

Microbial enzyme activity and carbon cycling in grassland soil fractions

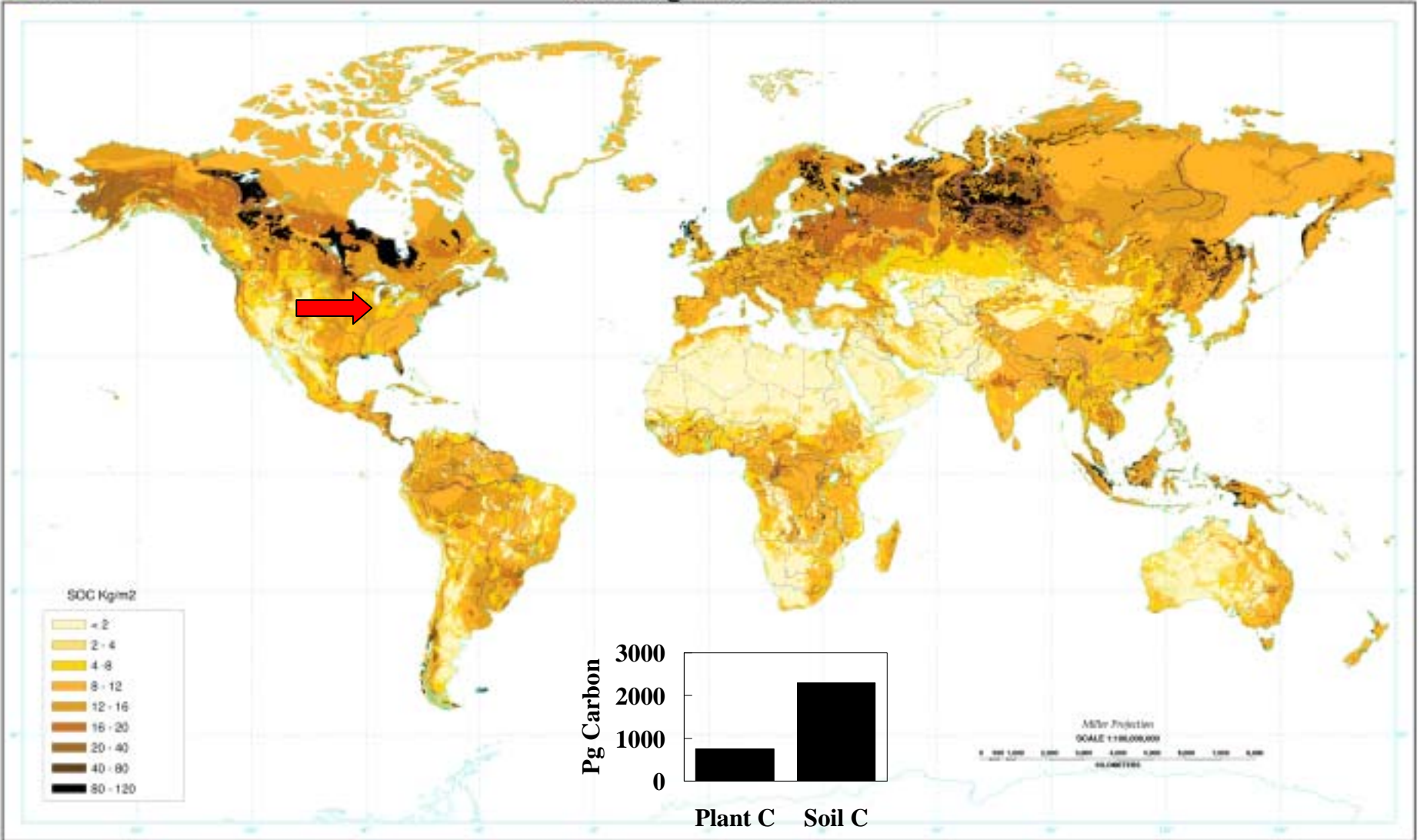
Steve Allison
GCEP Workshop
August 16, 2004

Primary research question

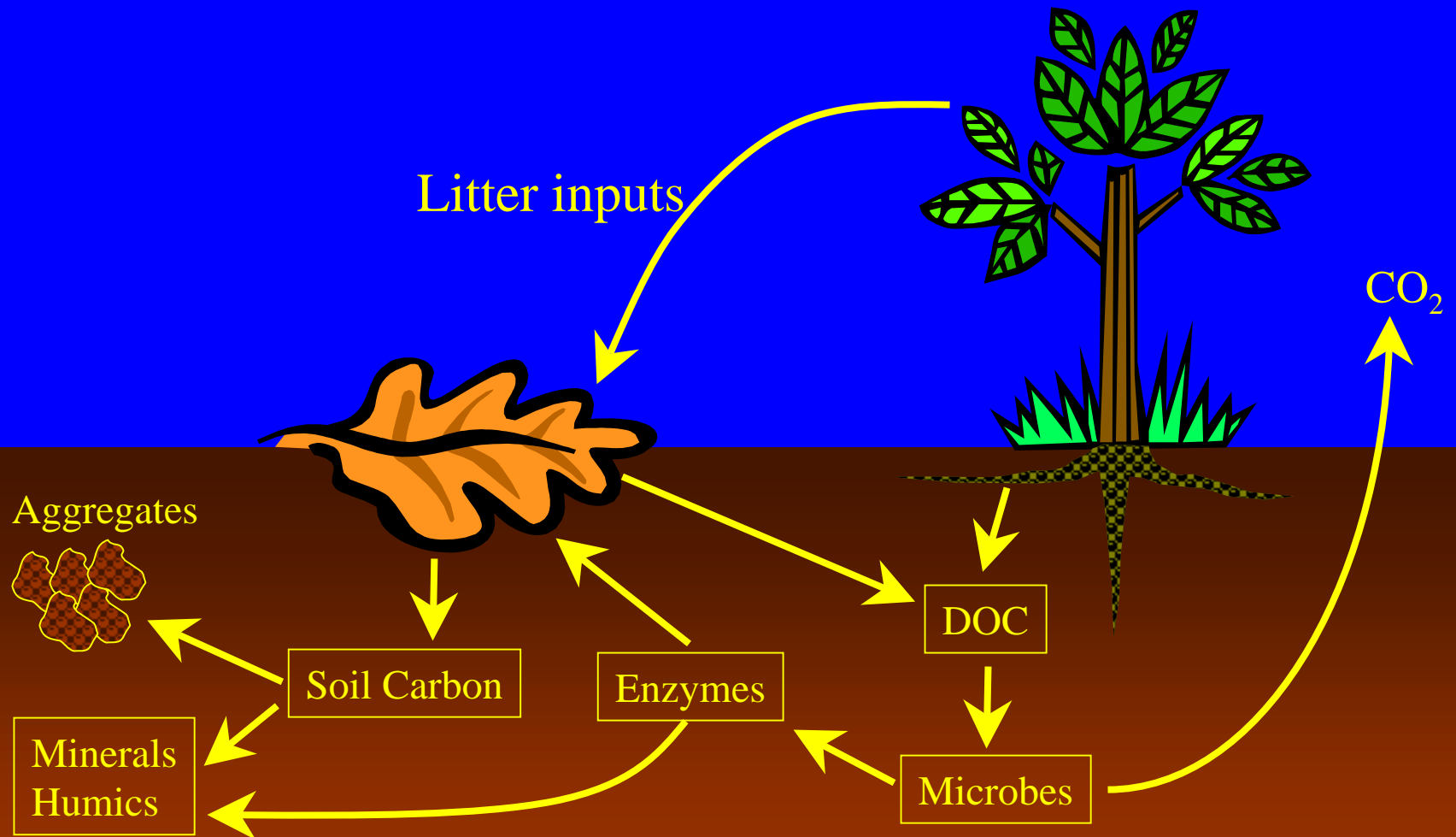
- Why does carbon remain in soil?

What prevents soil microbes from decomposing more carbon?

Soil Organic Carbon



The soil system



Soil Enzymes

- Produced by microbes to degrade and assimilate complex compounds
- First step to convert soil organic matter into CO₂ and inorganic nutrients
- Can affect soil carbon sequestration and CO₂ release

Enzymes and their functions

Enzyme	Function
Phosphatase	Releases PO_4^{3-}
Protease	Releases amino acids
Chitinase	Releases chitin monomers
Beta-glucosidase	Degrades cellulose
Cellobiohydrolase	Degrades cellulose
Polyphenol oxidase	Degrades lignin

An enzymic ‘latch’ on a global carbon store

A shortage of oxygen locks up carbon in peatlands by restraining a single enzyme.

Historically, northern peatlands have removed carbon dioxide from the atmosphere faster than it has been re-released, so they now contain 20–30% of the world’s soil carbon stock¹ (the equivalent of over 60% of the atmospheric carbon pool²). Here we show that the anaerobic conditions in peatlands prevent the enzyme phenol oxidase from eliminating phenolic compounds that inhibit biodegradation. This indicates that oxygen limitation on a single peatland enzyme may be all that prevents the re-release of a major store of global carbon into the atmosphere, with potentially serious implications for future global warming.

Mechanisms proposed to account for the slow decomposition rates in peatlands include the effects on microbial metabolism of low oxygen availability, low pH, low nutrient supply and low temperatures. But decomposition can be highly efficient in

Table 1 Effects on enzyme activities

	Control	Manipulated
Effect of oxygen on enzyme activity		
Sulphatase	66 ± 2.3	35 ± 1.4
Phosphatase	571 ± 2.4	387 ± 7.9
β-Glucosidase	237 ± 2.3	177 ± 12
Phenol oxidase	615 ± 93	4,350 ± 27
Effect of increasing phenol oxidase abundance		
Phenolics (µg l ⁻¹)	1,985 ± 55.4	1,444 ± 9.9
β-Glucosidase	1,677 ± 280	10,111 ± 380
Effect of phenolic removal on hydrolase activity		
Sulphatase	579 ± 36	849 ± 43
Phosphatase	3,707 ± 25	4,369 ± 180
β-Glucosidase	1,723 ± 120	2,183 ± 180
Xylosidase	116 ± 2.5	134 ± 5
Chitinase	243 ± 14	296 ± 3.5

Phenol oxidase activity (nmol 2-carboxy-2,3-dihydroindole-5,6-quinone formation min⁻¹ per g peat), hydrolase activities (nmol methylumbelliferone formation min⁻¹ per g peat) and phenolic compound concentrations (µg l⁻¹) are reported as mean ± s.e.

(Table 1). Lower water-tables, which are

field survey ($r=0.61$, $P<0.05$) that every doubling in phenol oxidase activity was accompanied by an approximate doubling in CO₂ production.

Taken together, our findings support the idea that oxygen constraints on a single enzyme, phenol oxidase, can minimize the activity of hydrolytic enzymes responsible for peat decomposition. This has profound implications in the context of climate change as a feedback to the process of intensified carbon loss. Increased peat aeration, as a result of droughts predicted by certain climate-change models¹³, has the potential to eliminate a critical mechanism restricting the re-release of CO₂ to the atmosphere. As such, phenol oxidase could be considered to represent a fragile ‘latch’ mechanism holding in place a vast carbon store of 455 gigatonnes.

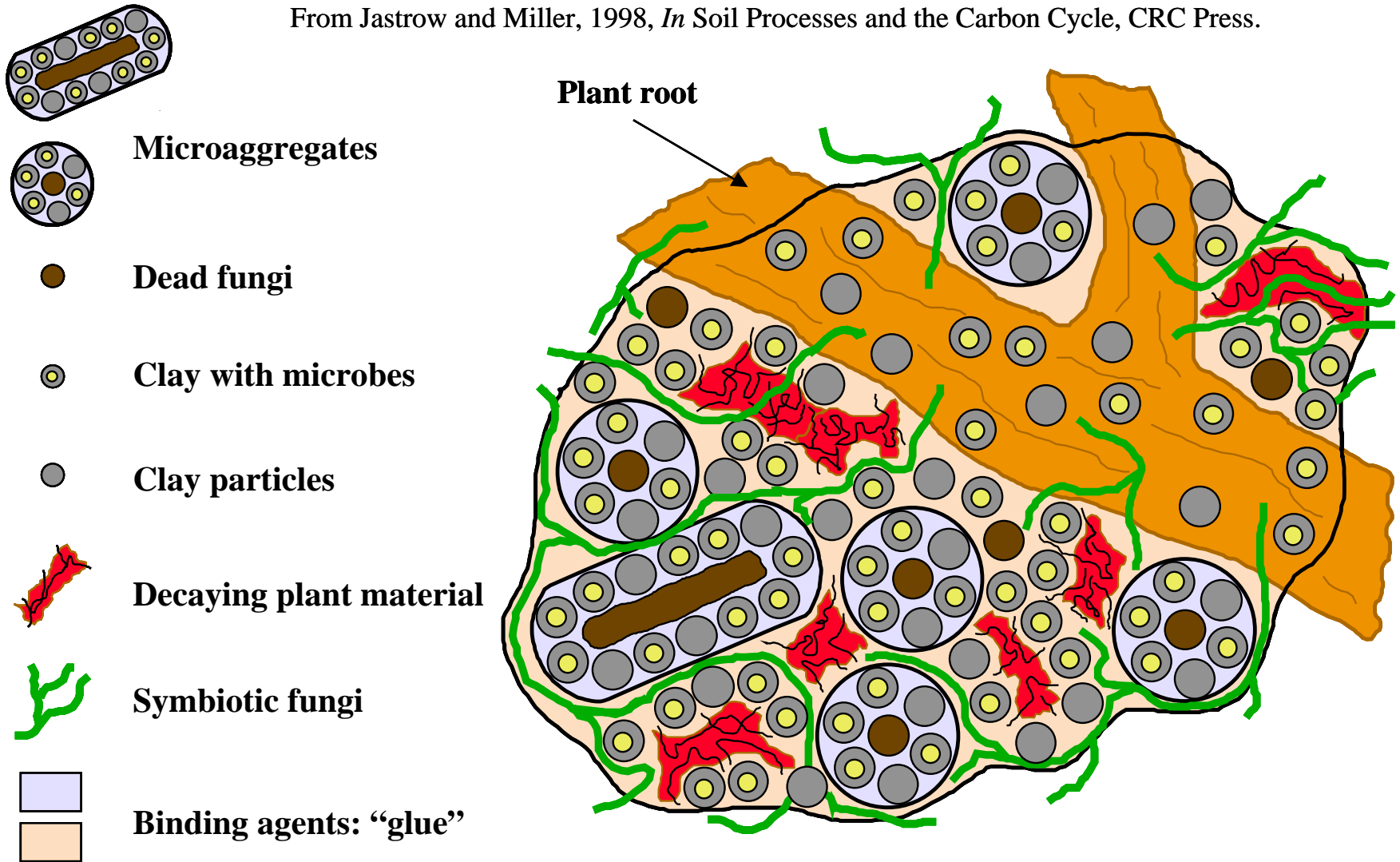
Chris Freeman*, Nick Ostle*†, Hojeong Kang*†

Questions for ANL work

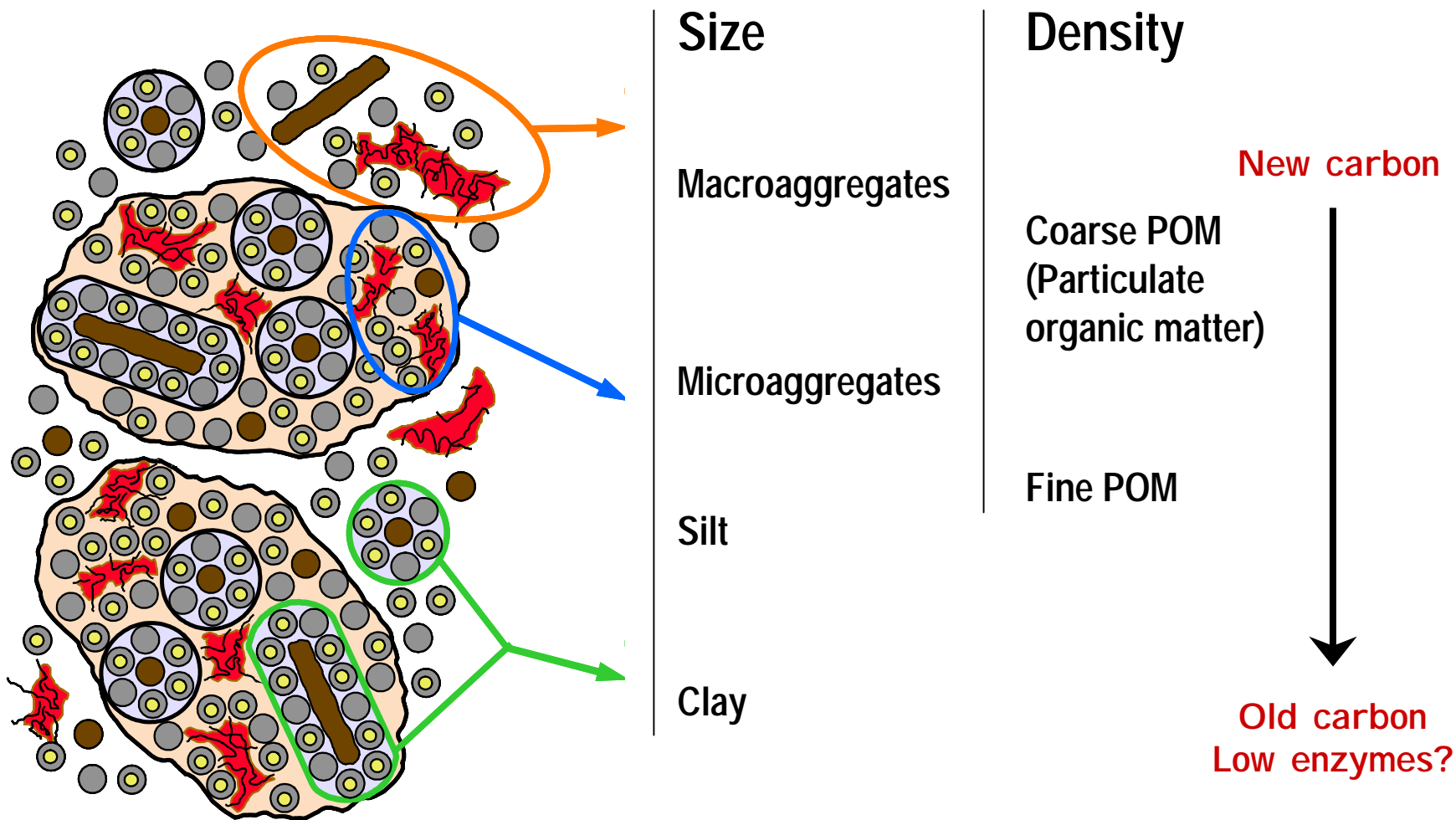
- Where is carbon located within the soil?
- Are enzymes excluded from aggregates and old carbon pools?
- How do enzymes change across restoration chronosequence?

CONCEPTUAL DIAGRAM OF AGGREGATE HIERARCHY

From Jastrow and Miller, 1998, *In Soil Processes and the Carbon Cycle*, CRC Press.



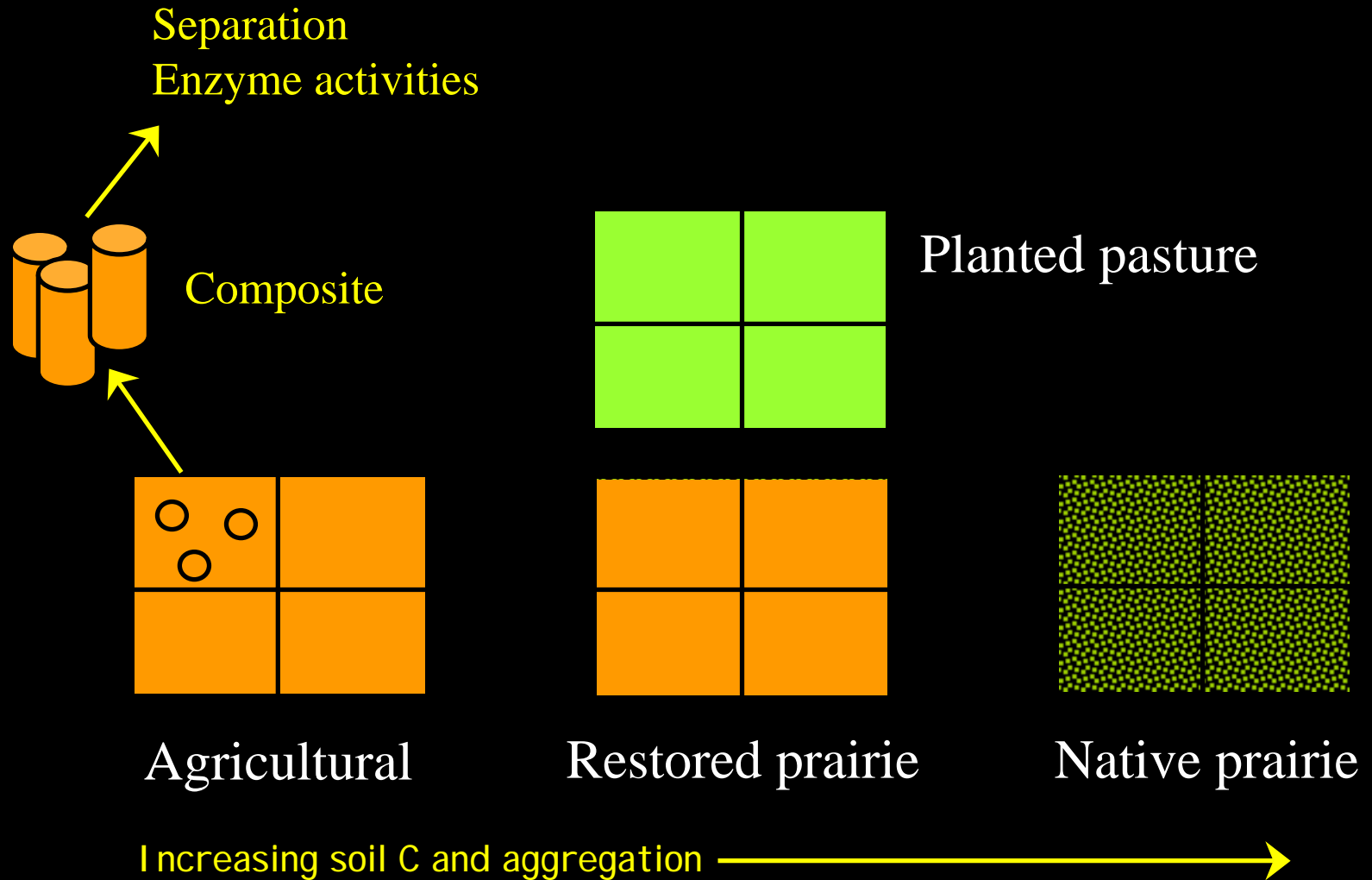
Fractionation of Soil Organic Matter Based on Aggregate Hierarchy



Predictions

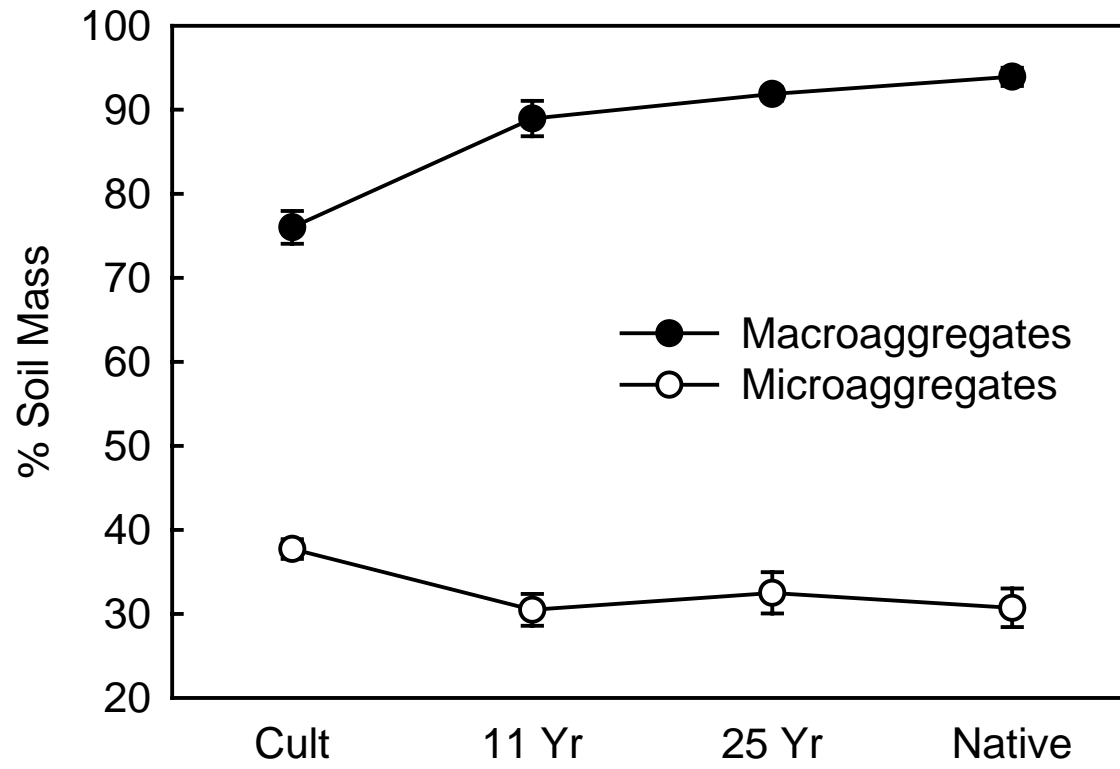
Fraction (Age)	Prediction
Light organic matter (6-31 yr)	Accessible substrates, high microbial activity, high enzyme activity
Macroaggregates (51 yr)	Includes some light organic matter; above average enzyme activity
Microaggregates (79 yr)	Physically shielded, below-average enzyme activity
Silt (74 yr)	Low enzyme activity
Clay (201 yr)	Lowest enzyme activity

Experimental Design

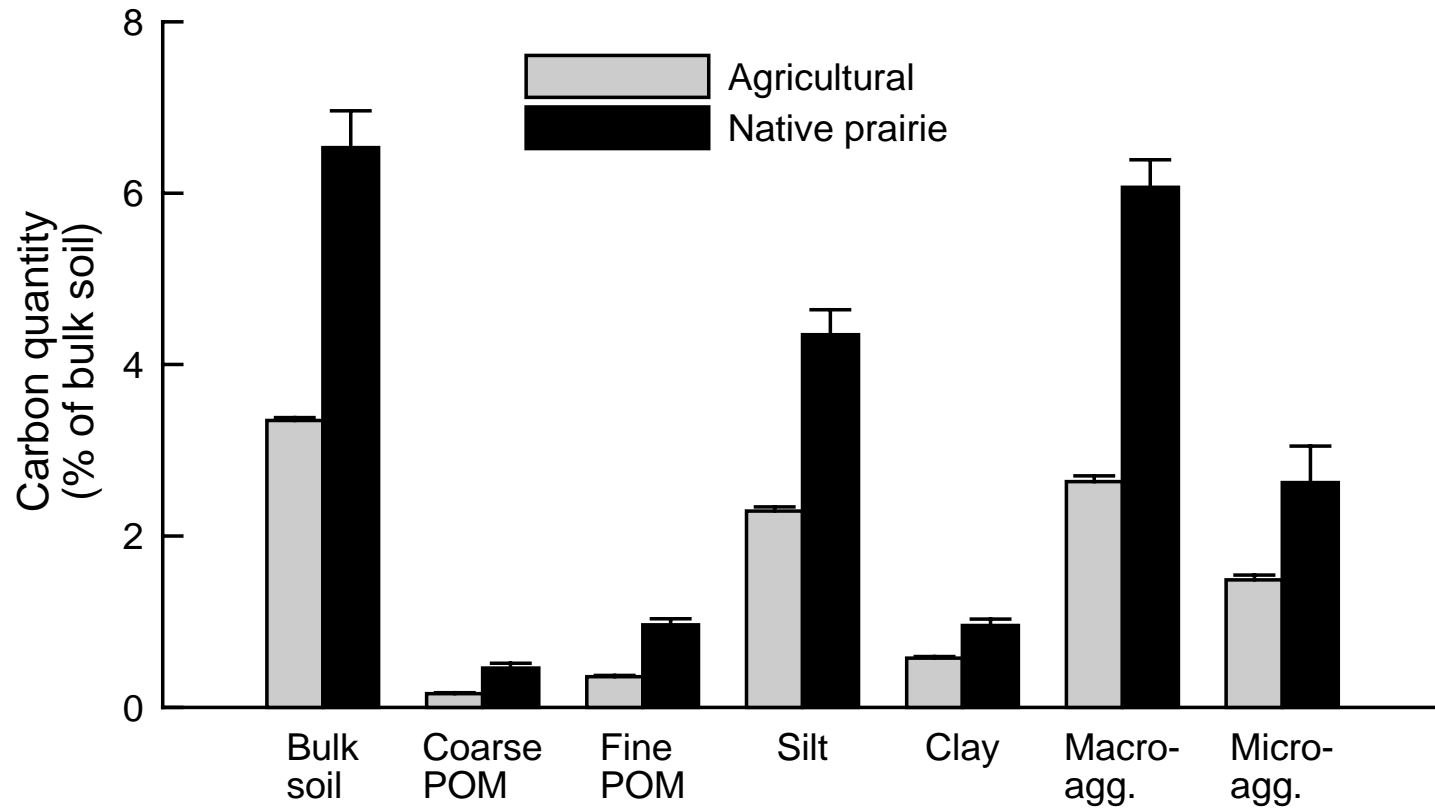




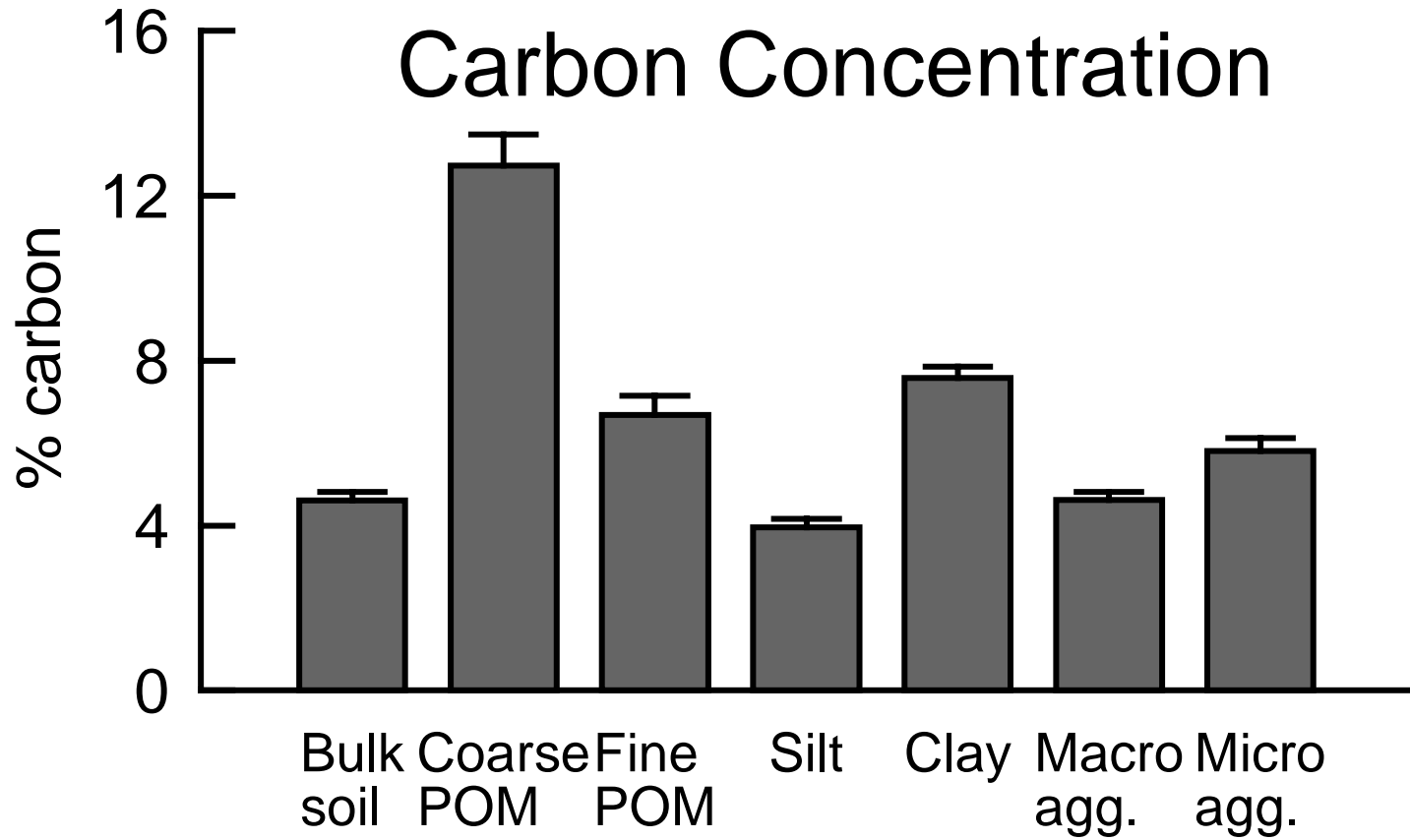
Macroaggregates increase over time



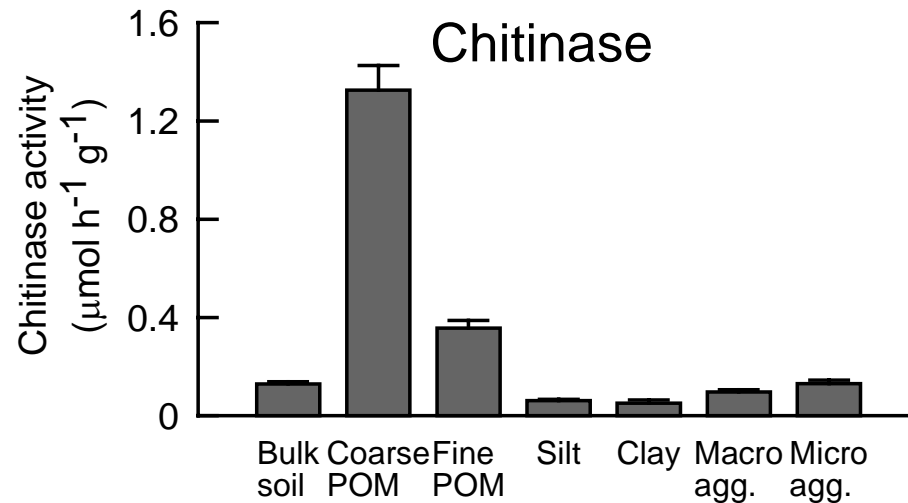
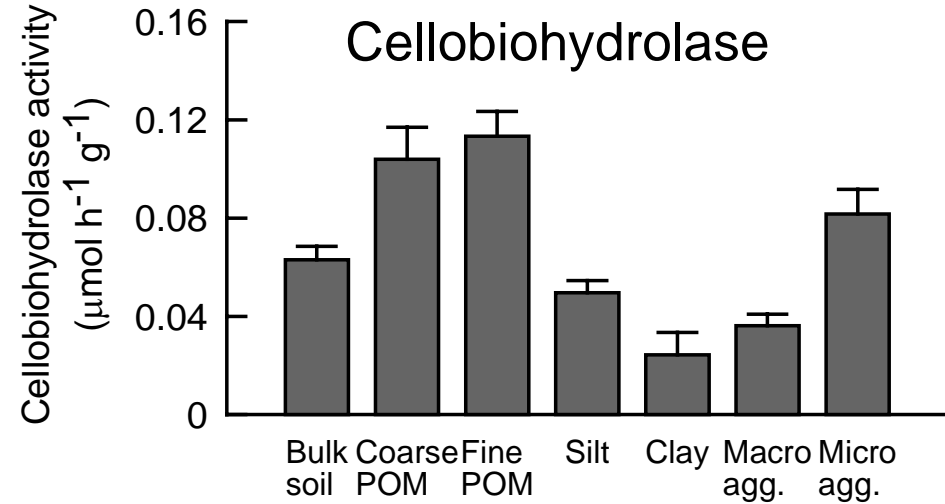
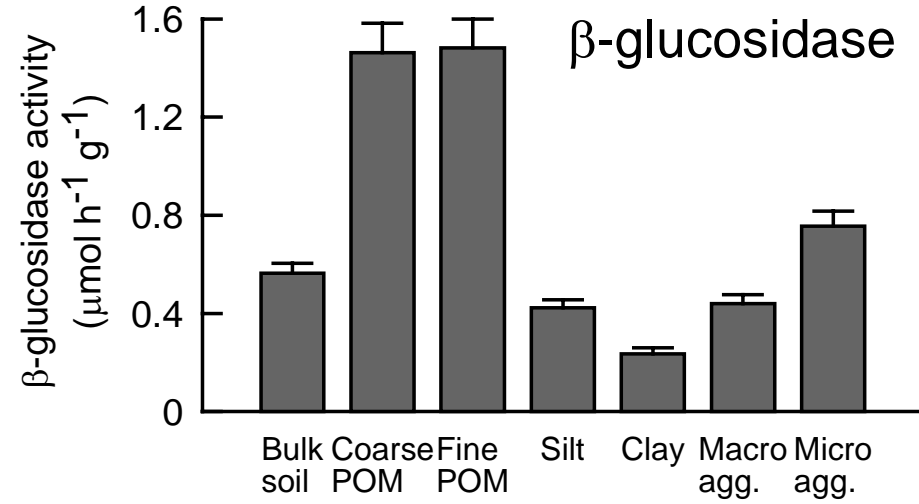
More C in native prairie soils



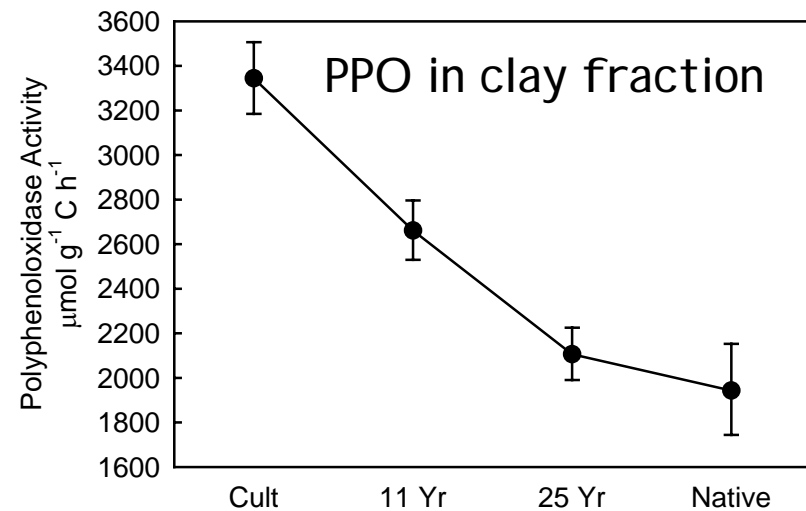
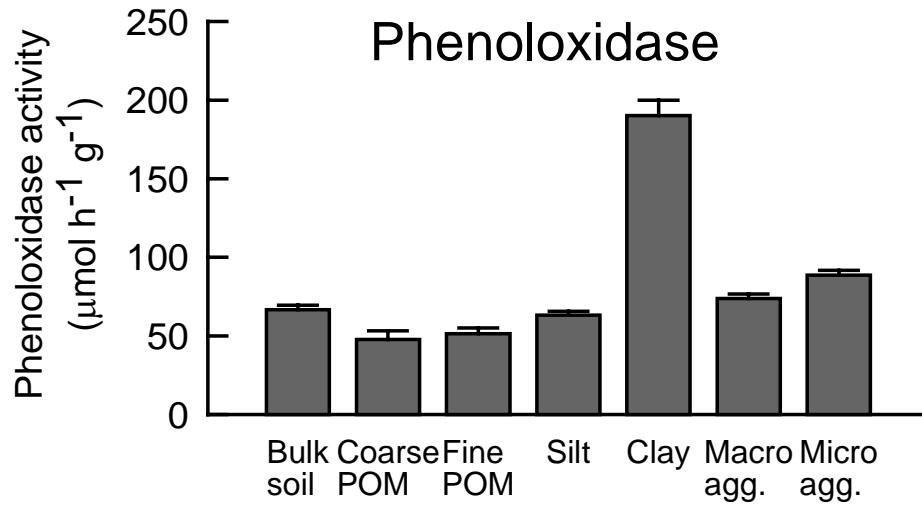
Highest C concentration in POM



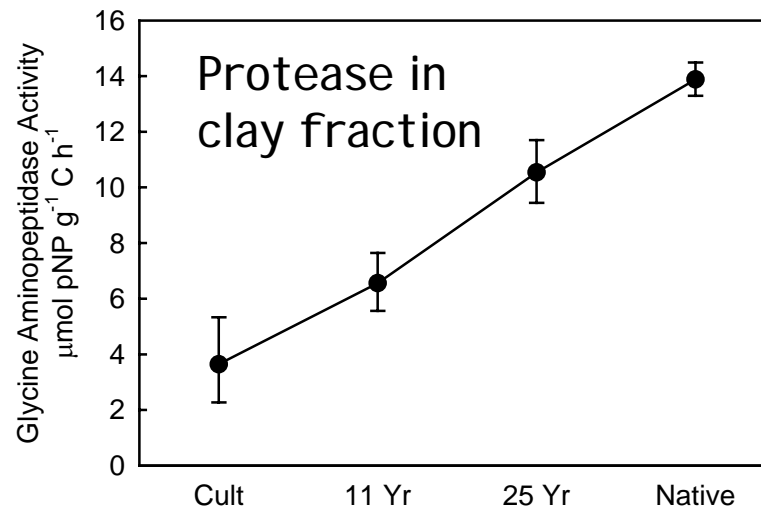
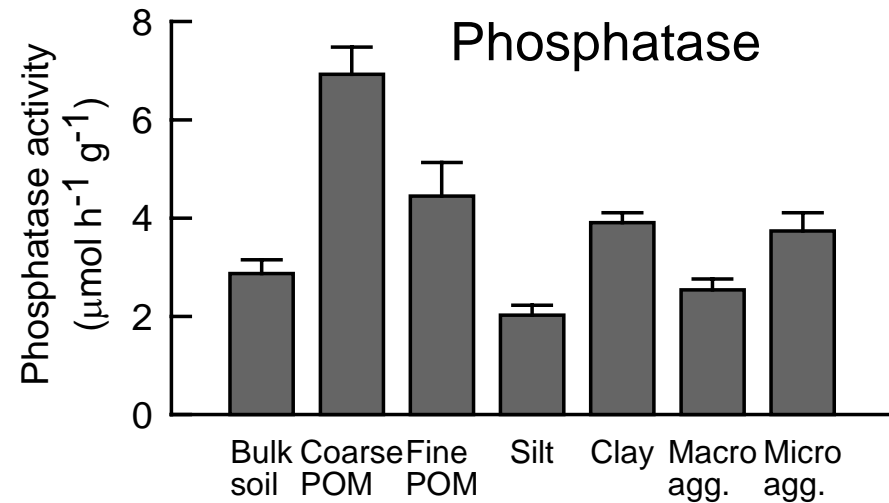
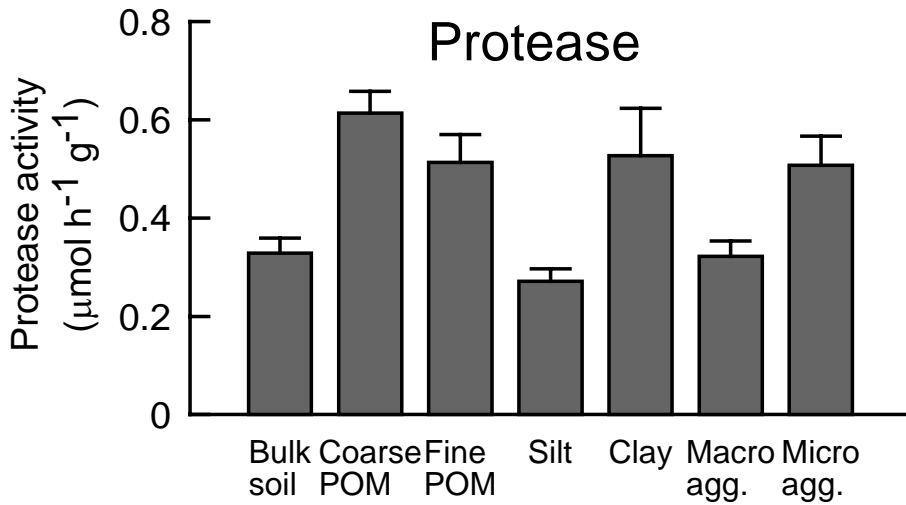
C-degrading enzymes are localized near their substrates



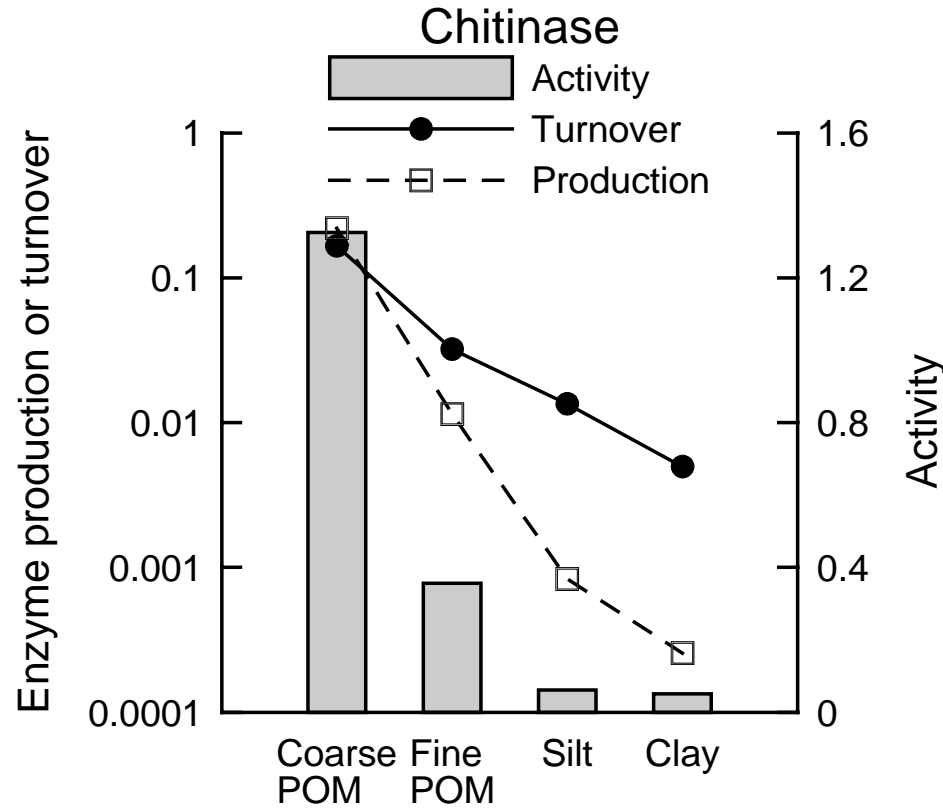
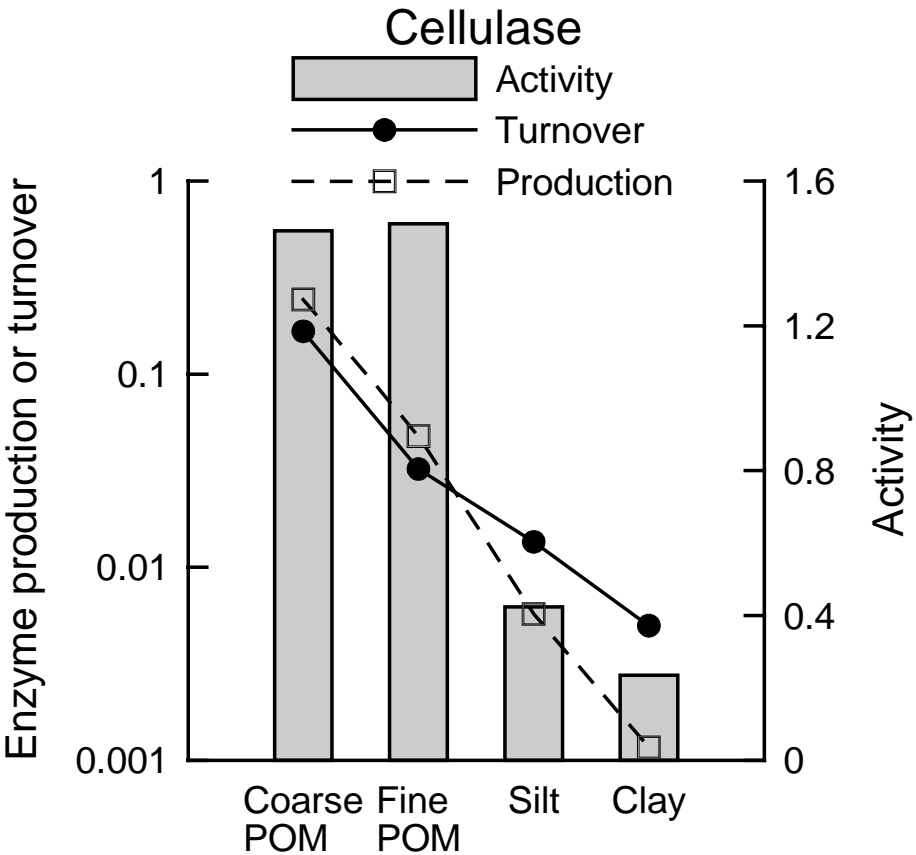
Polyphenoloxidase is localized in clay fraction



Nutrient enzymes produced across all fractions

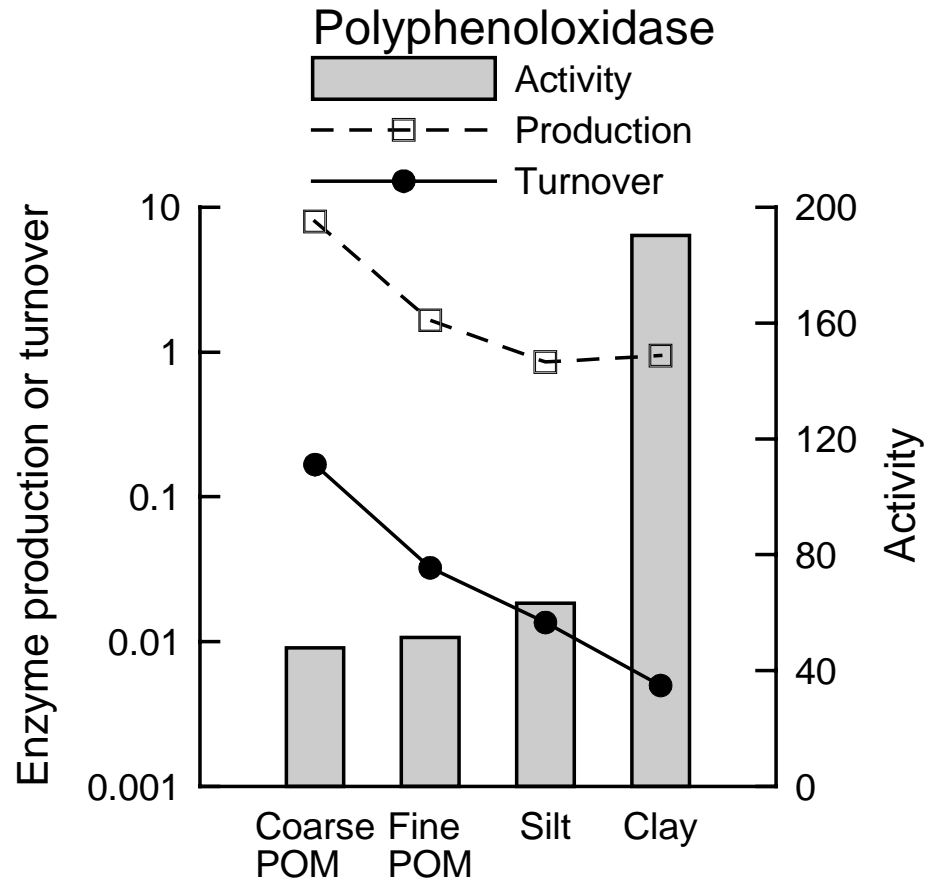


Enzyme turnover versus production



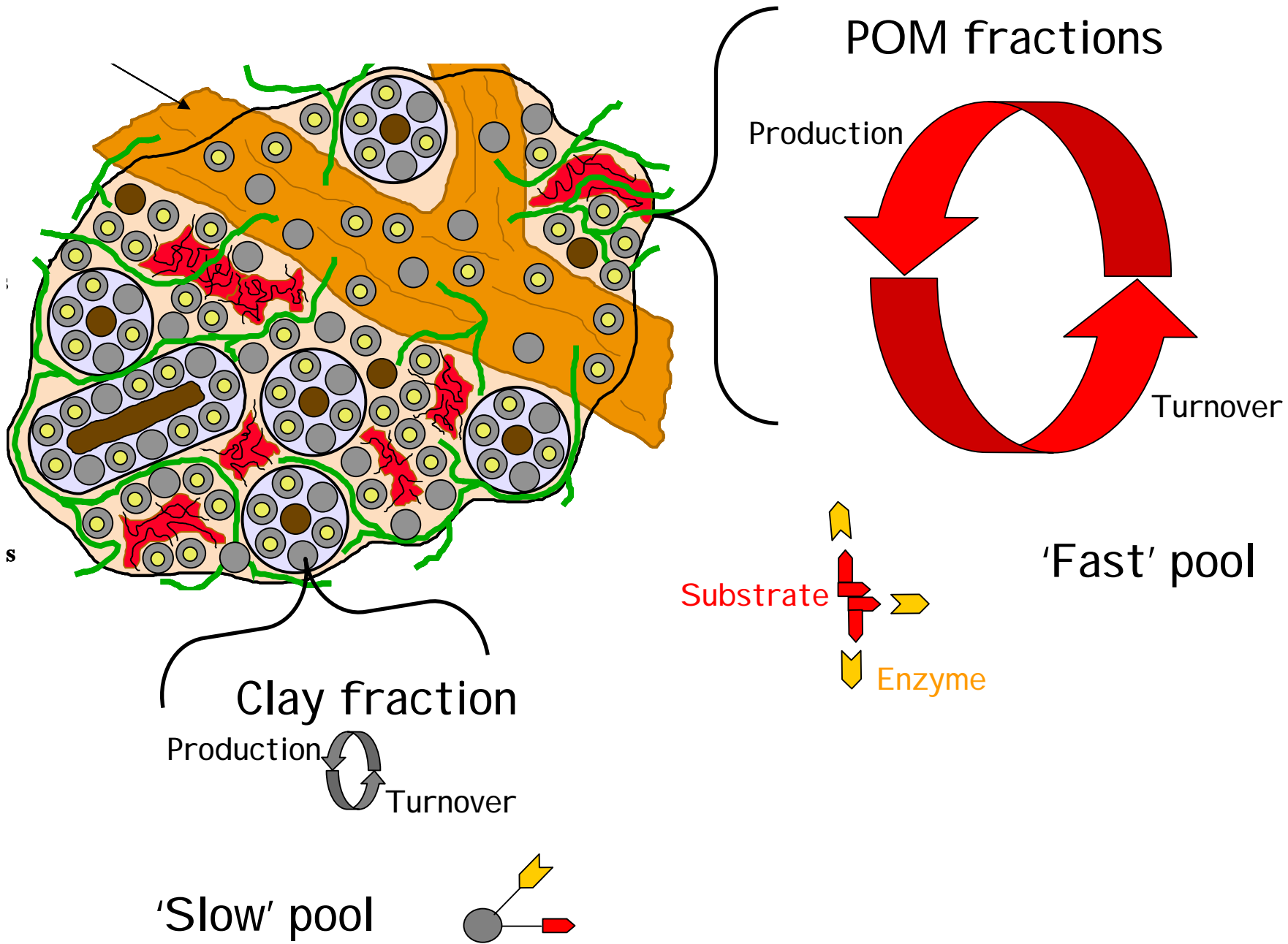
$$\text{Activity} = \frac{\text{Production}}{\text{Turnover}}$$

Enzyme turnover versus production



Concluding thoughts

- High production of specific enzymes on organic substrates
- Aggregates contain enzymes
- Substantial activity in old C fractions: conundrum?
- 'Two pool' model of enzyme activity?



Reasons for increasing C with restoration

- Increased/different C inputs
- Spatial isolation of enzymes and substrates
- Not a reduction in enzyme production or activity
- Importance of physical mechanisms

Thanks:

- DOE-GCEP
- Julie Jastrow
- Peter Vitousek