Lab 7: Energy and Respiration

April 10 & 11, 2013

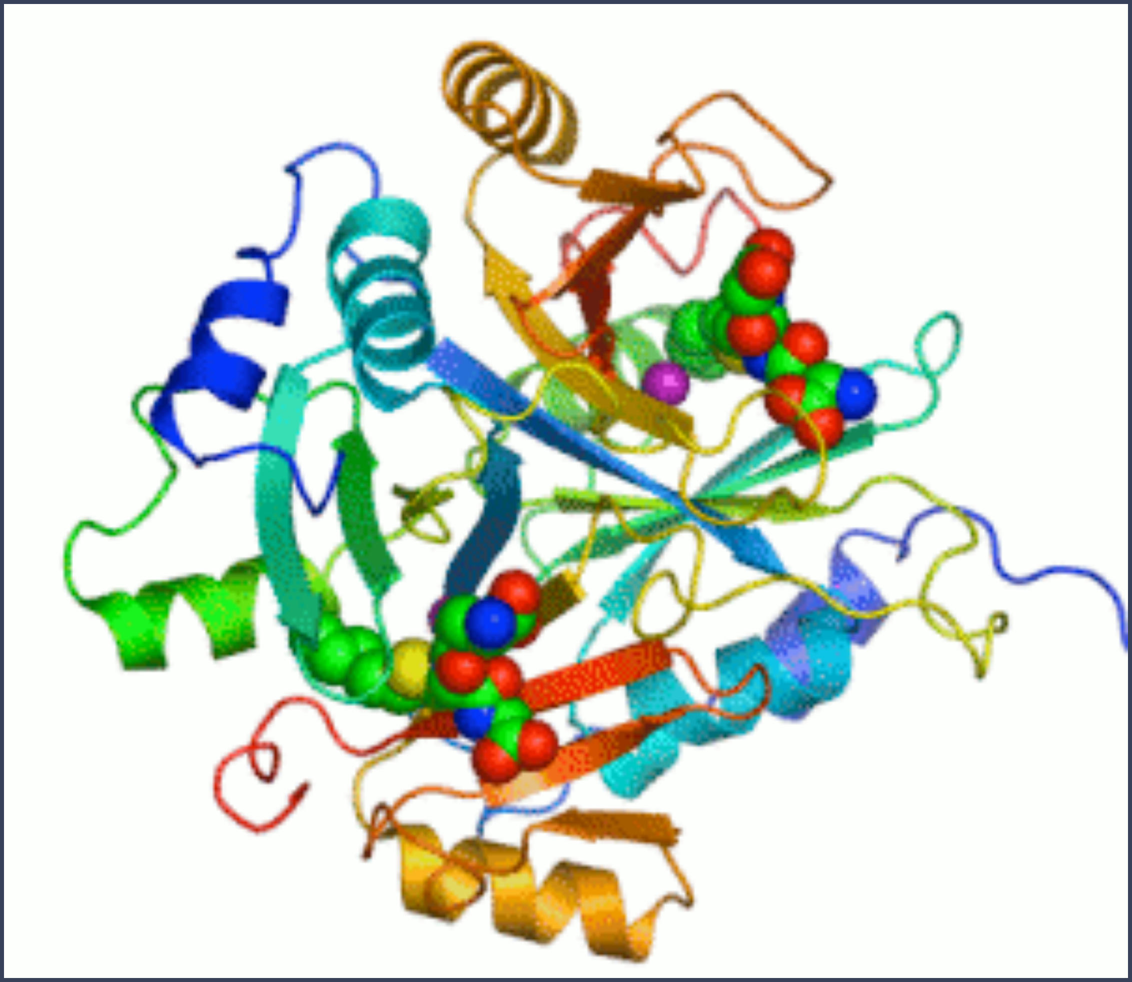
Lab Goals

* Think quantitatively
  * Enzyme kinetics

* Understanding at one level informs understanding at other levels
  * Scaling up

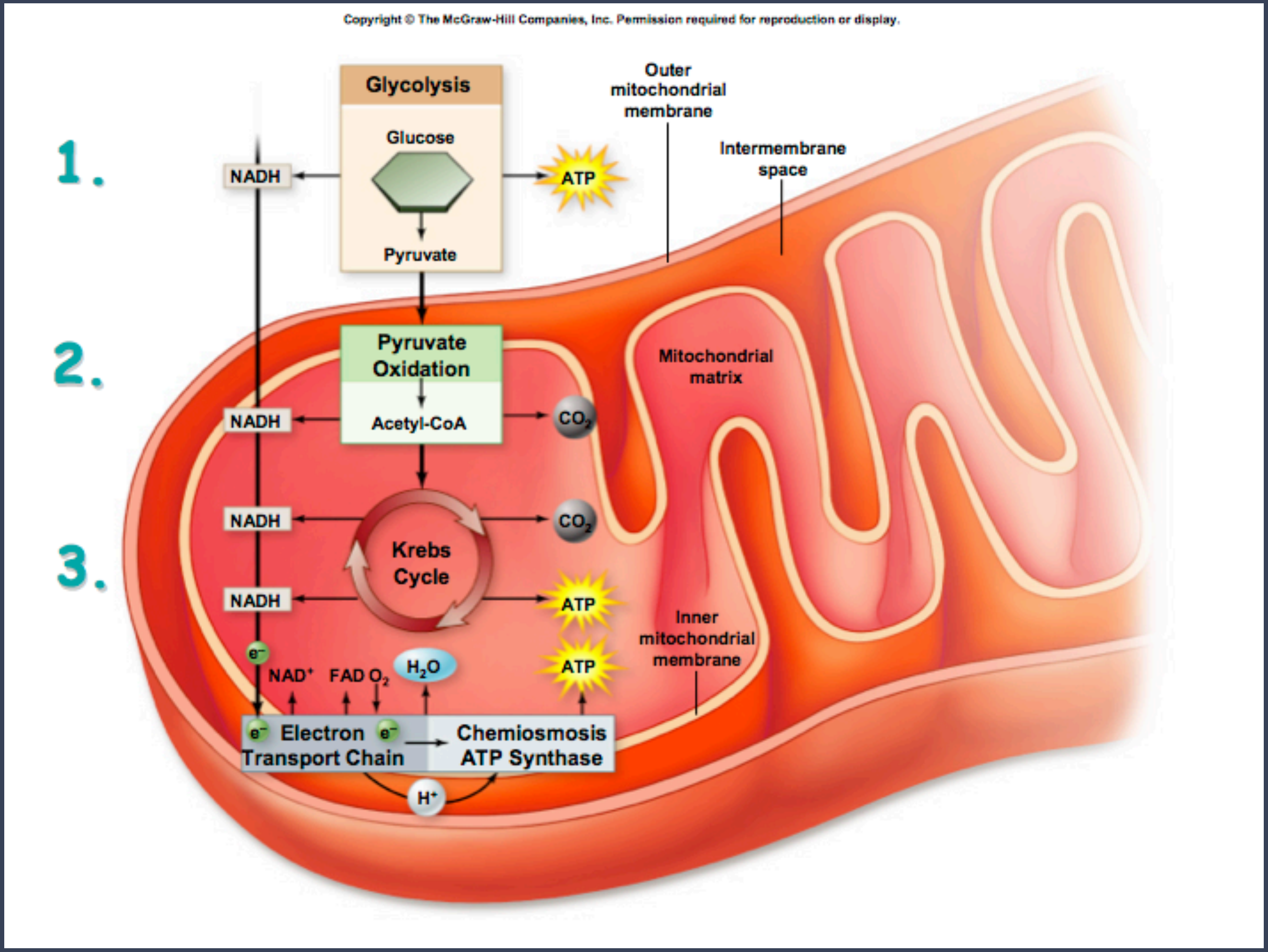
* Consider environmental influences
  * Variation
  * Global change

Section I. Enzymes



Respiration

$C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O + \text{Energy}$



Enzymes are proteins that accelerate chemical reactions

A typical enzyme (E) converts a substrate (S) into a product (P):

$$S + E \rightarrow E + P$$

However the rate at which S can be converted to P has a limit.

Factors Affecting Enzymatic Reactions

Factors Affecting Enzymatic Reactions

* Concentration of substrate(s)
* Concentration of enzyme
* pH
* Temperature
* Presence and/or concentration of inhibitor(s)
* Concentration of cofactors(s)

Enzyme Kinetics

Leonor Michaelis
1875-1949

Maud Menten
1879-1960

Michaelis – Menten equation:

$$v = \frac{V_{\max} S}{K_m + S}$$

v = velocity of reaction

V_{max} = maximal velocity of a reaction

S = substrate concentration

K_m = substrate concentration @ ½ Vmax

Experiment 1a: Soybeans

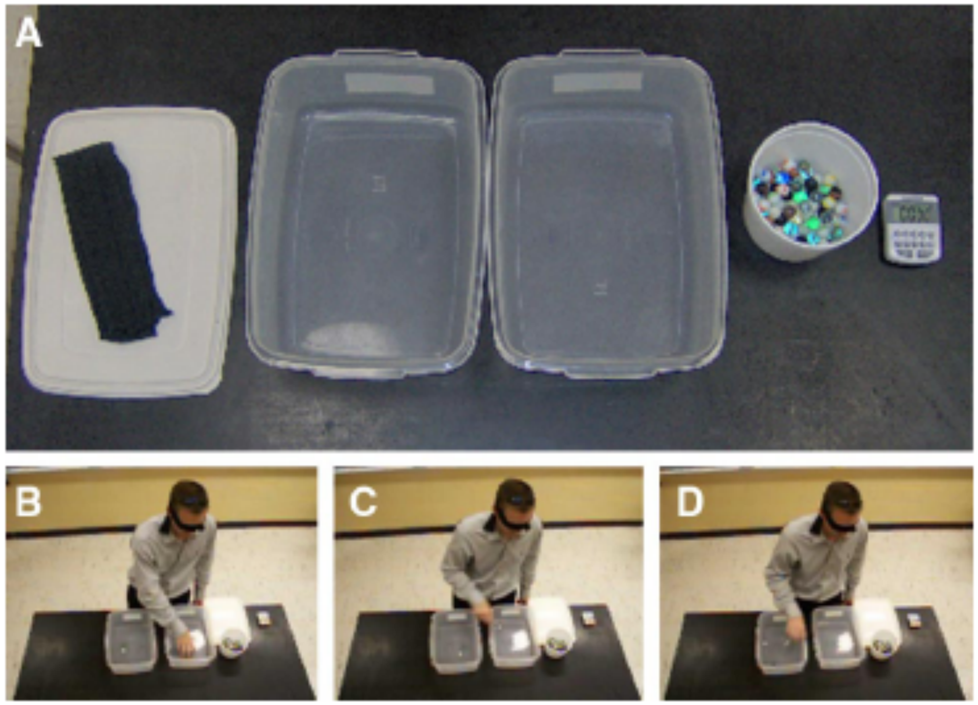


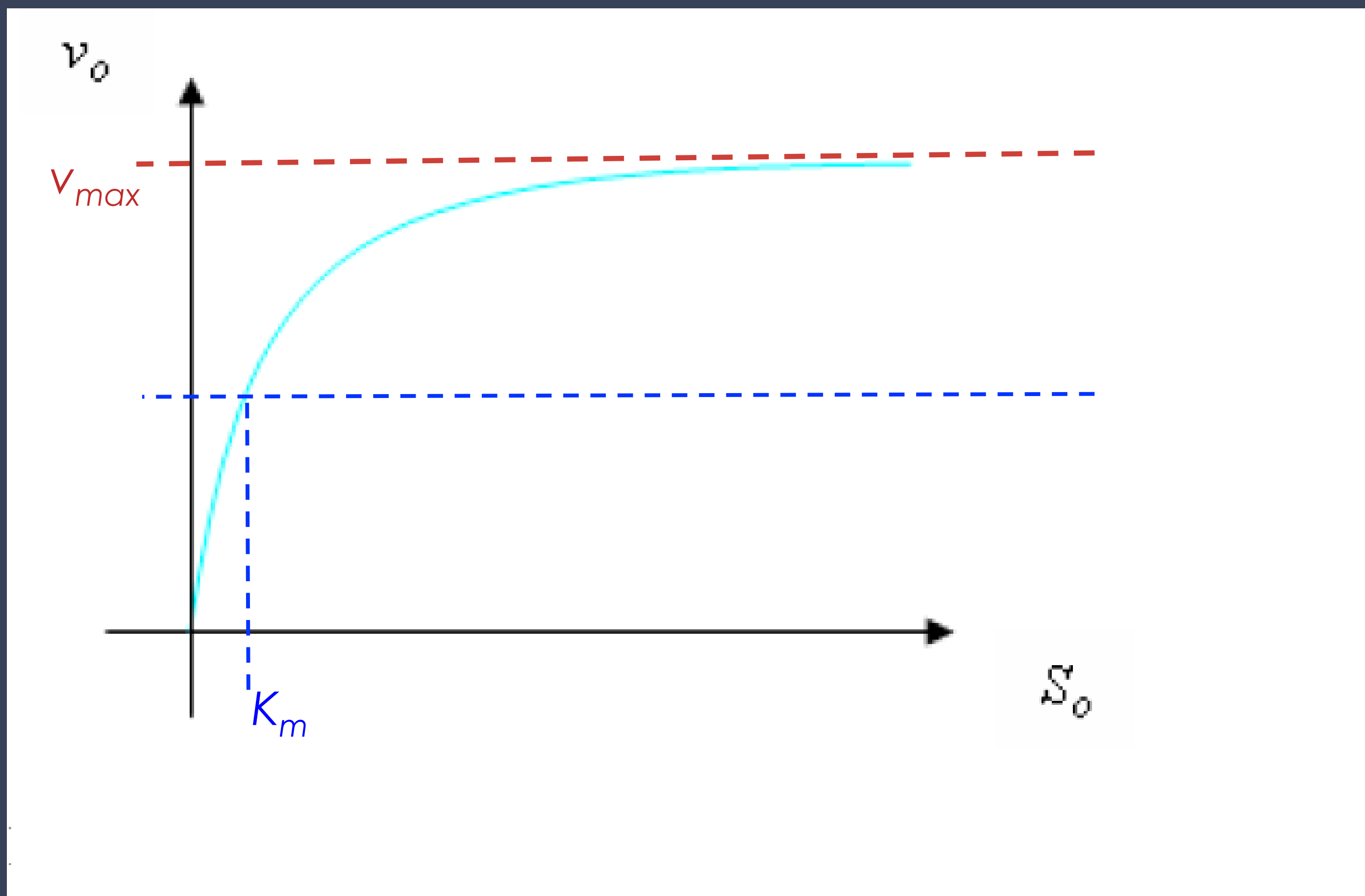
Figure 1. The experimental system. (A) Materials needed include a timer, marbles, two plastic containers, and a blindfold. (B–D) Photographs depicting a student performing one 10-s "run" of the exercise. (B) Finding a marble in the "substrate" container. (C) Transferring a marble to the "product" container. (D) Releasing the marble into the "product" container. The 13-s video of the student performing the exercise is available (see Supplemental Material).

Runge et al. 2006, CBE Life Science Education

V_{max} Describes how high the line goes

K_M Describes how curved the line is

$$v = \frac{V_{max} S}{K_m + S}$$



Steps

$$v = \frac{V_{\max} S}{K_m + S}$$

1. Hypothesize about how changing K_m and V_{max} influence the rate of a reaction (v)

2. Test your hypothesis graphing the equation in Excel.

Experiment 1b: Factors controlling reaction rates

We will consider how the hexokinase reaction rate changes after changing substrate levels and $V_{max.}$

Blood glucose levels in humans = 5 mM (before chocolate) to 12 mM (after chocolate)

Basal glucose transporters can uptake 1 mM glucose (K_m)

Let's look at how the reaction rate changes if:

a. K_m = 1 mM (Glucose); V_{max} = 15 reactions s^{-1}

Experiment 1b: Factors controlling reaction rates

Let's make our predictions:

* How will the reaction rate be affected by increasing/ decreasing K_m and V_{max}?

In Excel, calculate and observe how the reaction rate changes if:

b. K_m = 0.5, 1, and 2 mM; V_{max} = 15 reactions s^{-1}
c. K_m = 1 mM; V_{max} = 7.5, 15, and 30 reactions s^{-1}

Section II. Forest Tree Respiration and Climate Change

Forest tree respiration

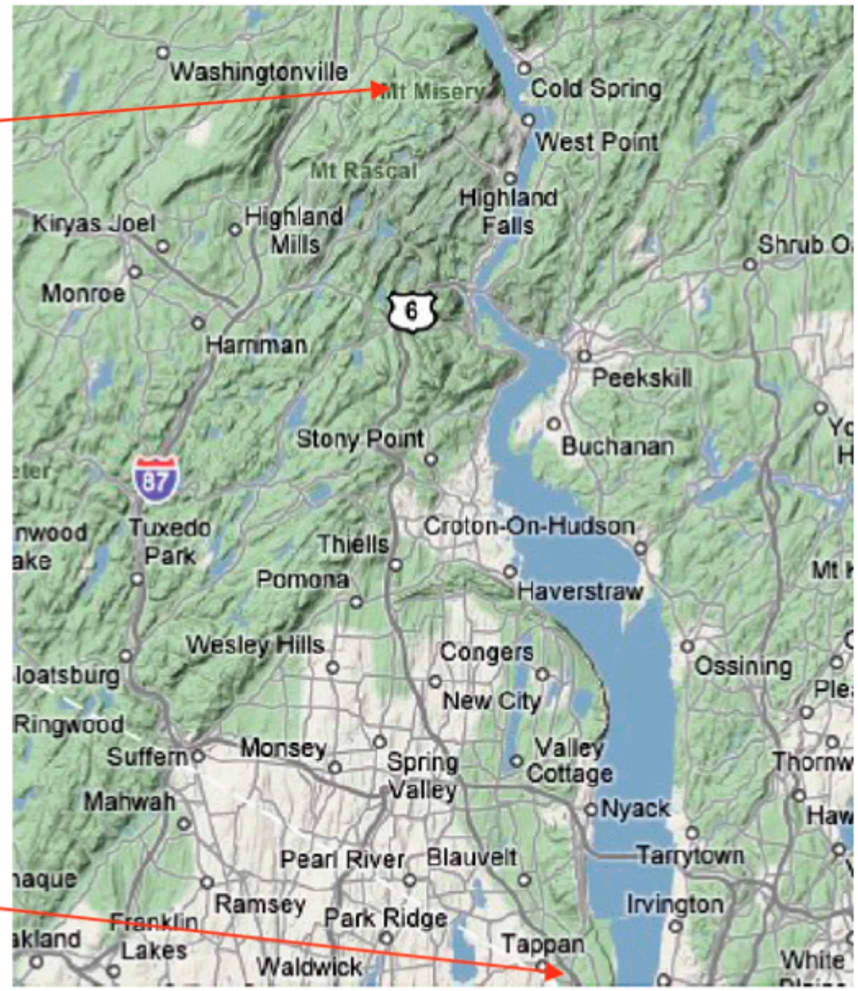
$C_6H_{12}O_6 + 6O_2 + 6H_2O \rightarrow 6CO_2 + 12H_2O + \text{Energy}$

Respiration is enzyme mediated, sugar consuming and energy producing.

It is strongly controlled by temperature but the temperature response can vary between species.

Black Rock Forest

www.blackrockforest.org



LAMONT DOHERTY EARTH OBSERVATORY
THE EARTH INSTITUTE AT COLUMBIA UNIVERSITY

http://blackrockforest.org/

Respiration can be measured instantaneously on a small scale.

How can we scale this up in space and time?

Steps

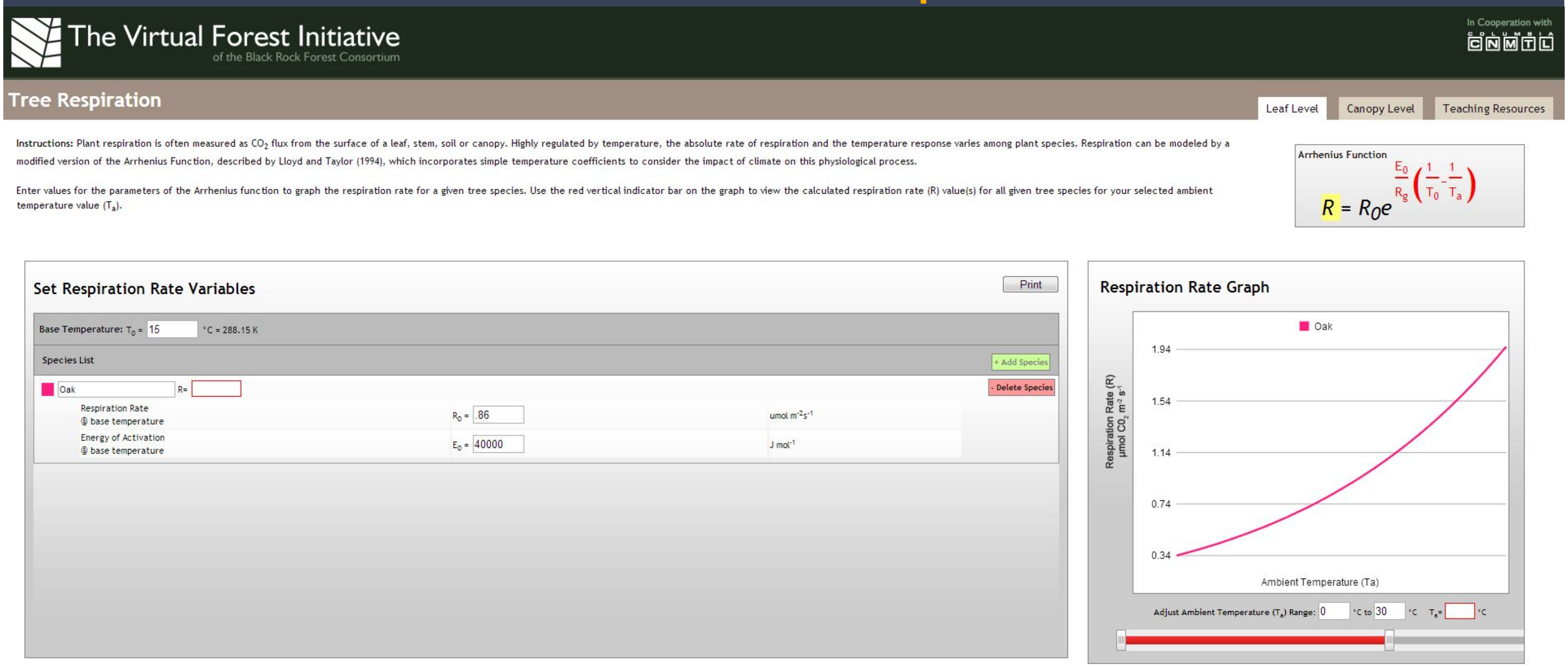
1. Use the respiration app. to evaluate the response of respiration of oaks and maples to temperature change: http://blackrock.ccnmtl.columbia.edu/ **USE FIREFOX!**

2. Use the **leaf-level** tab to evaluate the effect of changes in input variables on tree respiration **(Experiment 2a)**

3. Use the **canopy-level** tab to evaluate changes in seasonal respiration in the forest canopy **(Experiment 2b)**

4. State and test hypotheses on the relationship between input variables and respiration **(Experiment 3)**

Respiration Module
Leaf Level Respiration



Arrhenius function

$$R = R_0 e^{\frac{Eo}{R_g}\left(\frac{1}{T_o} - \frac{1}{T_a}\right)}$$

- R = Respiration
- R_g = *Ideal gas constant*
- R_0 = Base respiration, measured at the temperature T_0
- E_0 = Energy of activation for respiration, measured at the temperature T_0
- T_0 = Base temperature
- T_a = Ambient temperature → WE ASSUME THIS IS LEAF TEMPERATURE

To scale from leaf to canopy level, what do we need to consider?

Leaf Area Index (LAI)

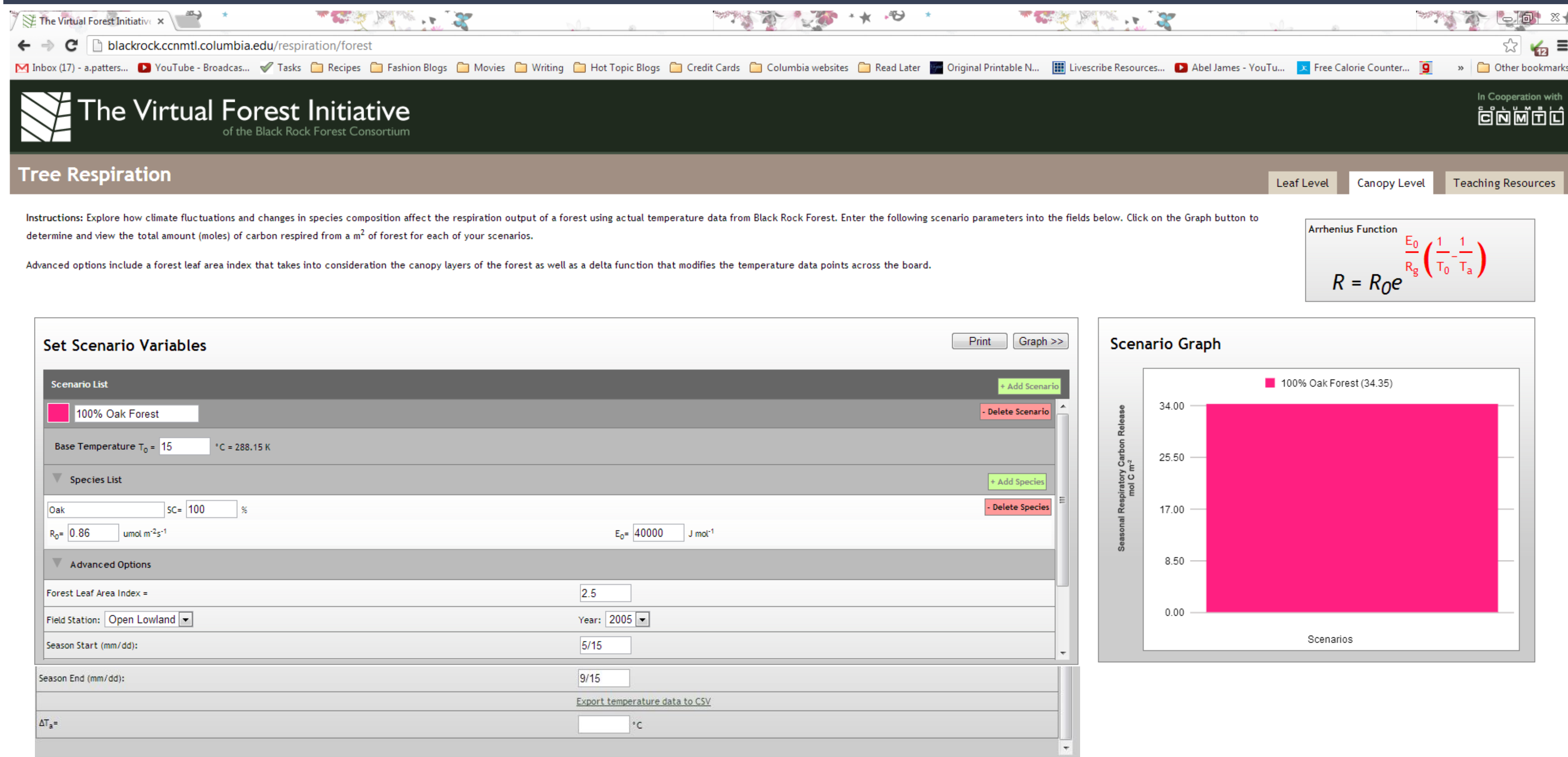
Ratio of leaf area to ground area

Expressed as m^2 leaf m^{-2} ground

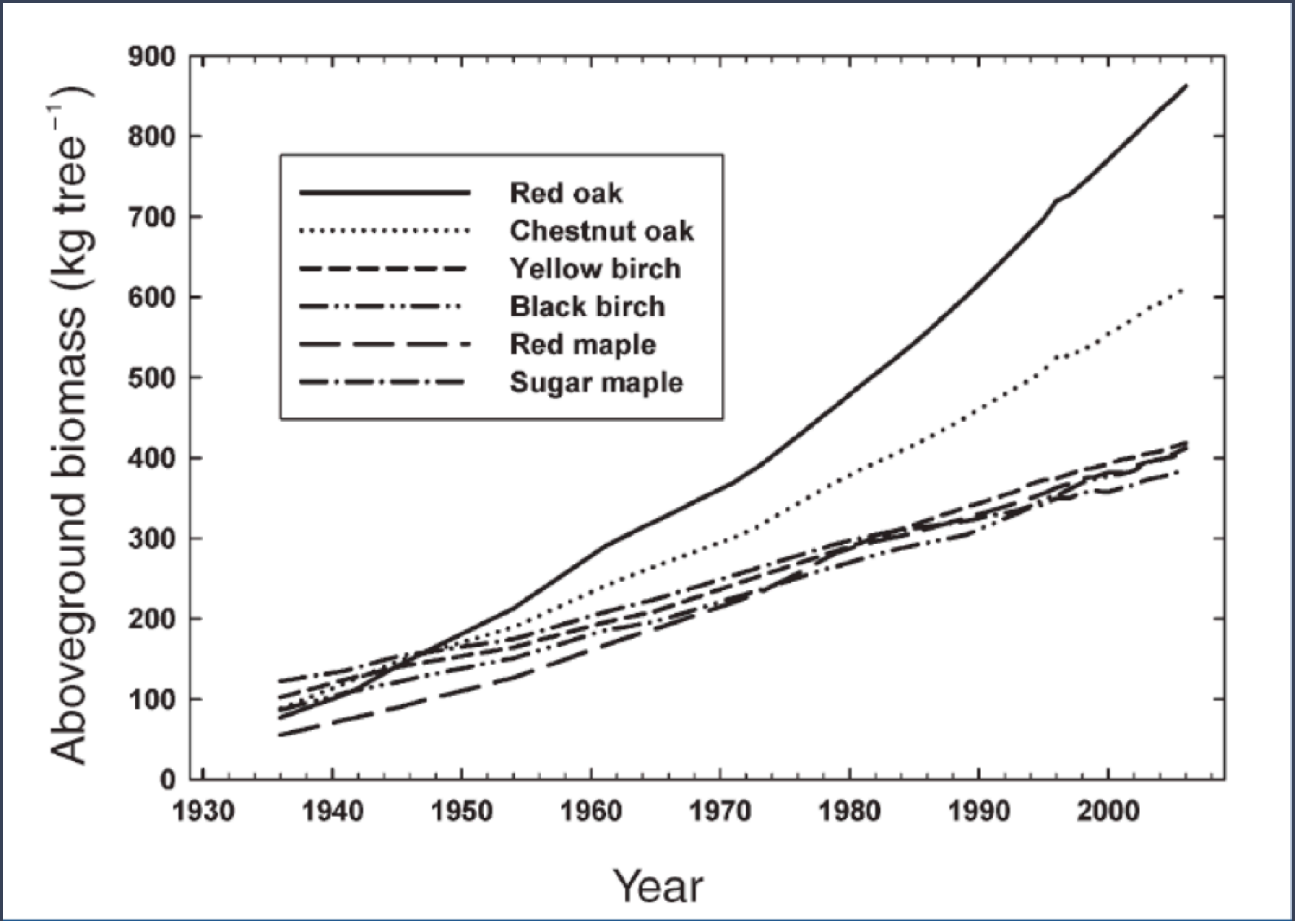
Ranges from 0 (bare leaves) to 15 (dense canopy) – BRF has an LAI of around 2.5

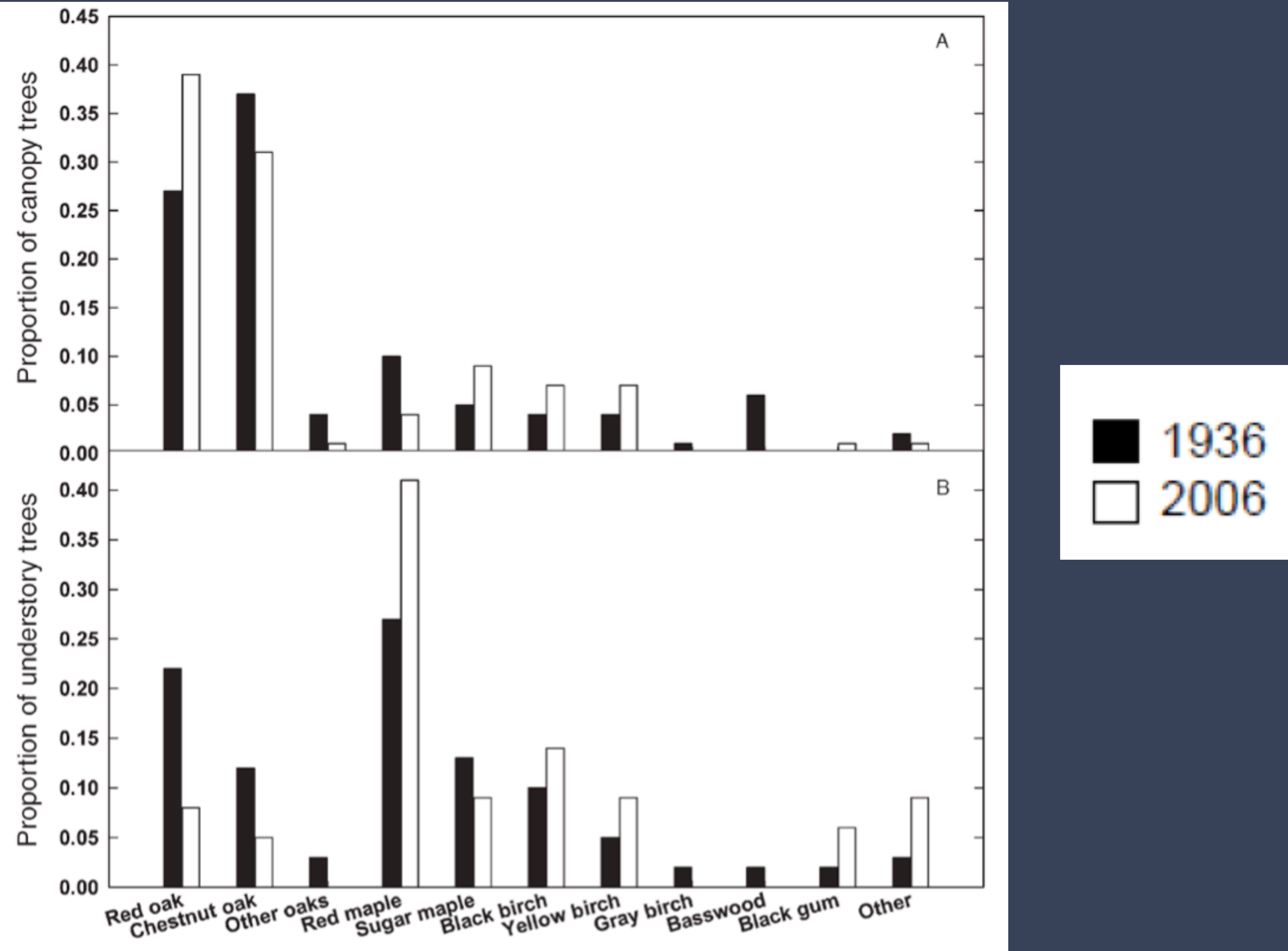
LAI plays important role in controlling interactions between terrestrial and atmospheric environmental variables

Respiration Module
Canopy Level Respiration

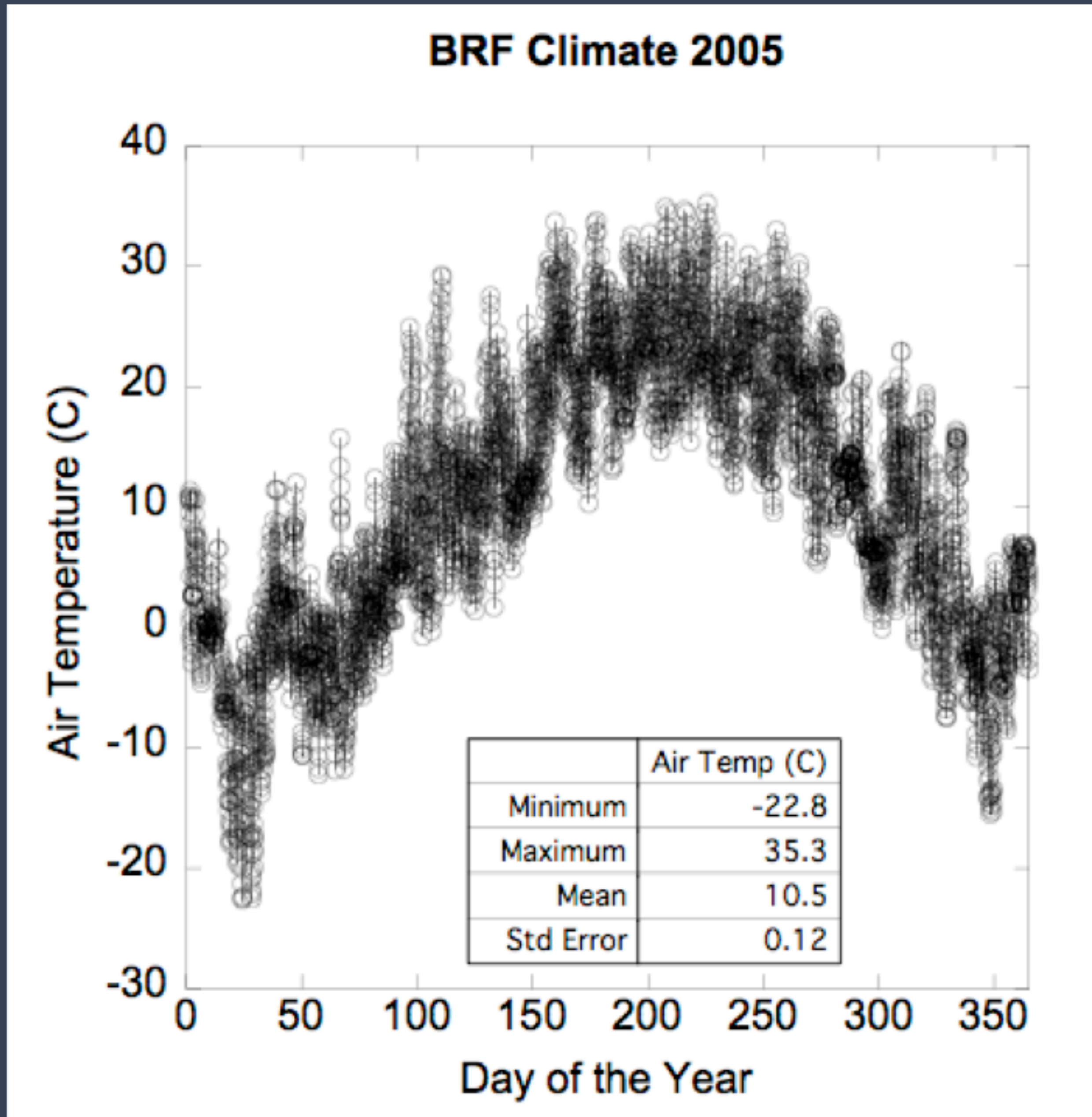


How might different tree species respond to climate change?





Proportion of canopy trees
Proportion of understory trees
A
B
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
Red oak
Chestnut oak
Other oaks
Red maple
Sugar maple
Black birch
Yellow birch
Gray birch
Basswood
Black gum
Other
1936
2006


BRF Climate 2005
Air Temperature (C)
40
30
20
10
0
-10
-20
-30
0
50
100
150
200
250
300
350
Day of the Year
Air Temp (C)
Minimum -22.8
Maximum 35.3
Mean 10.5
Std Error 0.12