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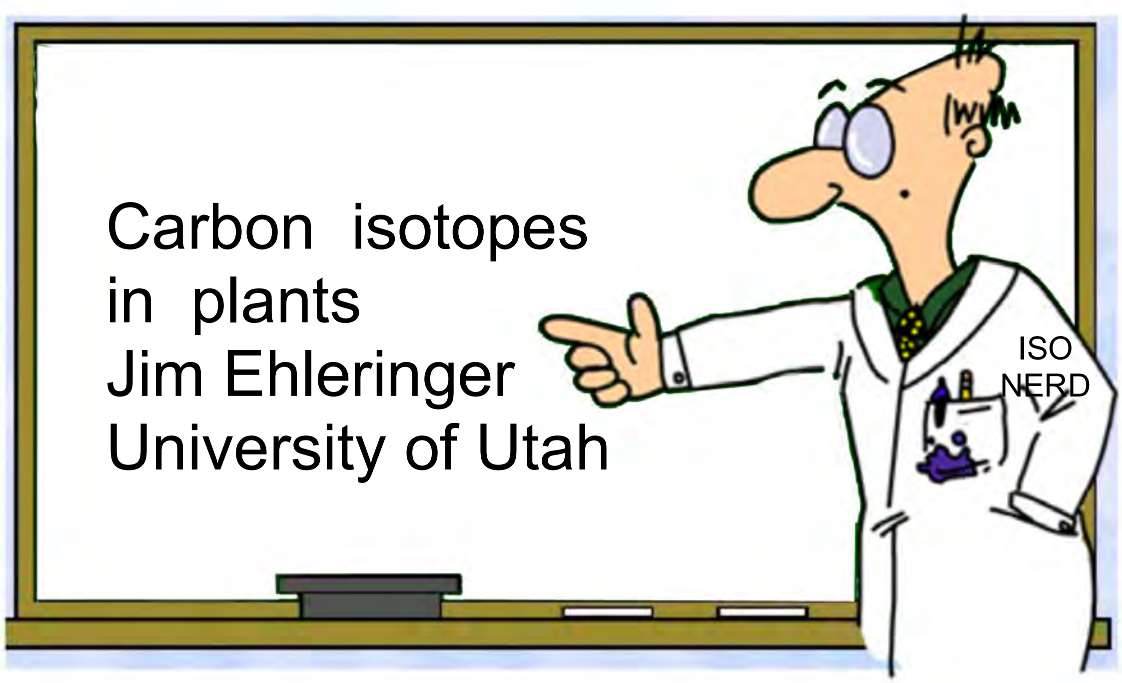


Carbon isotopes in plants

Jim Ehleringer

University of Utah

Goals

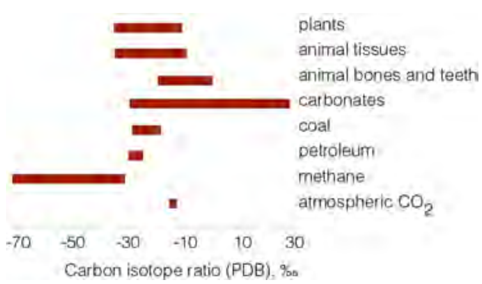
Goal 1
To establish a mechanistic foundation for understanding variations in the abundances of carbon isotopes in plants.

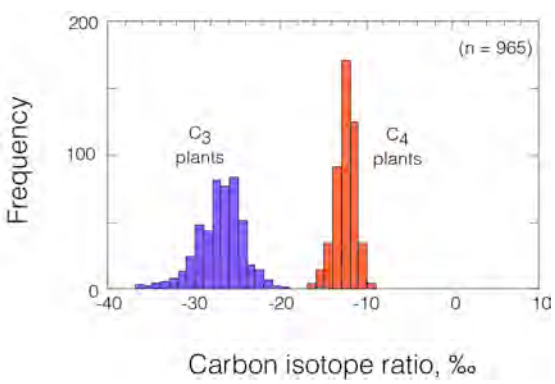
Goal 2
To predict patterns from a first-principle's basis and understanding of biochemistry and physiology.

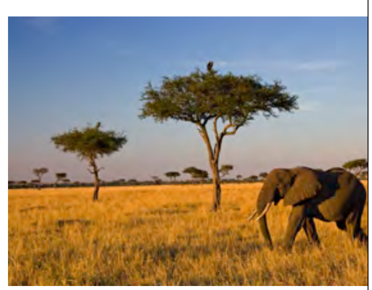
Background #1

What is the typical range of δ values for plant tissues?

-32‰ to -10‰ V-PDB scale



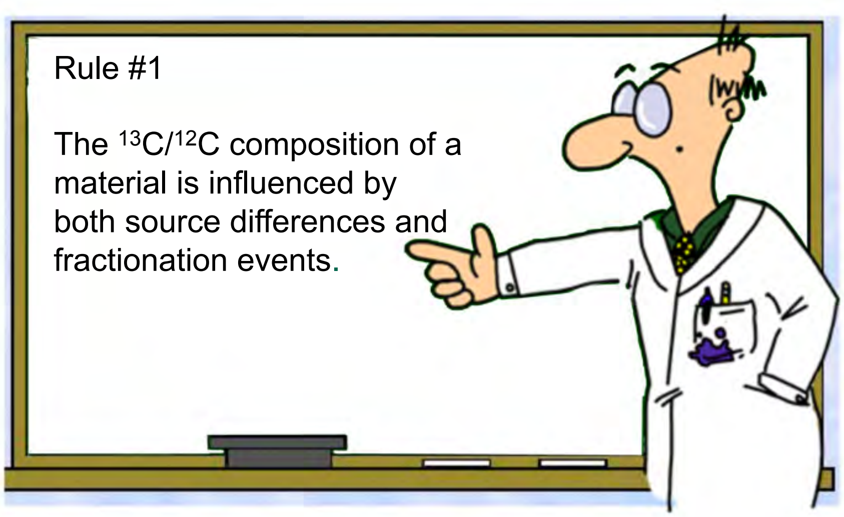




Background #2

Isotope effects in steps leading to CO_2 fixation in plants

Process	Isotope effect (α)	Discrimination	Symbol
Diffusion			
CO_2 diffusion in air	1.0044	4.4‰	a
CO_2 diffusion in water	1.0007	0.7‰	a_l
C_3 photosynthesis			
Rubisco fixation of CO_2	1.030	30‰	b_3
Net C_3 fixation with respect to c_i/c_a	1.027	27‰	b
C_4 and CAM photosynthesis			
Dissolution of CO_2 into water	1.0011	1.1‰	e_s
Hydration of CO_2 to HCO_3^- at 25°C	0.991	-9‰	e_b
Fixation of HCO_3^- by PEP carboxylase	1.0020	2.0	b_4^*



Why are there such large variations in the carbon isotope ratios of tomatoes?

tomato purchased from the store -45‰

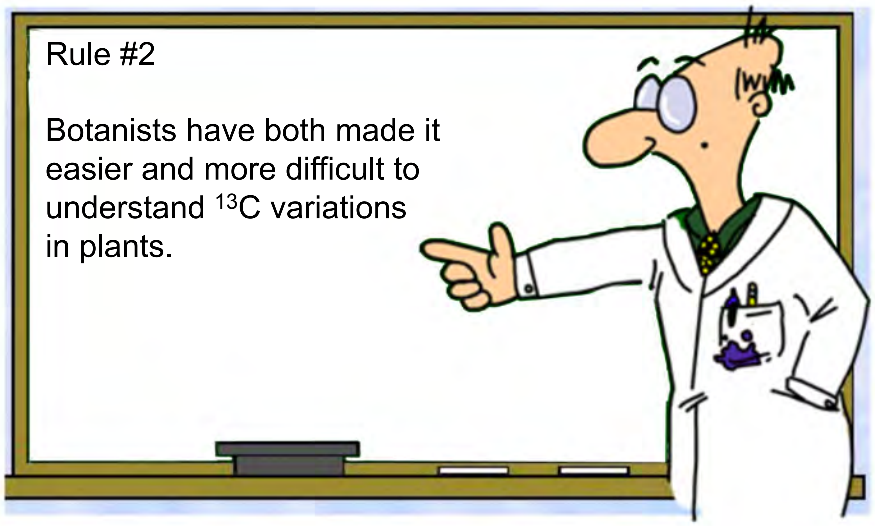
tomato grown in the field -25‰

Using this information, explain carbon isotope ratios of the plants we purchased from the nursery this year:

variegated strike -40.7‰

yellow marigold -34.9‰

zinnia -34.4‰



A traditional approach: 'delta' notation

$$\delta\ (‰) = (R_{plant}/R_{standard} - 1) \bullet 1000‰$$

$$\delta = \frac{R_{plant}}{R_{standard}} - 1$$

Plant biologists don't always use delta notation to describe isotope abundances

We then multiply by 1000 to get to familiar "per mil" unit

Defining isotope effect
as a discrimination - a la Farquhar et al.

Graham Farquhar

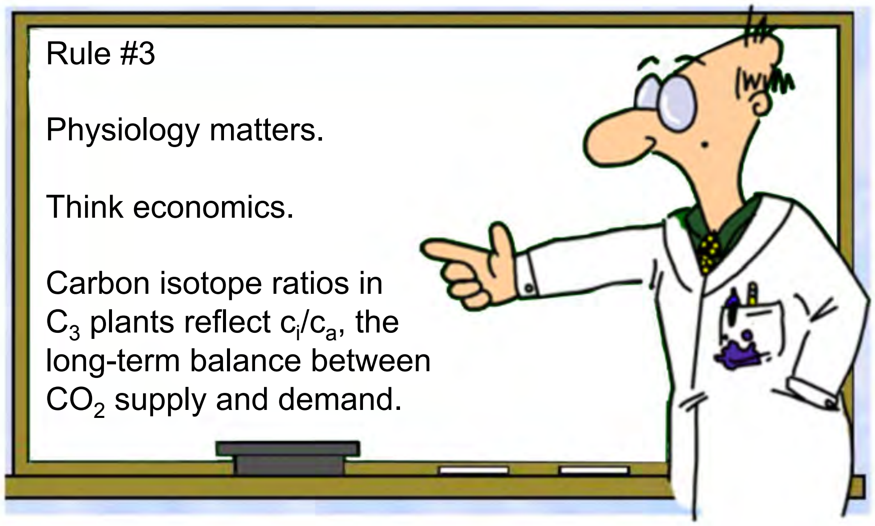
$$\delta = \frac{R_{plant}}{R_{standard}} - 1$$

$$\Delta = \frac{\delta_{air} - \delta_{plant}}{1 + \delta_{plant}}$$

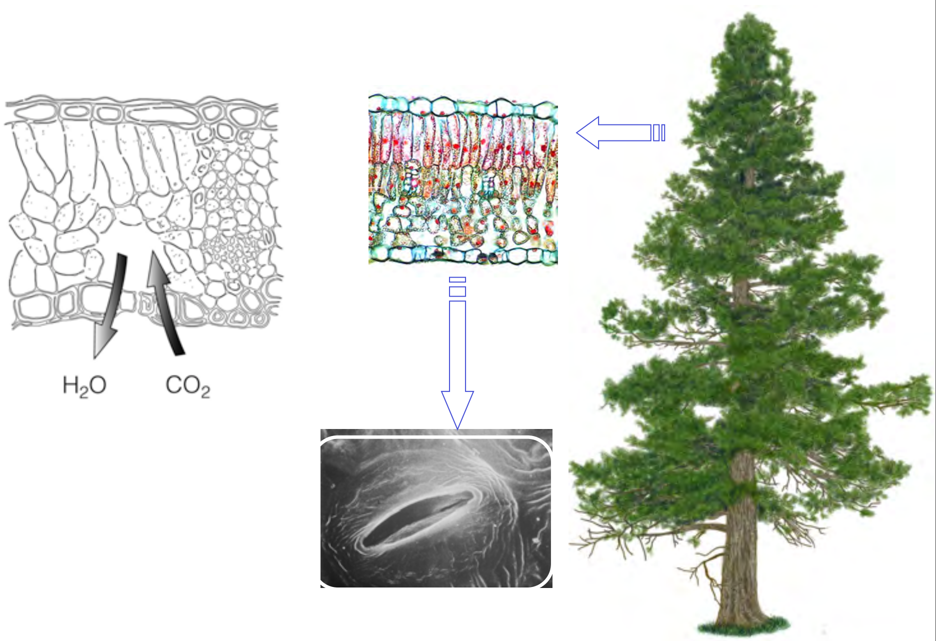
$$\alpha = \frac{R_{air}}{R_{plant}}$$

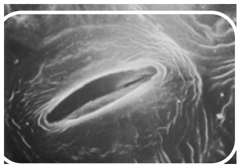
$$\Delta = \alpha - 1 = \frac{R_{air}}{R_{plant}} - 1$$

Questions?



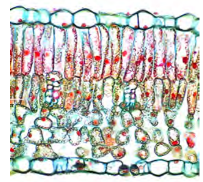
Carbon and water are interconnected at a common intersection- stomata

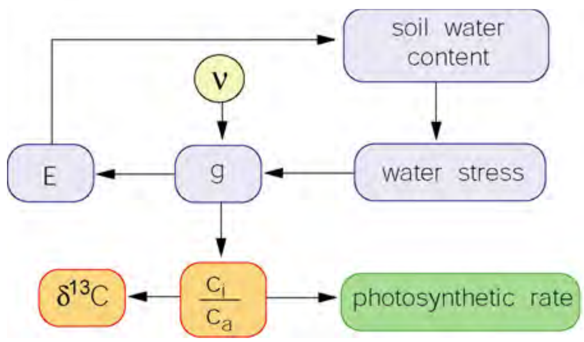




Carbon isotope ratio
is a long-term measure of a
metabolic set point

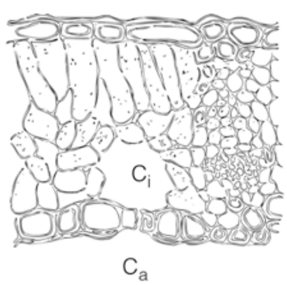
(the balance between CO_2
demand and supply)

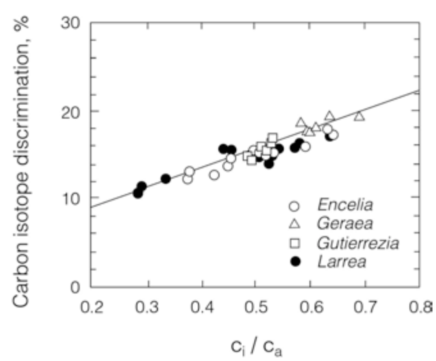




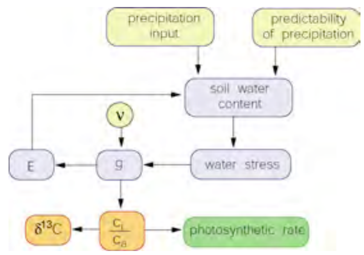
g = conductance
ν = leaf-to-air water gradient
E = transpiration

Carbon isotope discrimination in C_3 plants is driven by variations in stomata





$$\Delta = \underset{\downarrow \atop 4.4}{a}\frac{C_a - C_i}{C_a} + \underset{\downarrow \atop 27}{b}\frac{C_i}{C_a} = a + (b - a)\frac{C_i}{C_a}$$



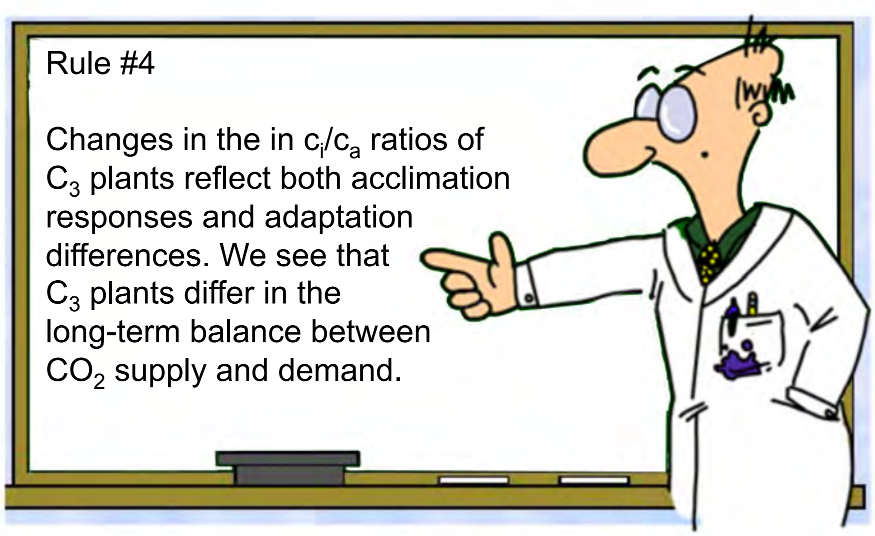
Metabolic set point as reflecting a balance between two opposing processes, each having both genetic and environmental controls

Consider some of the features associated with

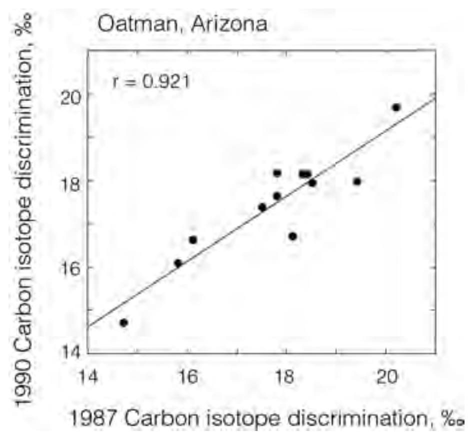
supply
- stomatal conductance
- hydraulic conductivity
- cavitation sensitivity
- rooting distribution

demand
- photosynthetic capacity
- nitrogen content
- leaf area index

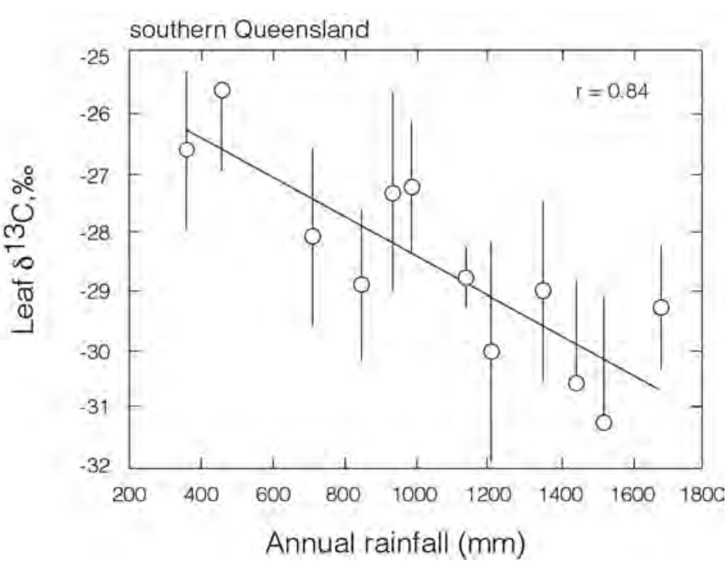
We should therefore expect to see many plant features correlated with leaf carbon isotope ratios.



Adaptation: rankings among genotypes remain fixed

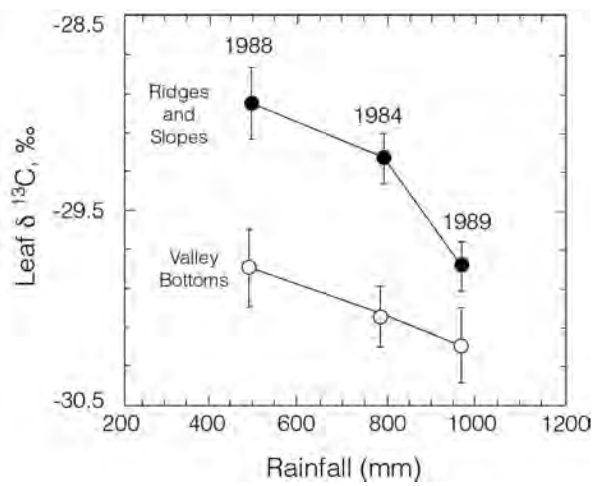


We observe a decrease in C_3 discrimination along precipitation gradients



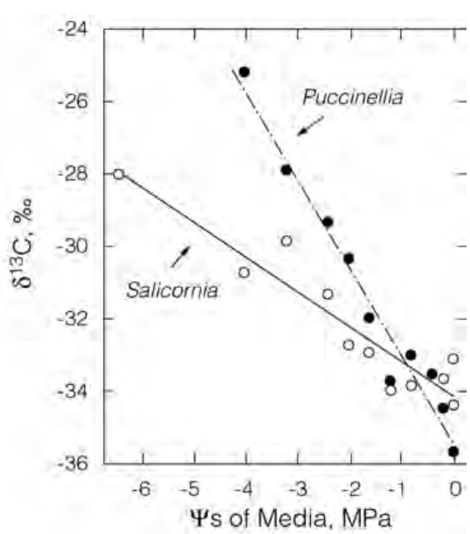
Stewart et al. (1995)

There is an adjustment response to current growth environment (acclimation), but ^{13}C rankings among plant species remain fixed.



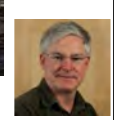
Garten and Taylor (1992)

Acclimation: C_3 plants discriminate less when exposed to water stress



high water stress

low water stress



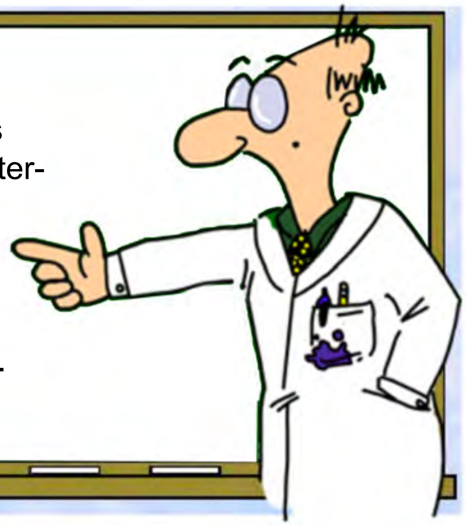
Guy et al. (1980)

Let's describe other ecological or environmental situations where plant carbon isotope ratios vary?

Questions?

Rule #5

Carbon isotope ratio does NOT directly measure water-use efficiency. Yet understanding ^{13}C is important for many agricultural breeding efforts and improvements.



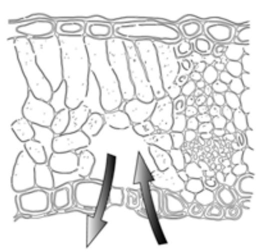
Water-use efficiency is the ratio of assimilation/transpiration (A/E)

$$A = (c_a - c_i)\frac{g}{1.6}$$

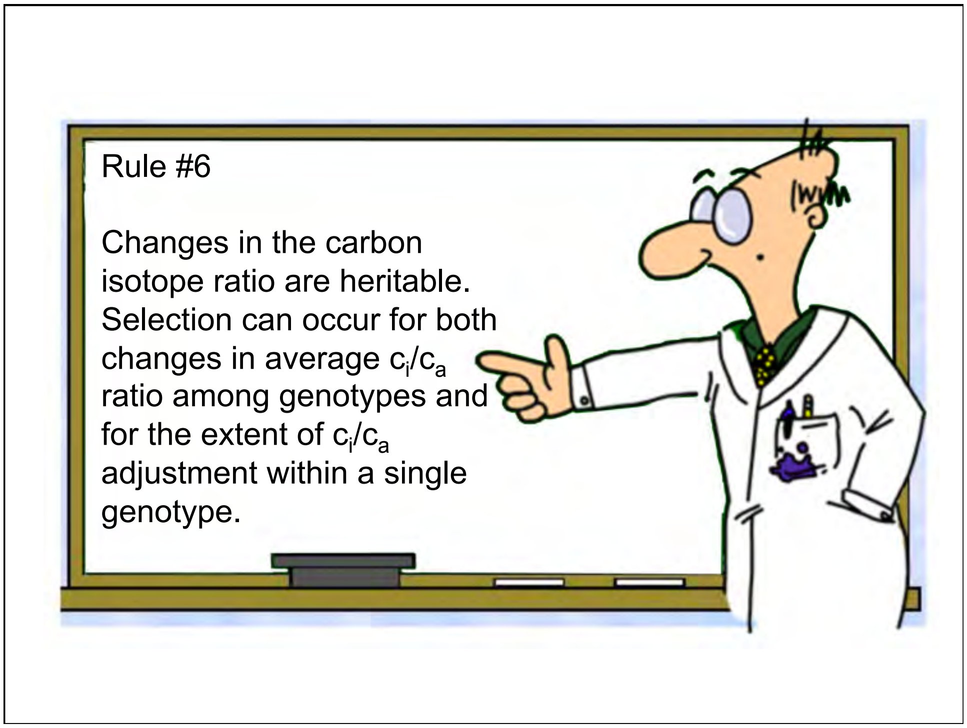
$$E = vg$$

$$\frac{A}{E} = \frac{(c_a - c_i)}{1.6v} = \frac{c_a\left(1 - \frac{c_i}{c_a}\right)}{1.6v}$$

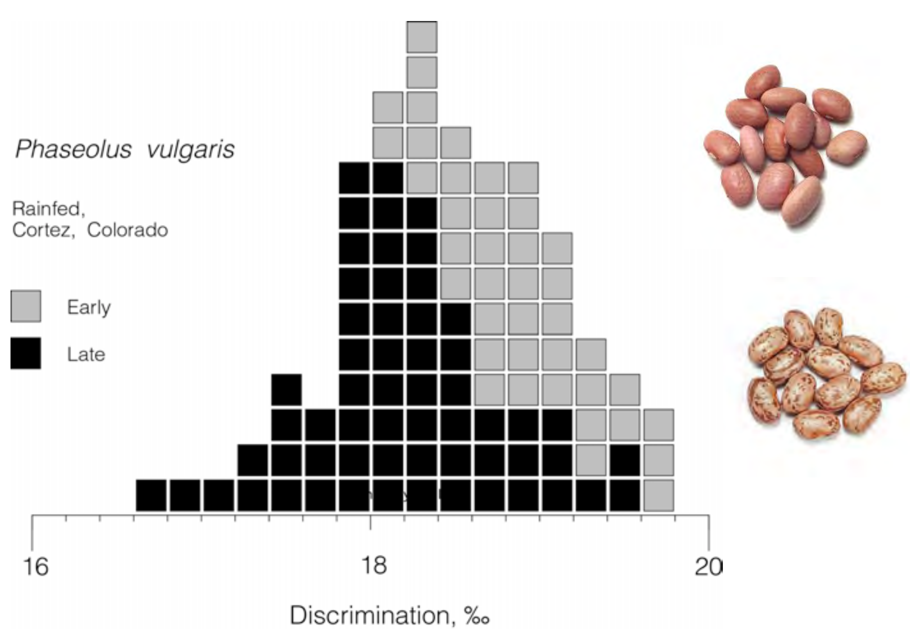
Leaf-to-air vapor pressure gradient (vpd) divided by P



intrinsic water-use efficiency $A/g = c_a \bullet (1 - c_i/c_a)/1.6$

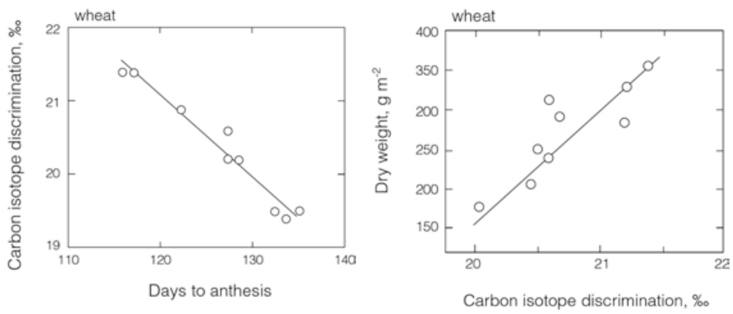


There is genetic variation in ^{13}C among agricultural genotypes



Genetic variation in ^{13}C exists and appears correlated with

- sensitivity to drought
- maturity date
- life expectancy
- biomass and growth rate
- leaf conductance




Richard Richards

Gutierrezia microcephala

Coleogyne ramioissima

cold desert

short - lived

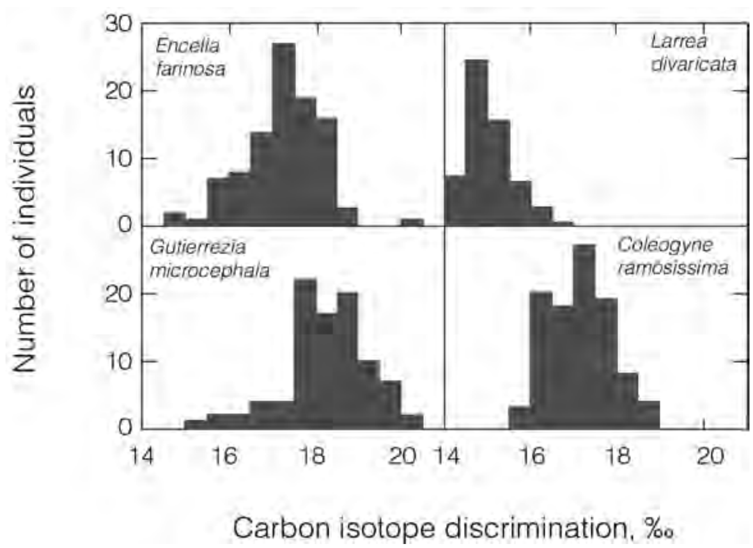
long - lived

warm desert

Encelia farinosa

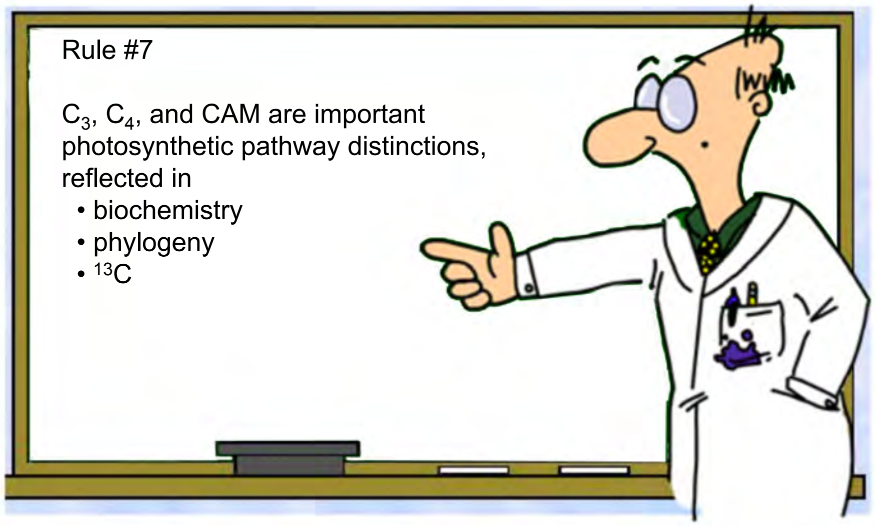
Larrea divaricata

There is genetic variation in ^{13}C among native-plant genotypes



Longer-lived species in an environment exhibit lower discrimination values

Questions?



Large ^{13}C differences occur between C_3 and C_4 photosynthetic pathway

C_3

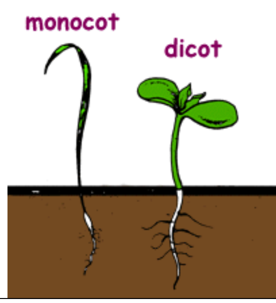
C_4

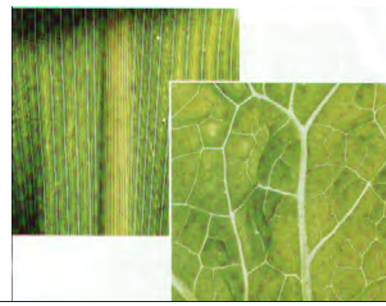
CAM

	% of all plants	% global productivity
C_3	> 95%	70-75%
C_4	< 5%	25-30%
CAM	< 1%	< 1%

Two primary groups within Angiosperms contain different abundances of C_3 and C_4 species

	C_3 species	C_4 species
Monocots	~ 6,000	~ 6,000
Dicots	~ 300,000	~ 2,000





C_4 plants are found only in the more derived Angiosperms

C_4 pathway appears to have evolved multiple, independent times

Families known to have C_4 photosynthesis

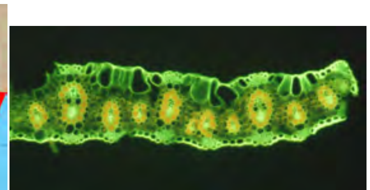
Acanthaceae	Cyperaceae
Aizoaceae	Euphobiaceae
Amaranthaceae	Nyctaginaceae
Asteraceae	Poaceae
Boraginaceae	Polygonaceae
Capparidaceae	Portulaceae
Chenopodiaceae	Scrophulariaceae
Cleomaceae	Zygophyllaceae

C_3 and C_4 species appear in a genus several times, suggesting multiple independent evolution events

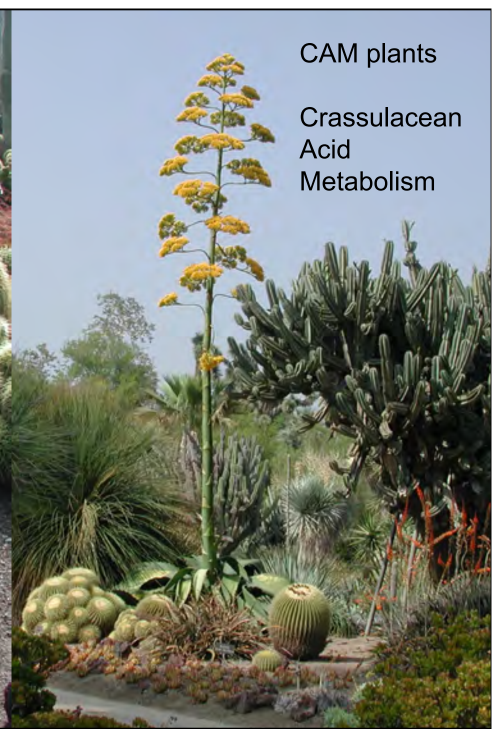
Family	Genus
Aizoaceae	*Mollugo*
Amaranthaceae	*Aerva, Alteranthera*
Asteraceae	*Flaveria, Pectis*
Boraginaceae	*Heliotropium*
Chenopodiaceae	*Atriplex, Bassia, Kochia, Suaeda*
Cyperaceae	*Cyperus, Scirpus*
Euphorbiaceae	*Chamaesyce, Euphorbia*
Nyctaginaceae	*Boerhaavia*
Poaceae	*Alloteropsis, Panicum*
Zygophyllaceae	*Kallstroemia, Zygophyllum*

Mollugo verticillata

Flaveria bidentis



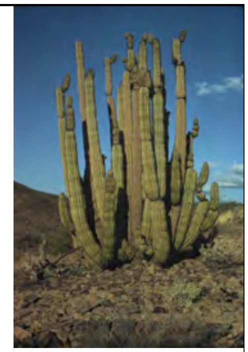
Alloteropsis



CAM plants

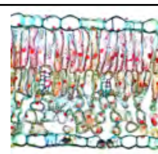
Crassulacean Acid Metabolism

CAM present in ferns, gymnosperms, and angiosperms

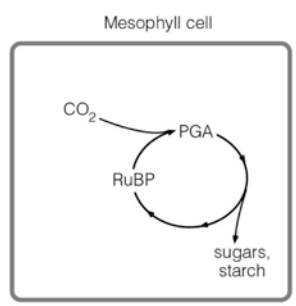


independent evolution probably also occurred

Polypodiales	Polypodiaceae
Gymnospermae	Welwitschiaceae
Monocotyledonae	Agavaceae, Bromeliaceae, Liliaceae, Orchidaceae
Dicotyledonae	Aizoaceae, Asclepiadaceae, Bataceae, Cactaceae, Capparidaceae, Caryophyllaceae, Chenopodiaceae, Compositae, Crassulaceae, Cucurbitaceae, Didiereaceae, Euphorbiaceae, Geraniaceae, Labiatae, Oxalidaceae, Passifloraceae, Piperaceae, Plantaginaceae, Portulacaceae, Tetragoniaceae, Vitaceae

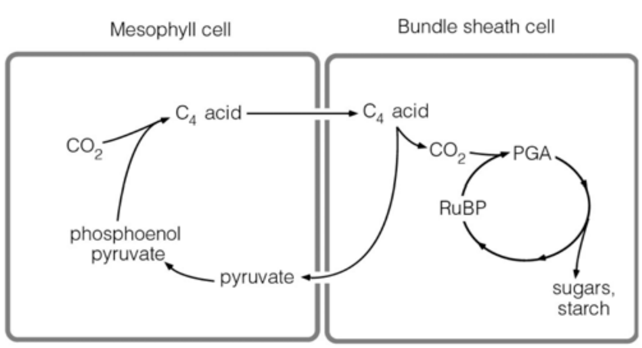


C_3 plants

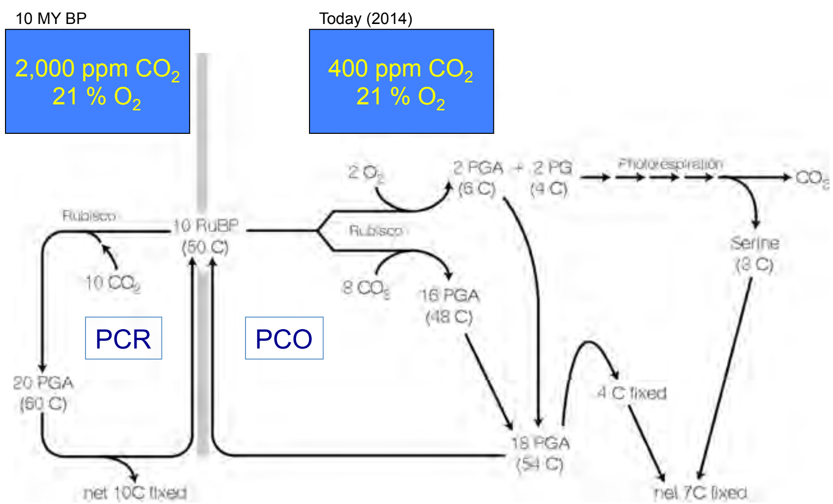


C_4 pathway requires
• spatial separation of carboxylases
• regeneration of PEP from pyruvate

C_4 plants

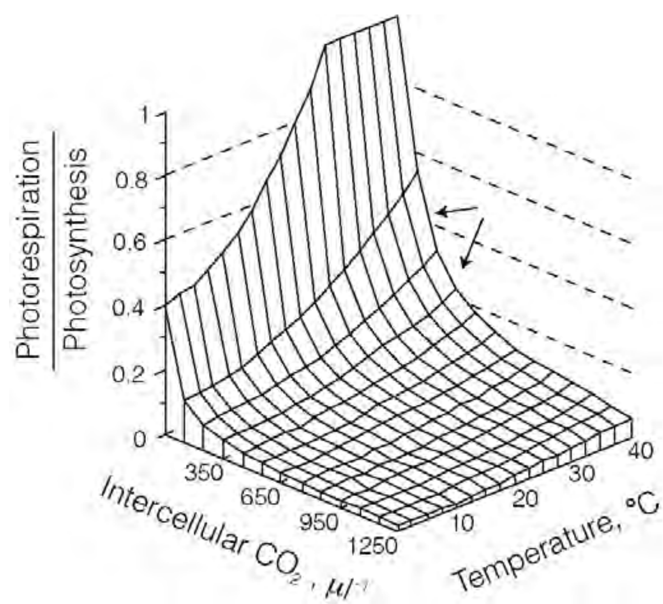


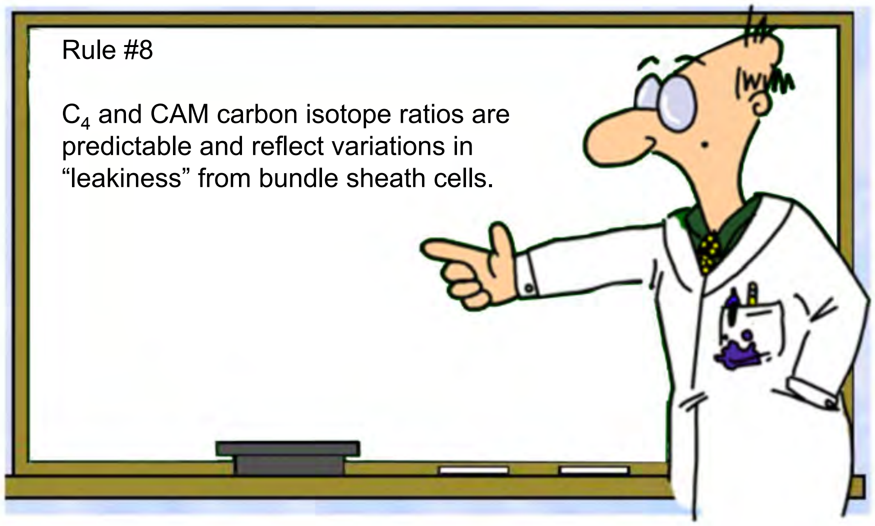
A change in atmospheric CO_2 affects the PCR/PCO ratio



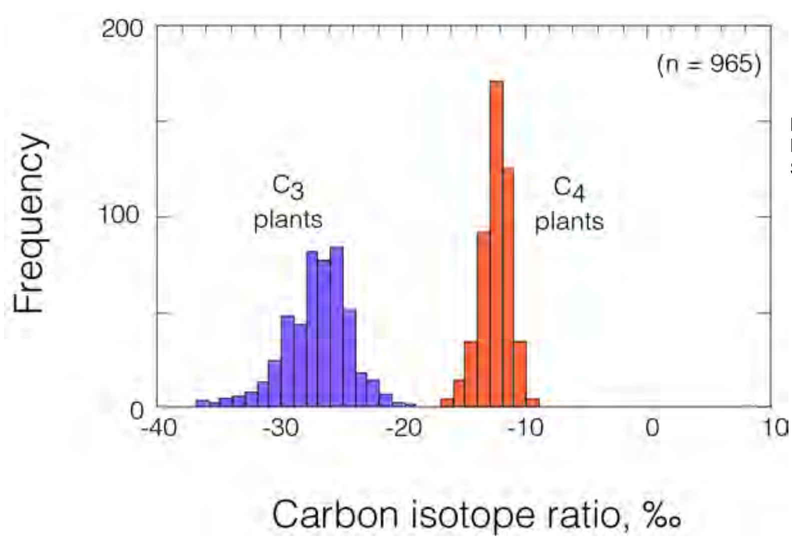
Rubisco oxygenase activity in PCO is known as photorespiration

Photorespiration to photosynthesis is a function of both temperature and $[CO_2]$





C_3 and C_4 plants differ in their carbon isotope ratios



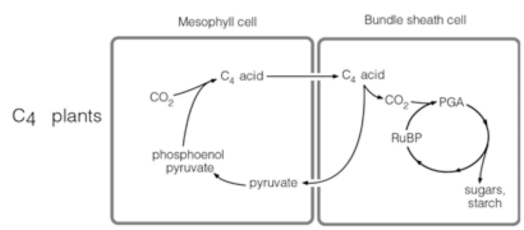
Phylogeny
Environment
Stress

Cerling et al (1997)

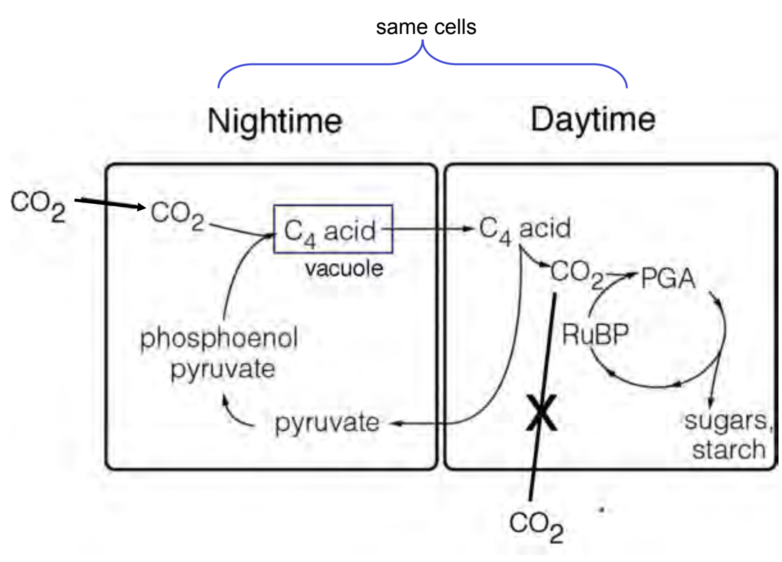
Carbon isotope discrimination in C_4 plants follows sample principles as we saw in C_3 carbon isotope discrimination, except that the first substrate is bicarbonate and not carbon dioxide.

$$b_4 = e_s + e_b + b_4^*$$

$$\Delta = a\frac{c_a - c_i}{c_a} + (b_4 + b_3\phi - a)\frac{c_i}{c_a} = a + (b_4 + b_3\phi - a)\frac{c_i}{c_a}$$



How the CAM cycle differs from C_4 photosynthesis



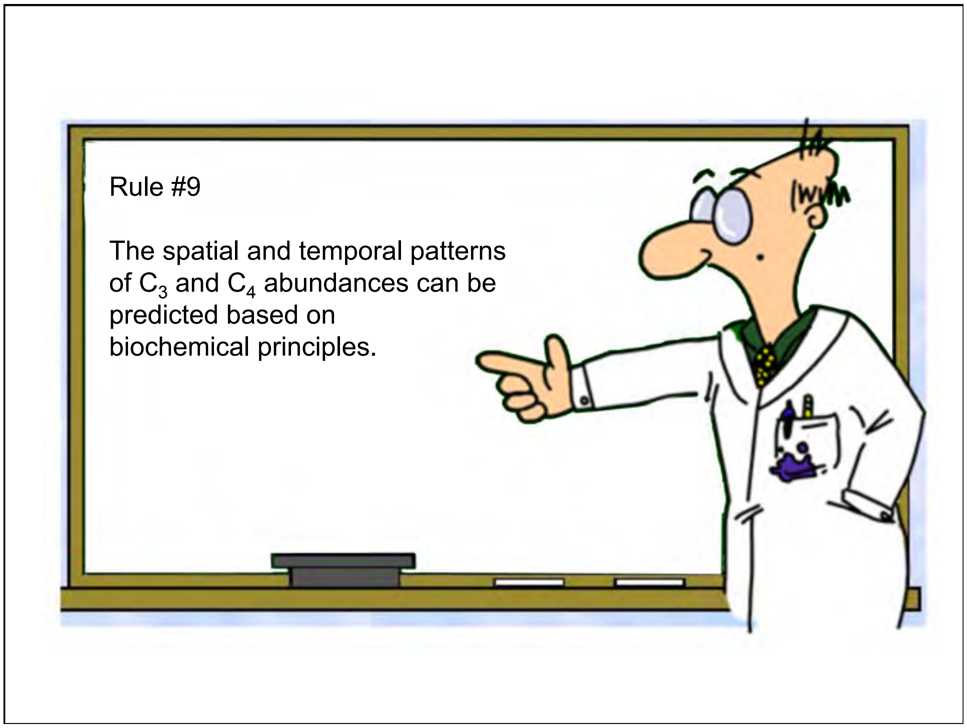
stomates open at night

stomates closed during day

Carbon isotope discrimination in CAM plants

$$\Delta = a + (b_4 - a)\frac{c_i}{c_a}$$

Questions?

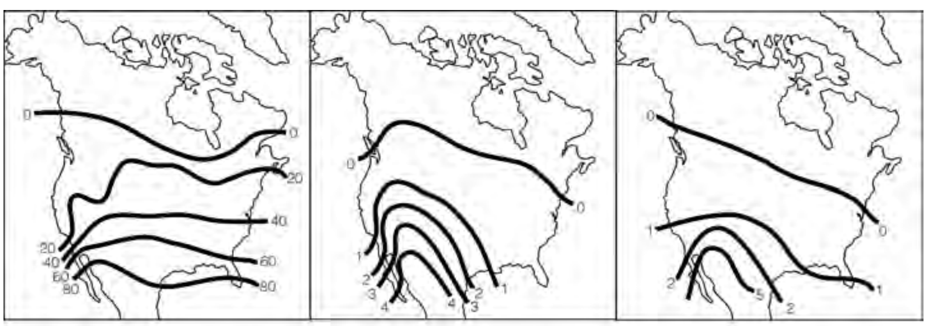

Rule #9
The spatial and temporal patterns of C_3 and C_4 abundances can be predicted based on biochemical principles.


Compare within a life form ...
C_3 grasses
C_4 grasses
… or between life forms
C_3 trees
C_4 grasses
Remember when you have soil organic matter, you may or may not know what lived aboveground in the past.

Common C_3/C_4 landscapes in tropical and subtropical sites

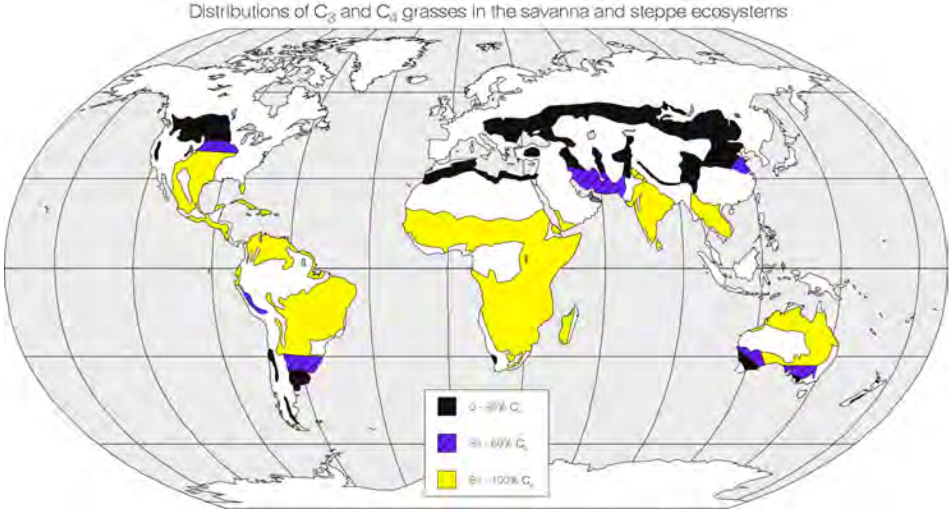
- C_3 trees
- C_4 crops and grasses

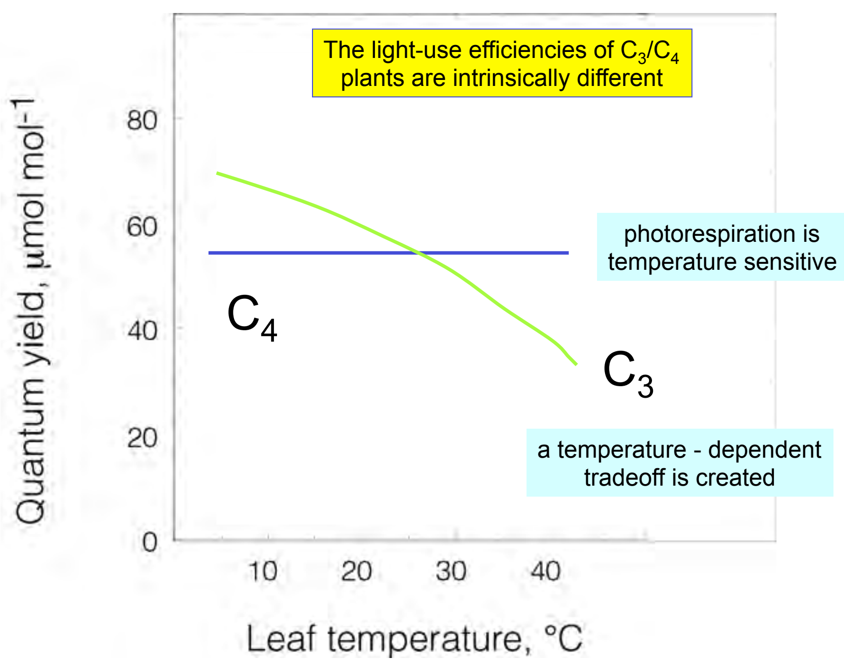
The abundance of C_3/C_4 plants varies along environmental gradients



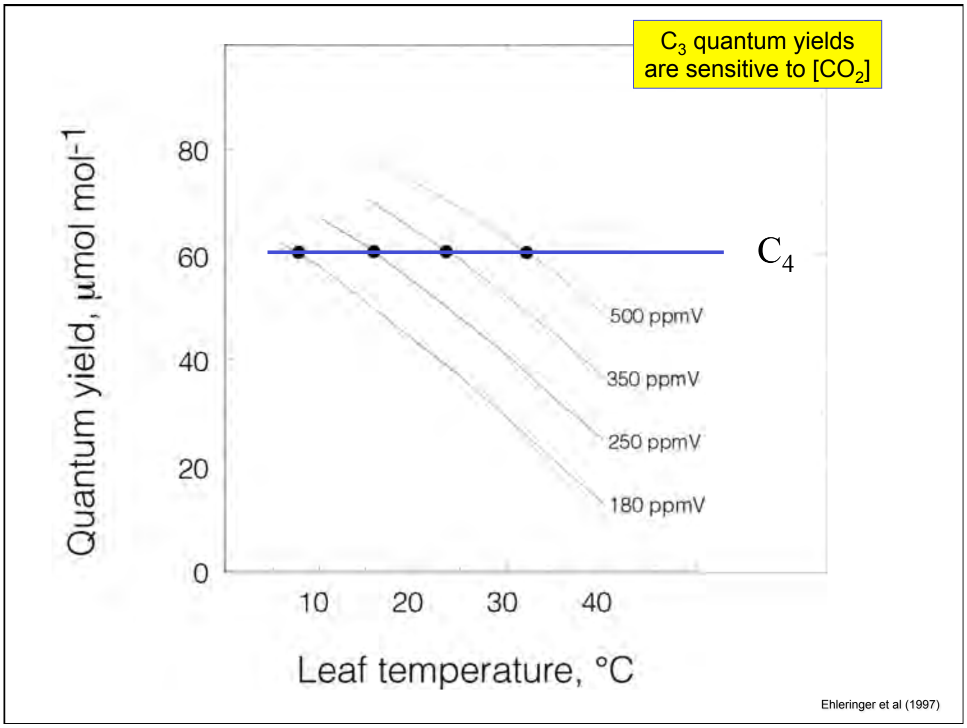
Ehleringer (1979)

Distributions of C_3 and C_4 grasses in the savanna and steppe ecosystems

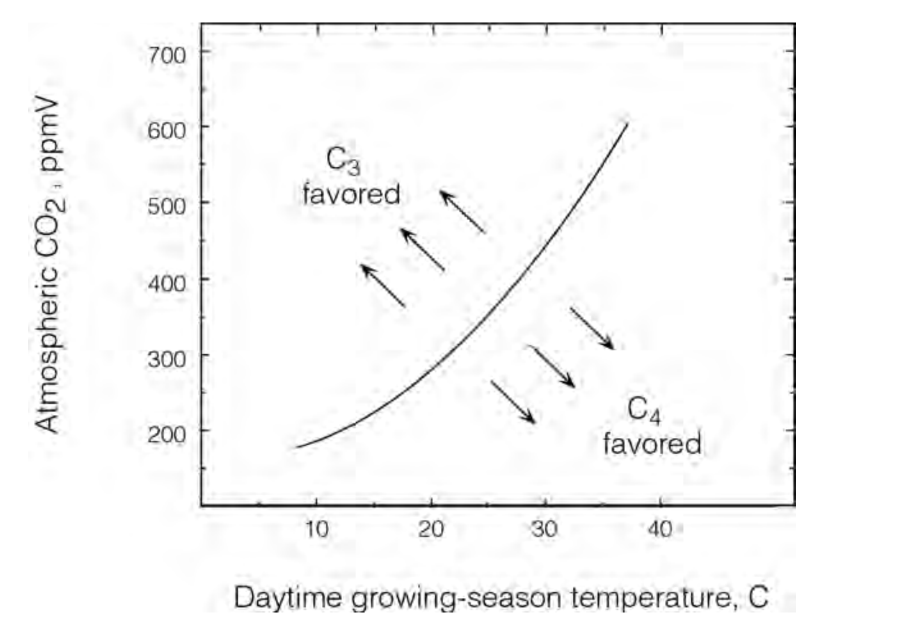




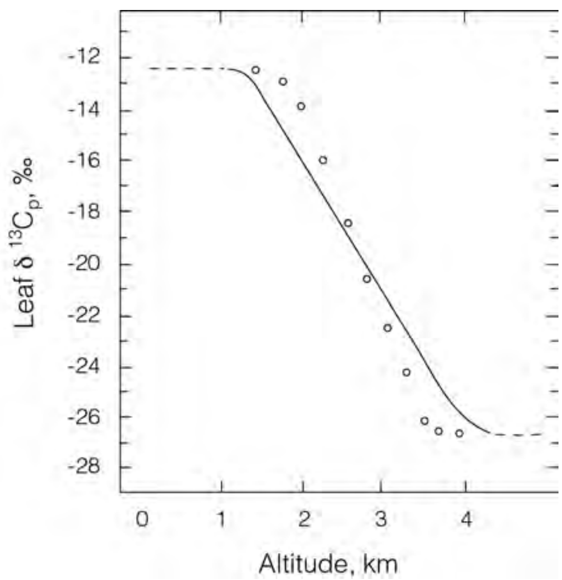
Ehleringer and Bjorkman (1977)



The distribution of C_3/C_4 plants relates to CO_2 and temperature

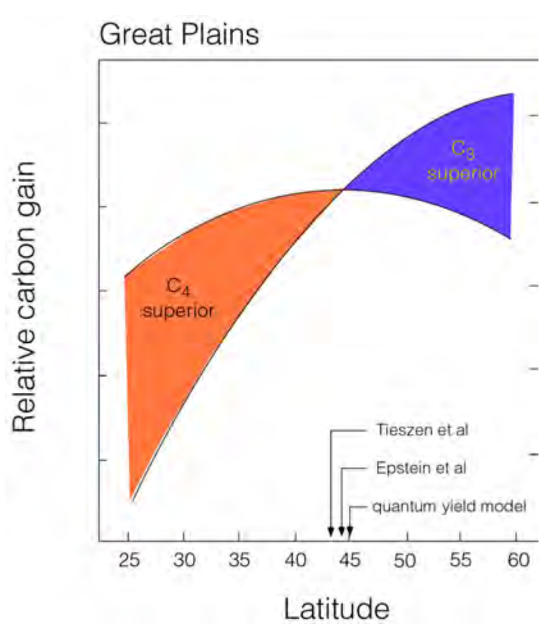


The abundance of C_3/C_4 plants varies along environmental gradients



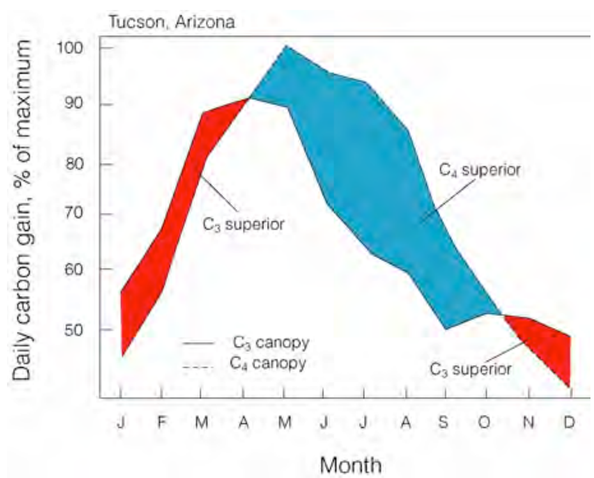
Tieszen et al (1979)

C_4 abundance is predicted to decrease with increasing latitude



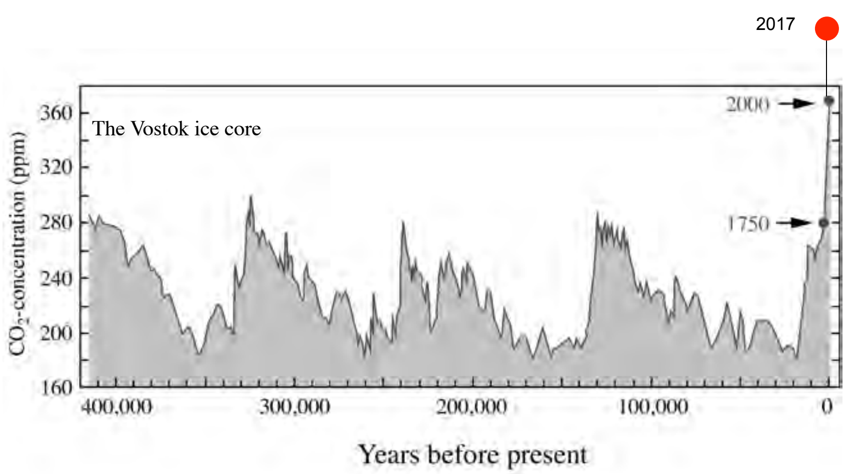
Ehleringer (1978)

C_4 abundance is predicted to decrease in cool growing seasons

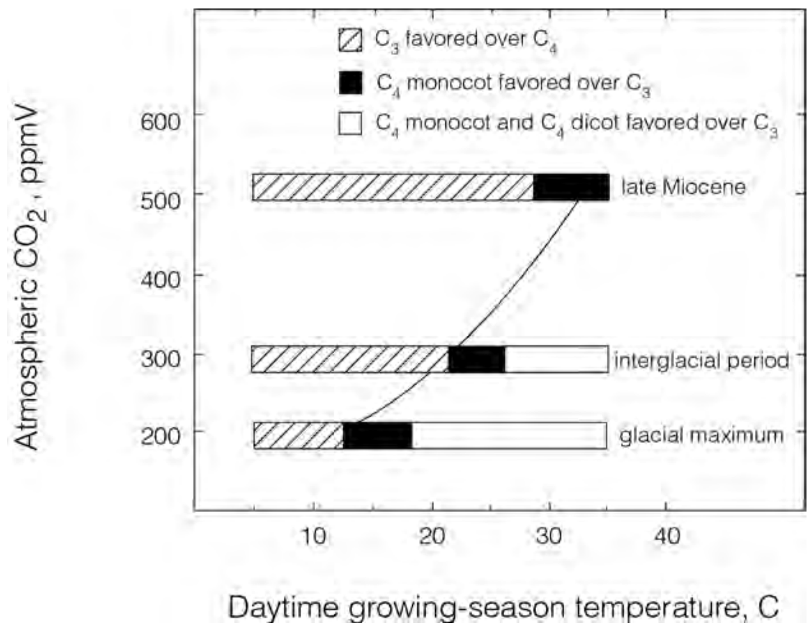


Ehleringer (1978)

What climate drivers are important for photosynthesis relationships between C_3/C_4 photosynthesis and climate?

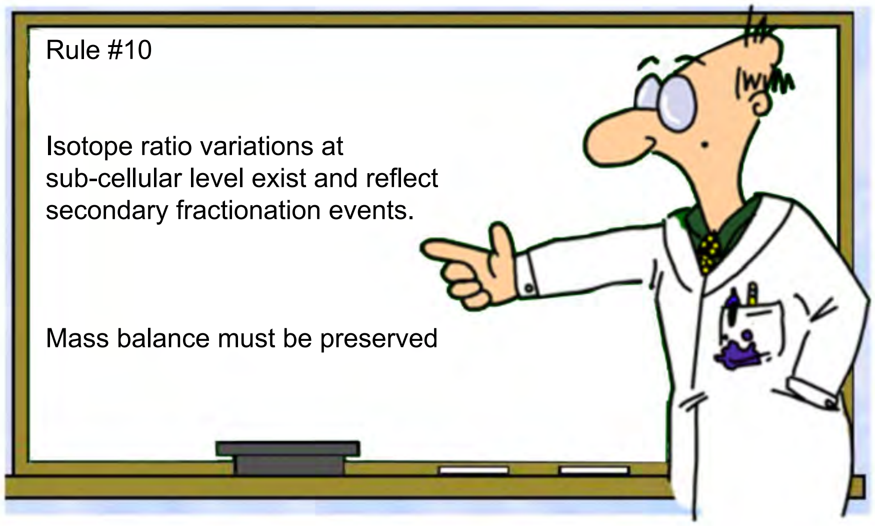


C_4 plant distributions are predicted to expand as CO_2 decreases



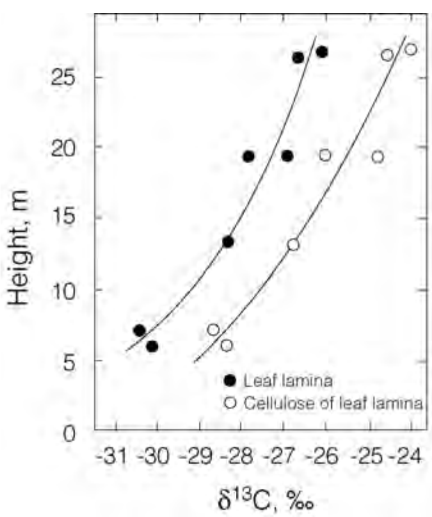
Ehleringer et al (1997)

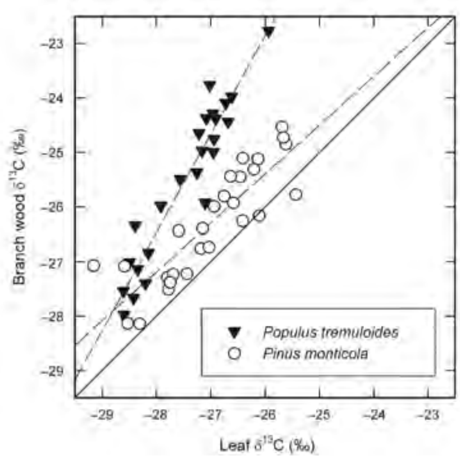
Questions?



The are predictable intra-plant variations in ^{13}C

beech tree

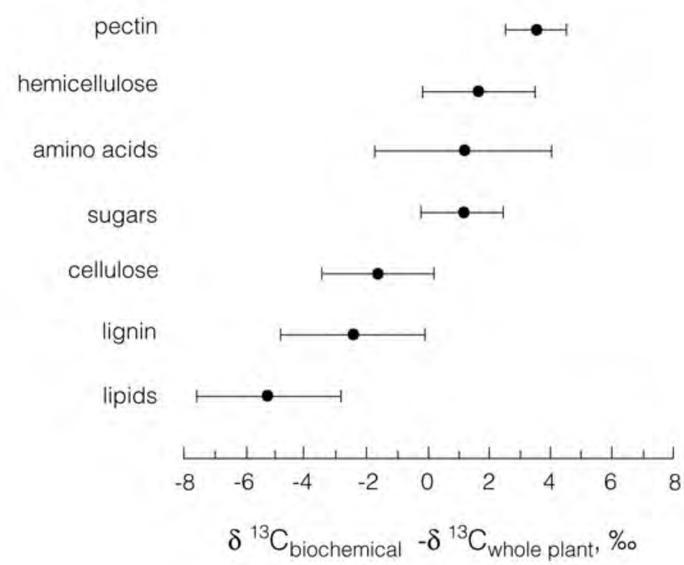




Leavitt and Long (1982)

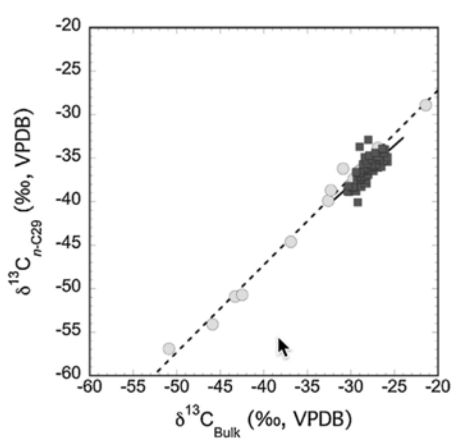
Cernusak et al. (2009)

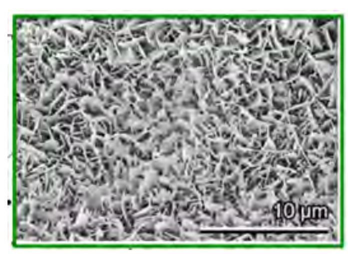
Intermolecular variations in ^{13}C composition

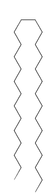


Boutton (1996)

Leaf cuticular wax and whole-leaf ^{13}C composition in *Cannabis*



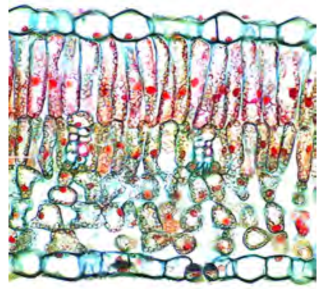




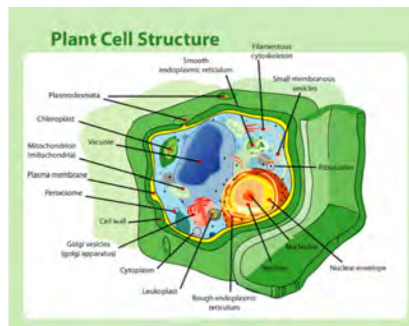
Leaf waxes (biomarker) do not turnover once formed, even though there may be extensive changes in water stress, carbohydrate 13-C, etc.

Tipple et (2015)

Appreciate that different leaf compounds have different turnover rates.



L**ow** turnover rates:
- cellulose (climate indicator)
- waxes (biomarker)
- nucleic acids (biomarker)



H**igh** turnover rates:
- carbohydrates (sugars and starches)
- lipids
- proteins
- metabolites
- sugars

How can respired CO_2 be ^{13}C-enriched with respect to substrate $\delta^{13}C$?

glucose: **C-C-C-C-C-C** = -25 ‰

^{13}C-enriched

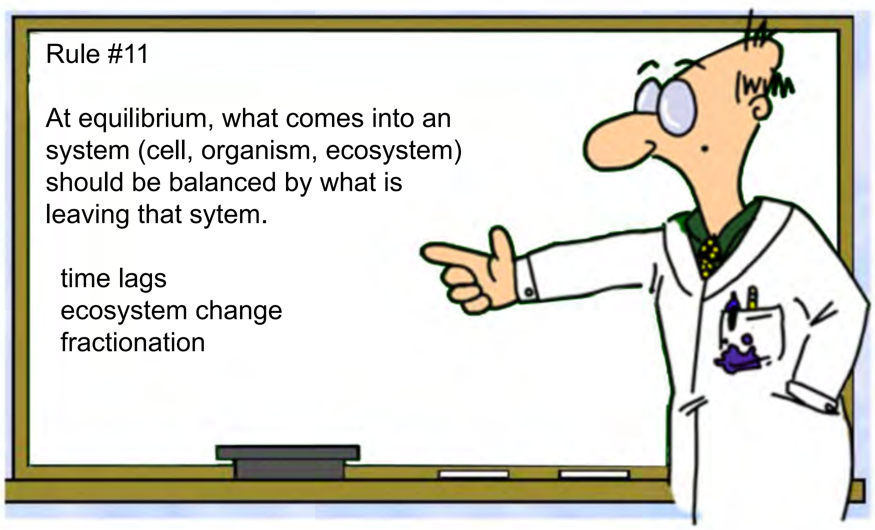
pyruvate **C-C-C**

PDH → CO_2 ^{13}C-enriched = -21‰ (C-3 and C-4 positions)

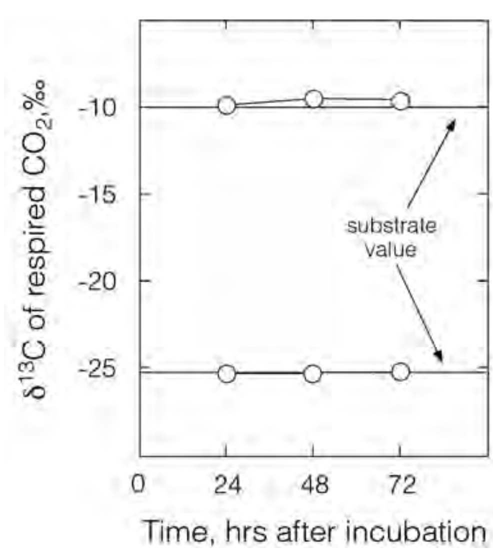
acetyl-CoA ⇄ e.g. Lipids = -33‰

Krebs Cycle → 2 CO_2 (-26‰) (C-2 and C-5)
(-28.5‰) (C-1 and C-6)

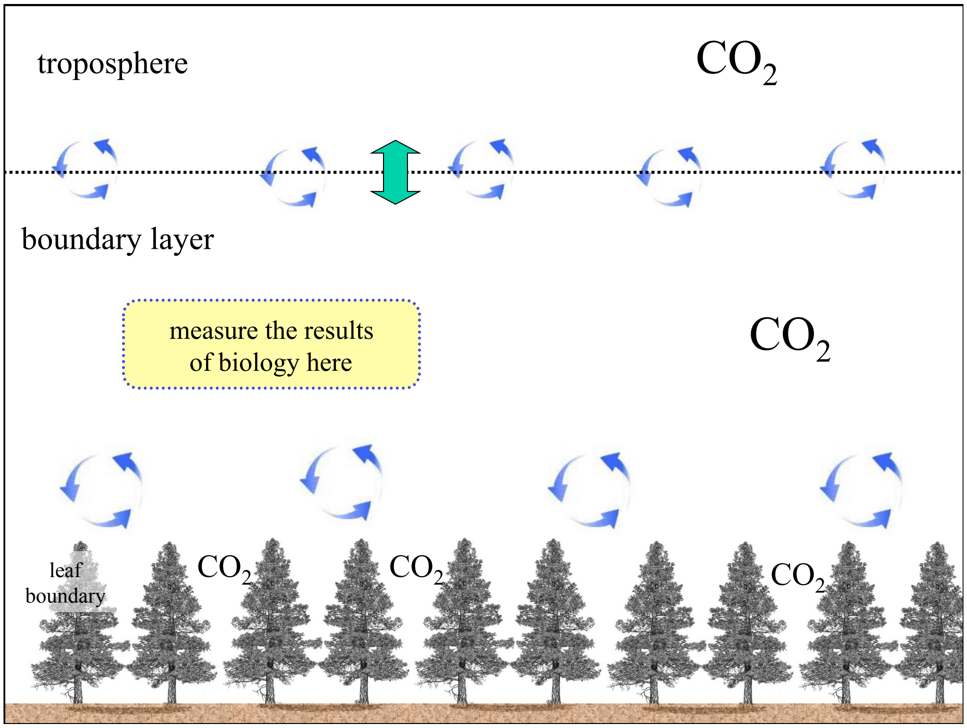
Short-term variation in metabolic fluxes may induce variability of isotope ratio of dark respired CO_2

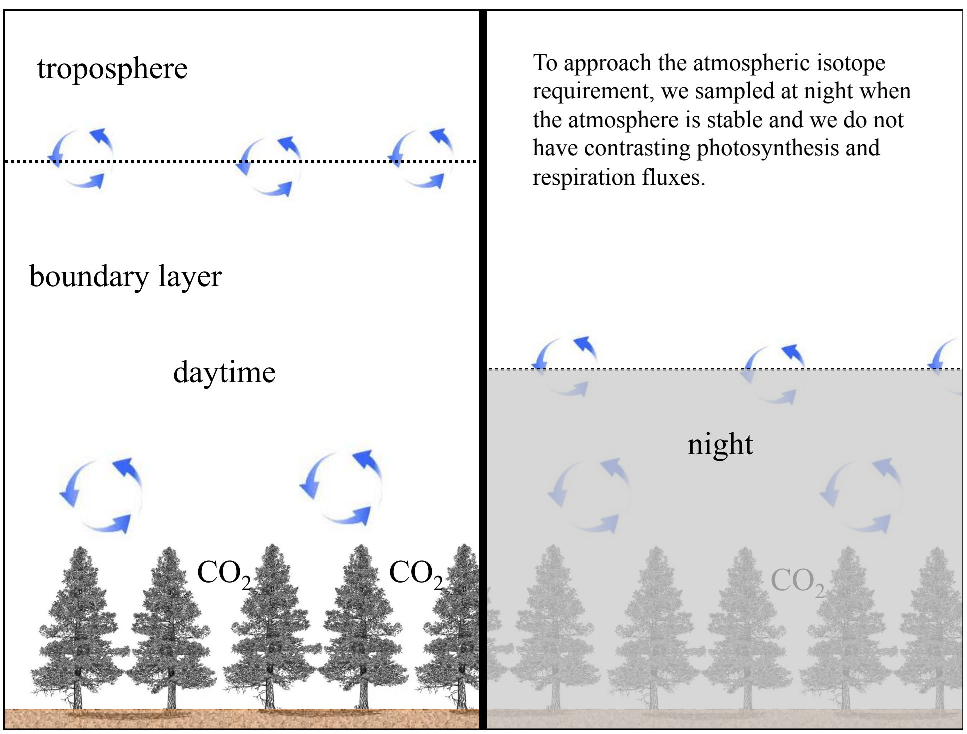


During cellular respiration, there appears to be no fractionation
(what goes in = what comes out)

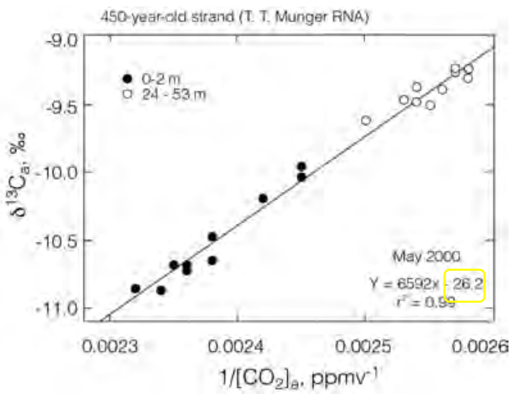


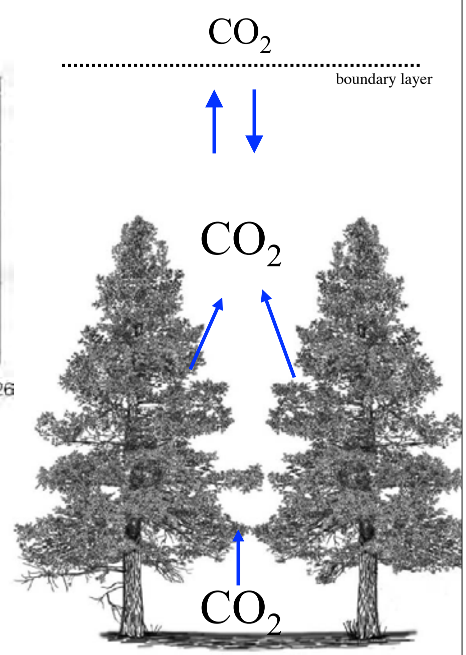
Guang-hui Lin


troposphere
CO_2
boundary layer
measure the results of biology here
CO_2
leaf boundary
CO_2
CO_2
CO_2


troposphere
boundary layer
daytime
CO_2
CO_2
To approach the atmospheric isotope requirement, we sampled at night when the atmosphere is stable and we do not have contrasting photosynthesis and respiration fluxes.
night
CO_2

a Keeling plot

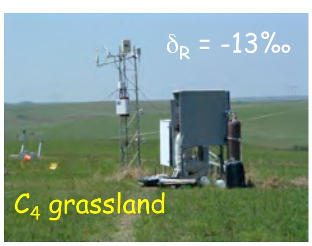


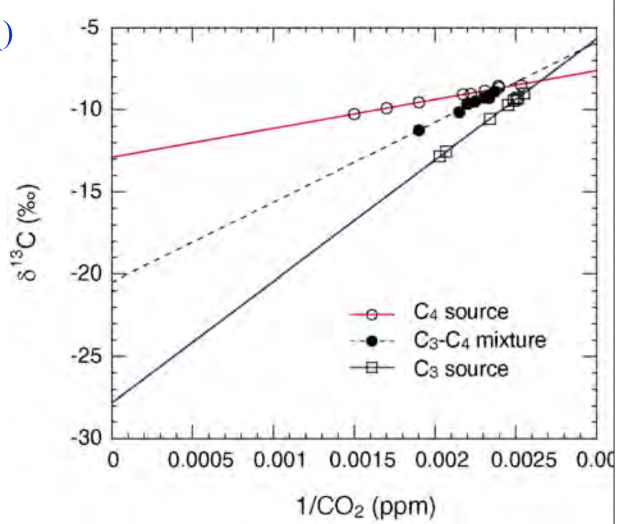


$$\delta^{13}C_{leaf} = \delta^{13}C_{air} - a - (b - a)\ c_i/c_a$$

$$A \approx (1 - c_i/c_a) \bullet c_a \bullet g_{CO_2}$$

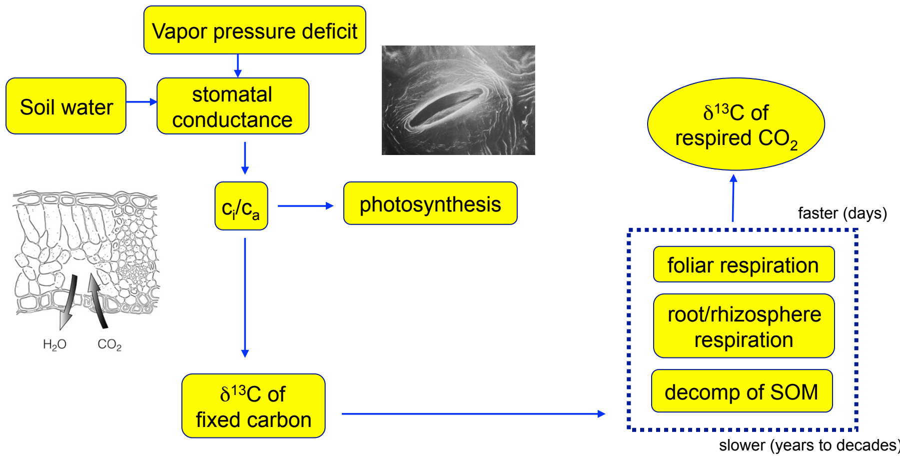
$\delta^{13}C$ of respiration (δ_R)



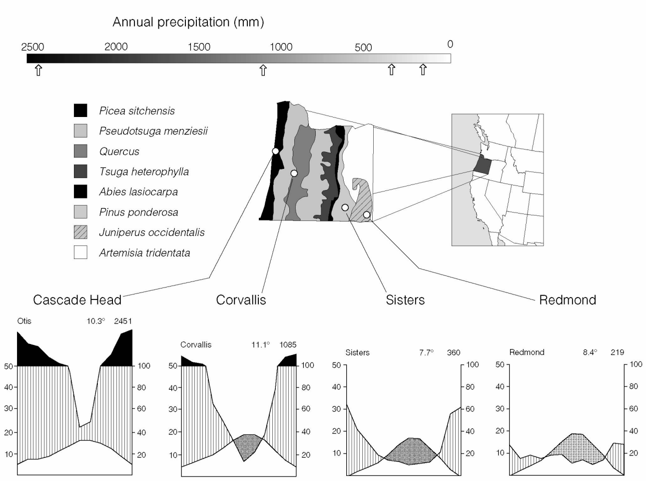


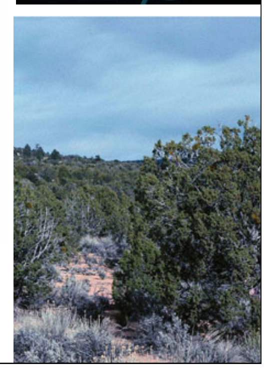
Chun-Ta Lai

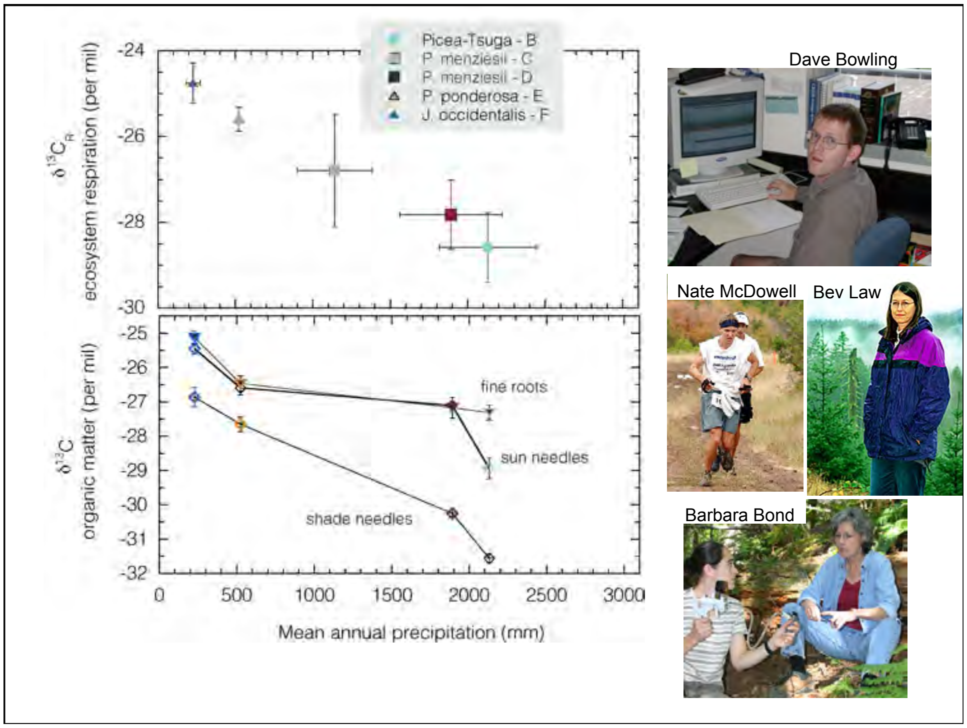
Linkage between gas exchange activity and $\delta^{13}C_R$



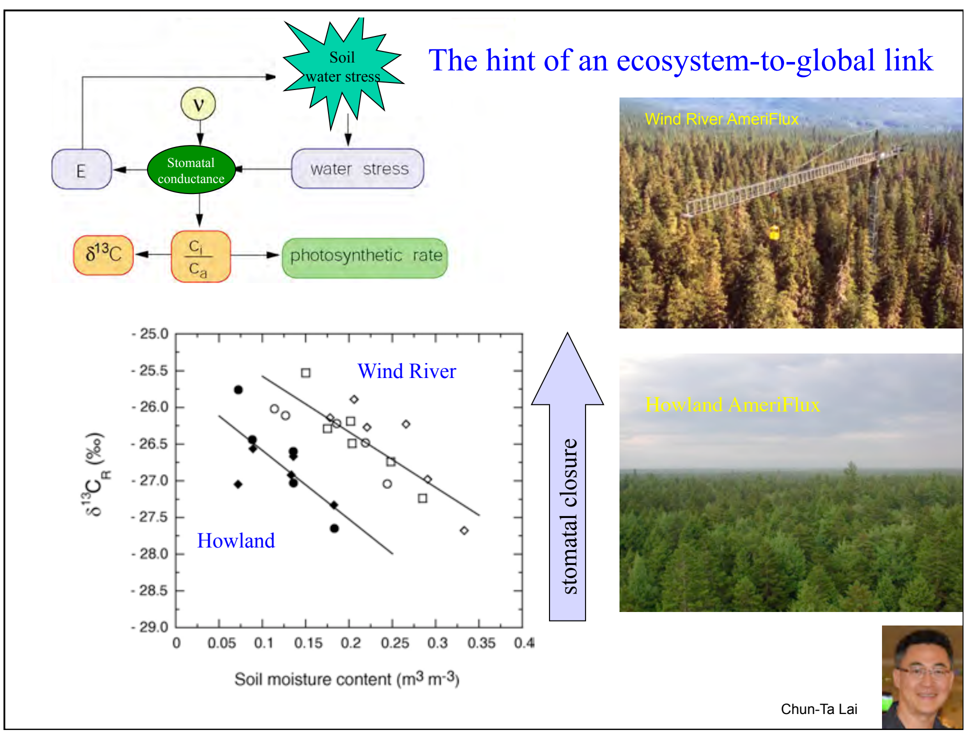
OTTER: a precipitation transect across Oregon







Picea-Tsuga - B
P. menziesii - C
P. menziesii - D
P. ponderosa - E
J. occidentalis - F
δ13CR ecosystem respiration (per mil)
-24
-26
-28
-30
δ13C organic matter (per mil)
-25
-26
-27
-28
-29
-30
-31
-32
fine roots
sun needles
shade needles
0
500
1000
1500
2000
2500
3000
Mean annual precipitation (mm)
Dave Bowling
Nate McDowell
Bev Law
Barbara Bond



The hint of an ecosystem-to-global link
Soil water stress
V
E
Stomatal conductance
water stress
δ13C
Ci/Ca
photosynthetic rate
Wind River AmeriFlux
Howland AmeriFlux
δ13CR (‰)
- 25.0
- 25.5
- 26.0
- 26.5
- 27.0
- 27.5
- 28.0
- 28.5
- 29.0
Wind River
Howland
0
0.05
0.1
0.15
0.2
0.25
0.3
0.35
0.4
Soil moisture content (m3 m-3)
stomatal closure
Chun-Ta Lai

Precipitation also impacts fluxes in rainforest ecosystems