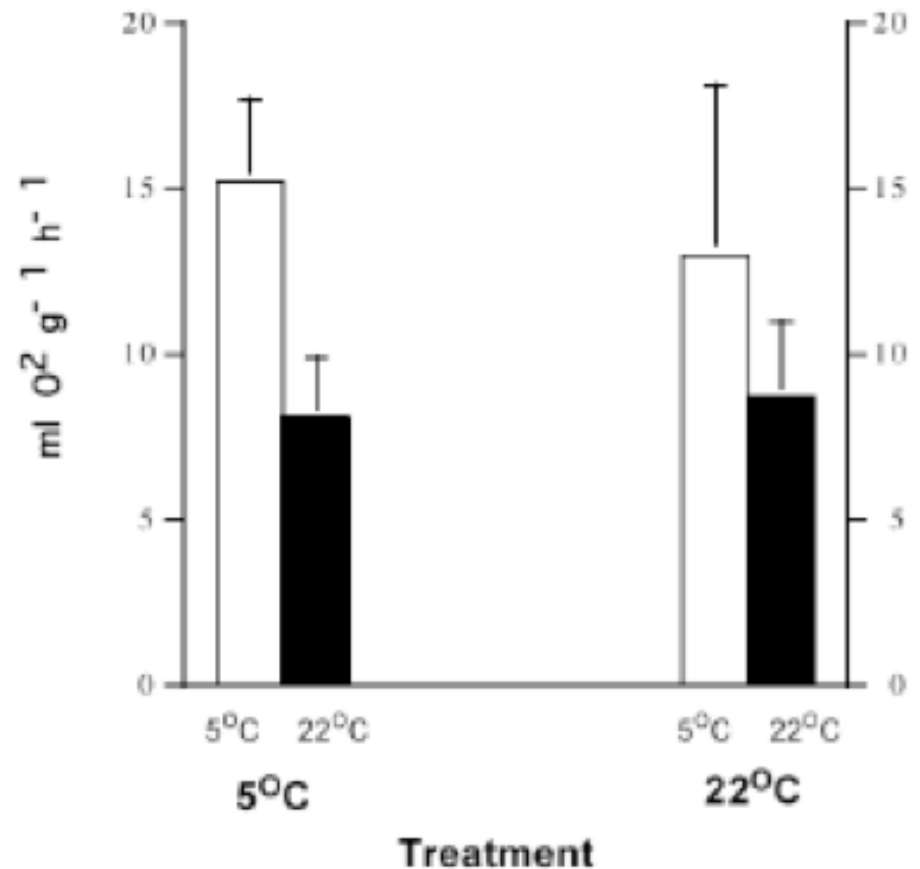
# Isotopic Incorporation Rate $\approx$ Metabolic Rate?

60 days on a C<sub>3</sub> (wheat) diet  
( $\delta^{13}\text{C} = -25.5\text{‰}$ ,  $\delta^{15}\text{N} = 4.3\text{‰}$ )

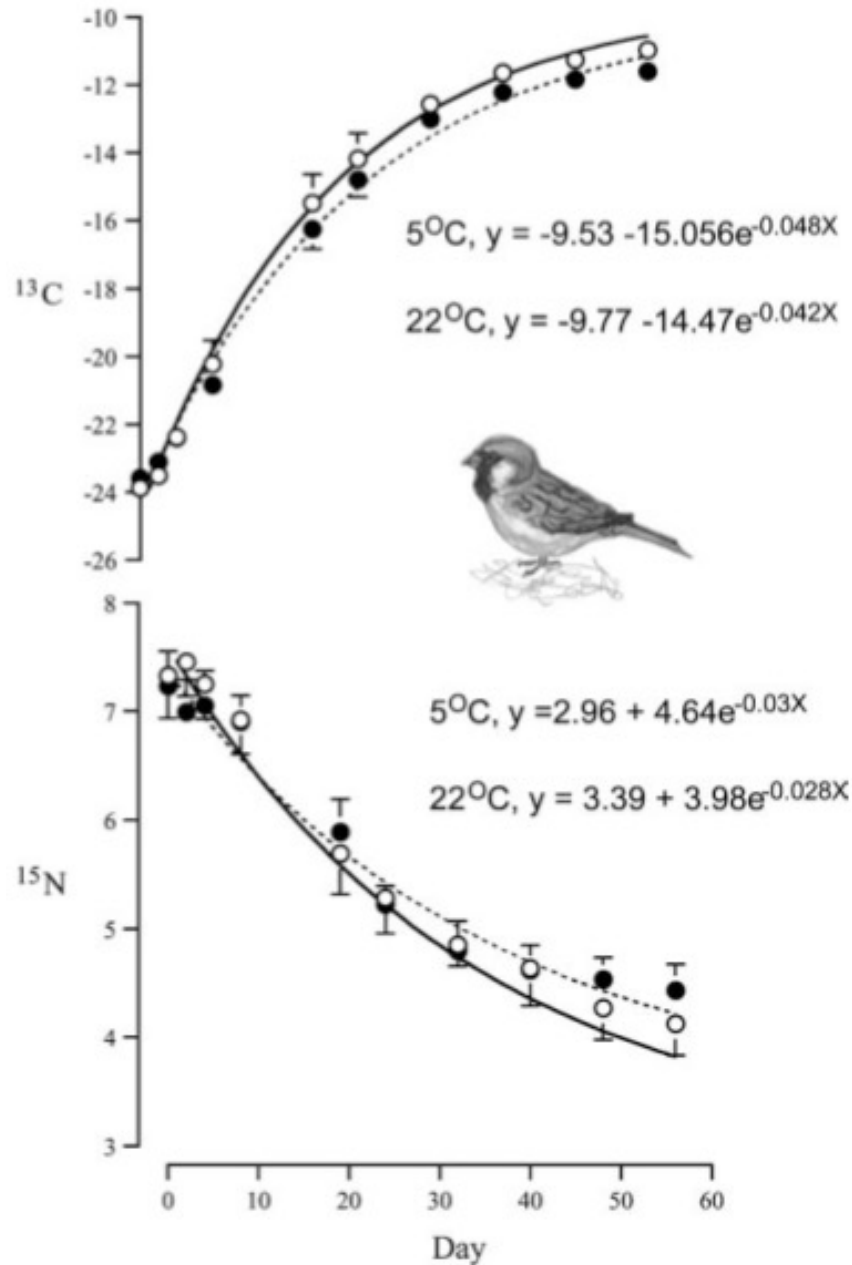


60 days on a C<sub>4</sub> (corn) diet  
( $\delta^{13}\text{C} = -11.5\text{‰}$ ,  $\delta^{15}\text{N} = 1.5\text{‰}$ )

House Sparrow Experiment:  
2 Treatments; 5°C and 21°C  
(2X change in metabolic rate)



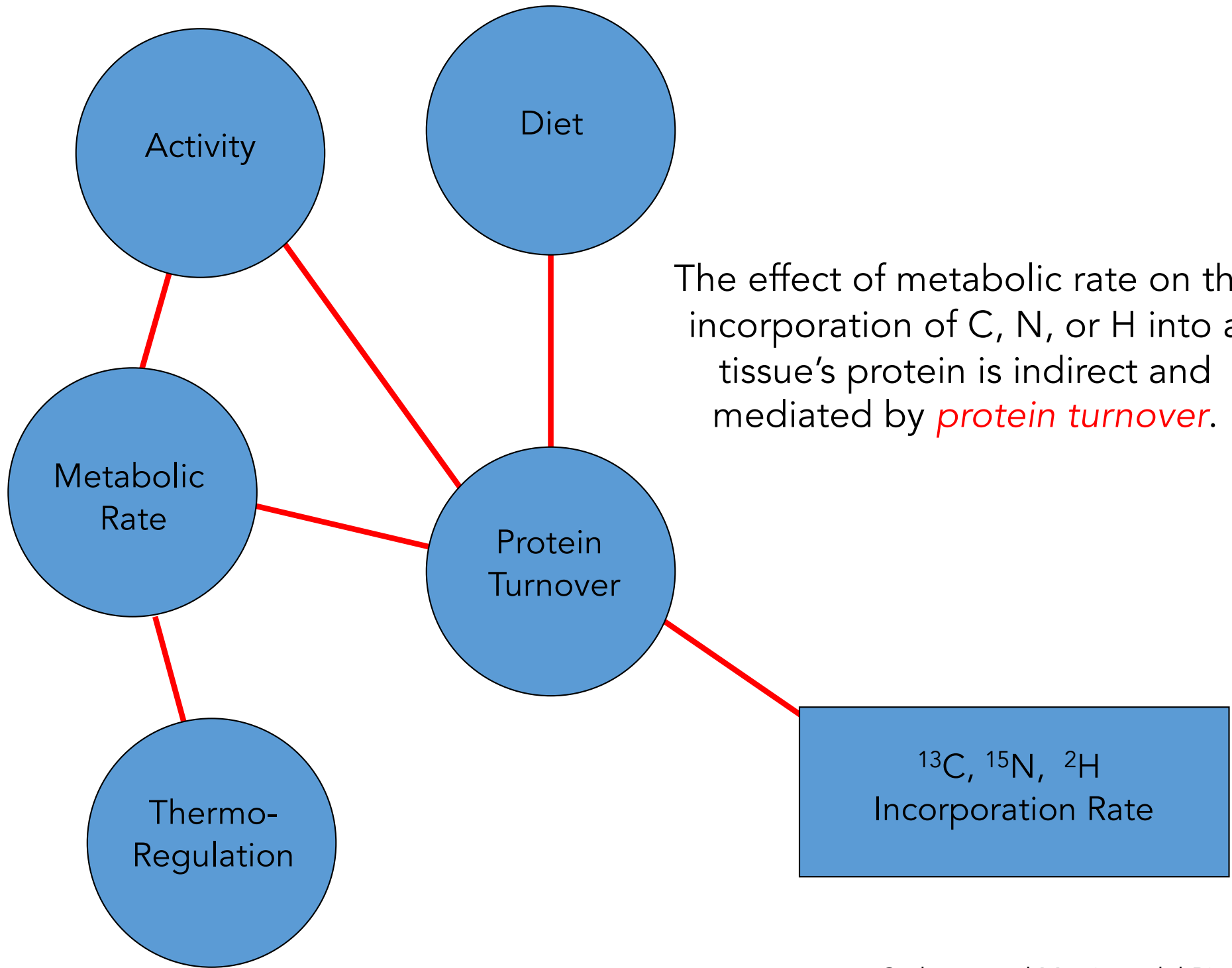
# House Sparrow Results



No significant effect of a cold-induced increase in metabolism on the rate of C and N incorporation.

We can uncouple the relationship between metabolic rate (MR) and incorporation rate (r)

What factor(s) (besides body mass) drive isotopic incorporation?



The effect of metabolic rate on the incorporation of C, N, or H into a tissue's protein is indirect and mediated by *protein turnover*.