

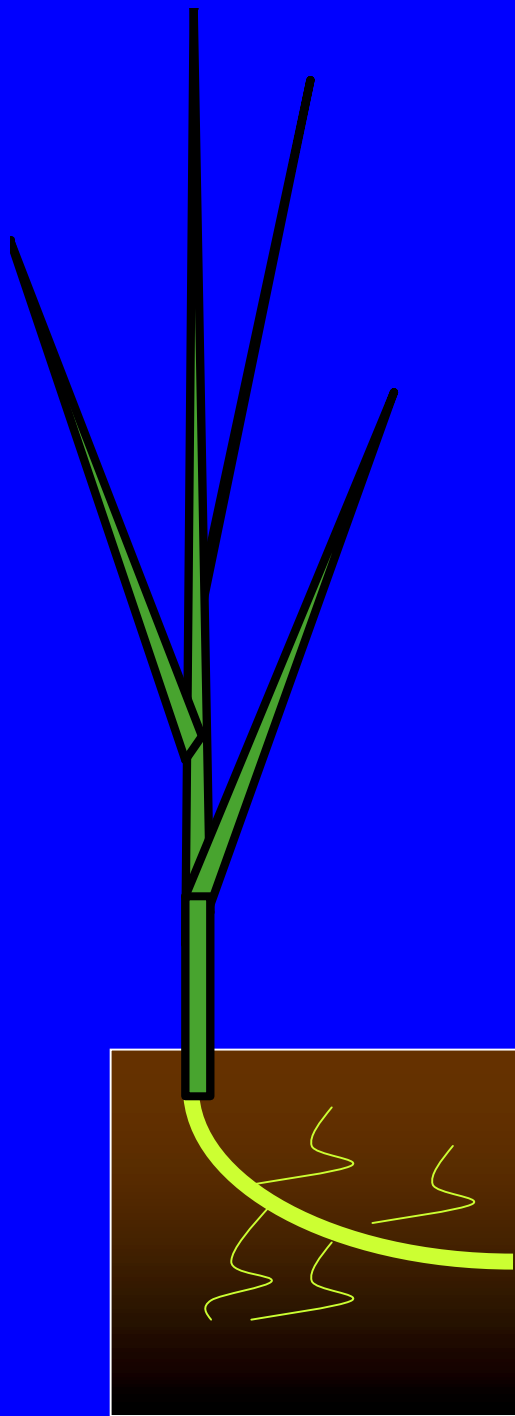
Oxidation of sediments by  
*Spartina* marsh grasses: Potential  
facilitation of pollutant  
degradation

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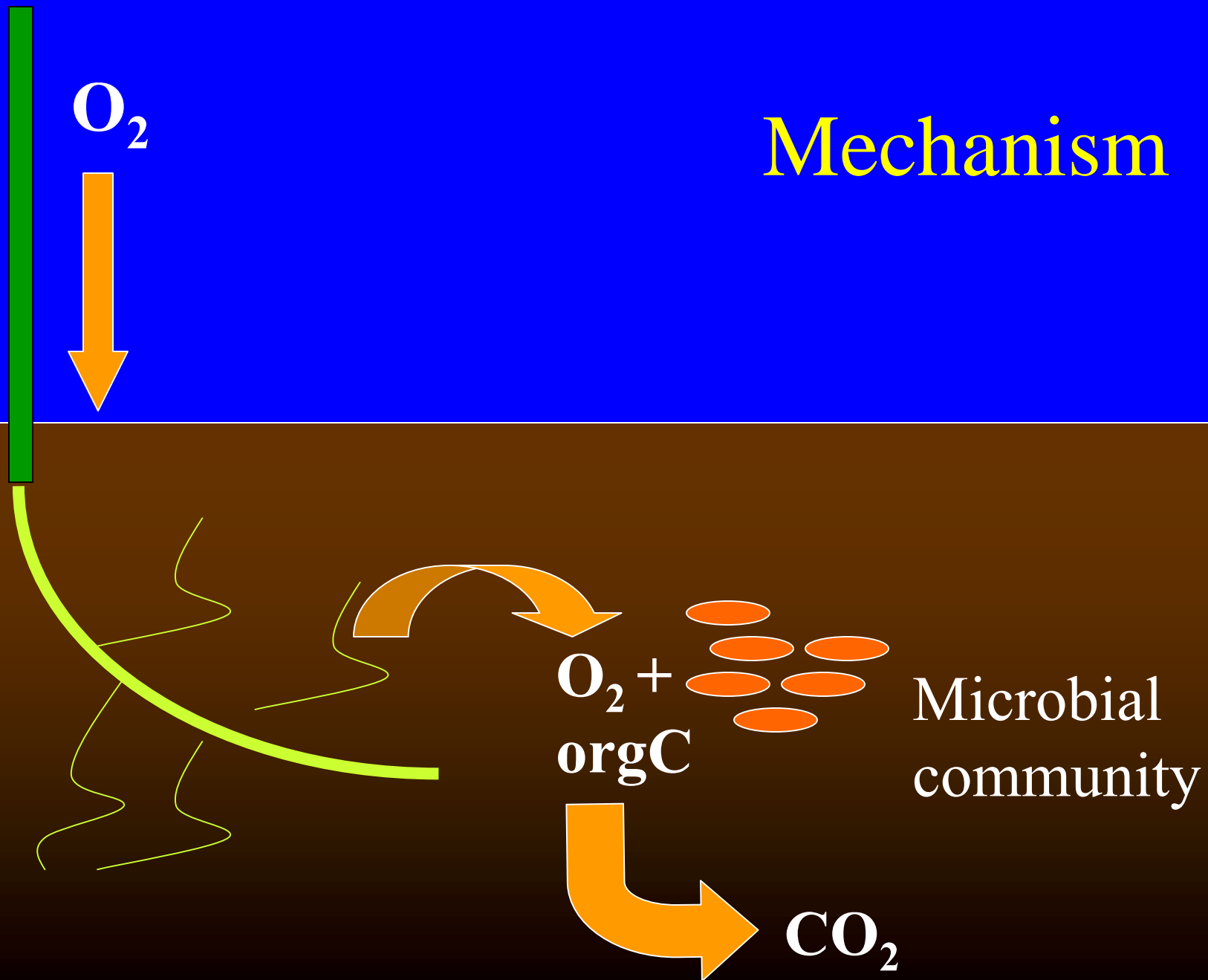
Marsh grasses may transport oxygen to the rhizosphere that can facilitate aerobic degradation of organic pollutants



Oxidized zone

Anoxic zone

# Mechanism



# Oxidation of sediments by *Spartina* marsh grasses: Potential facilitation of pollutant degradation

- Introduction - a highly vigorous plant
- Gas conducting tissues
- Gas transport
- Testing for accelerated degradation

# *Spartina alterniflora* and *Spartina anglica*



- Saltmarsh grasses native to the Eastern U.S. (*S. alterniflora*) and British Isles (*S. anglica*).
- Invasive species in Puget Sound and Willapa Bay in Washington State.

# Spartina are physiologically resilient and vigorous

- Physiological tolerance
  - Wide range of salinities
  - Waterlogged soils
    - Anoxia
    - Hydrogen sulfide
- Fast rate of spread - average 10-20% per year (can be much greater)



**One Year-old Seedling**



**Two Year-old Seedlings**



**Ten Year-old Meadow**



**Three to Four Year-old Clone**





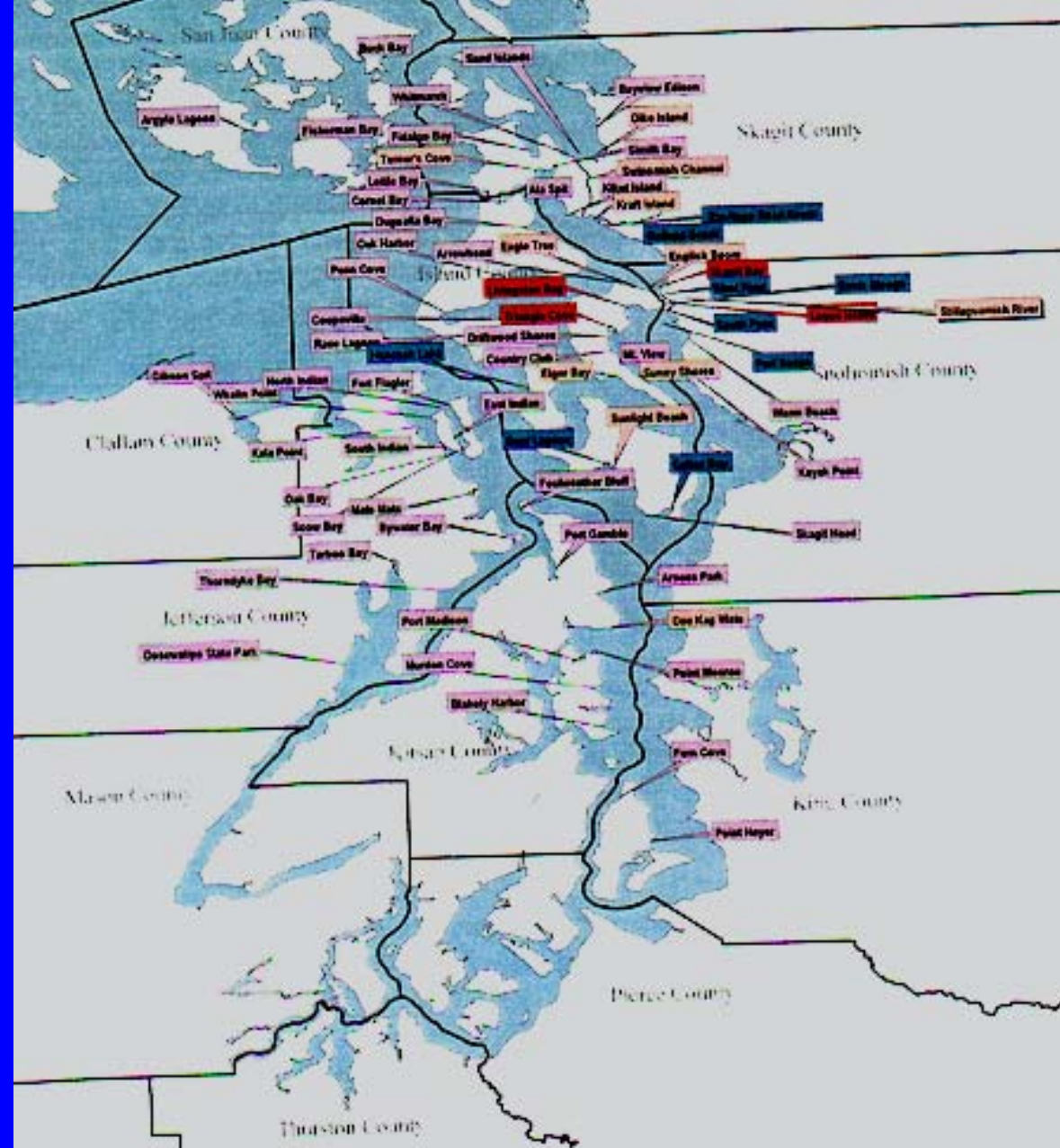
# Willapa Bay

- *Spartina alterniflora*
- 10,000 to 25,000 acres affected
- Nearly 4000 solid acres



# Puget Sound

- *Spartina anglica*
- $\approx 7000$  acres affected
  - Solid acres:
    - Pink= $<1$
    - Peach= 1-5
    - Blue=6-100
    - Red= $>100$

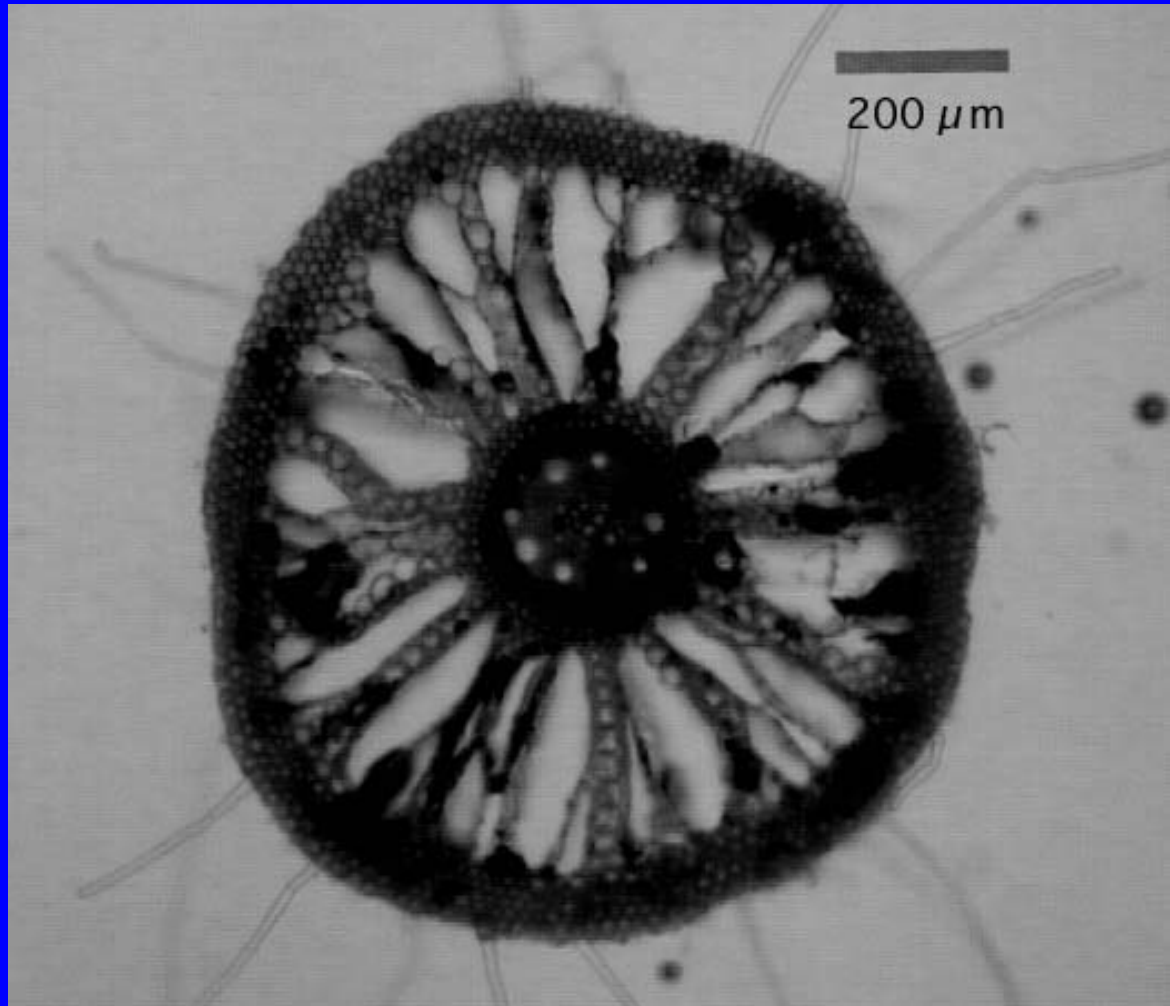


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# Quantification of gas conducting structures

# Tolerating anoxic sediments

- Aerenchyma
- Anaerobic metabolism
  - Alcohol dehydrogenase
- Sulfide oxidation



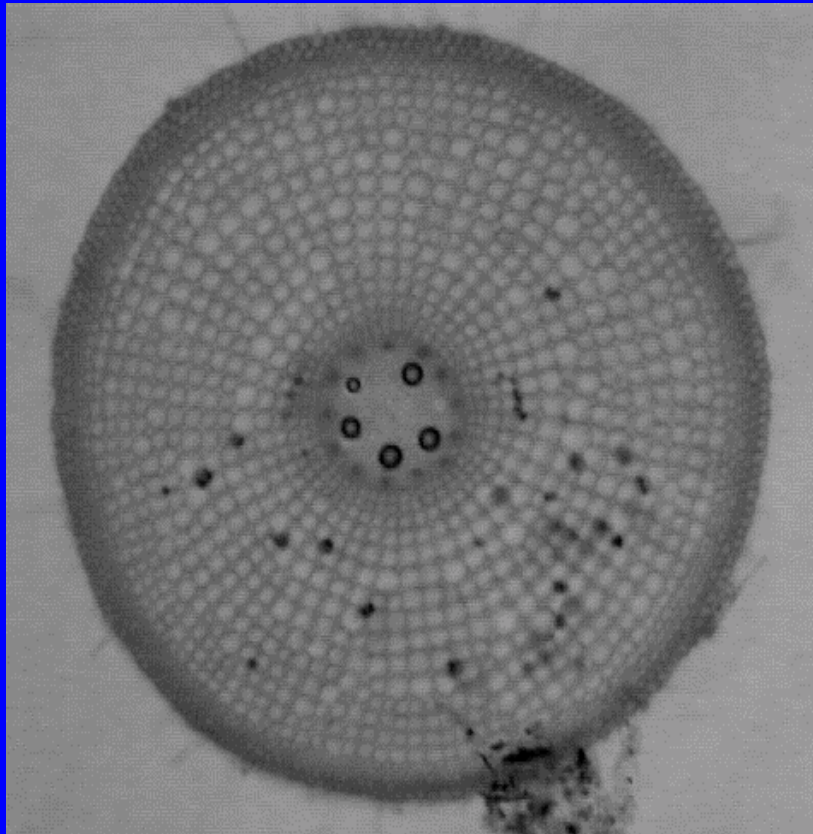
*Spartina anglica* root

# Root Structure During Flooding

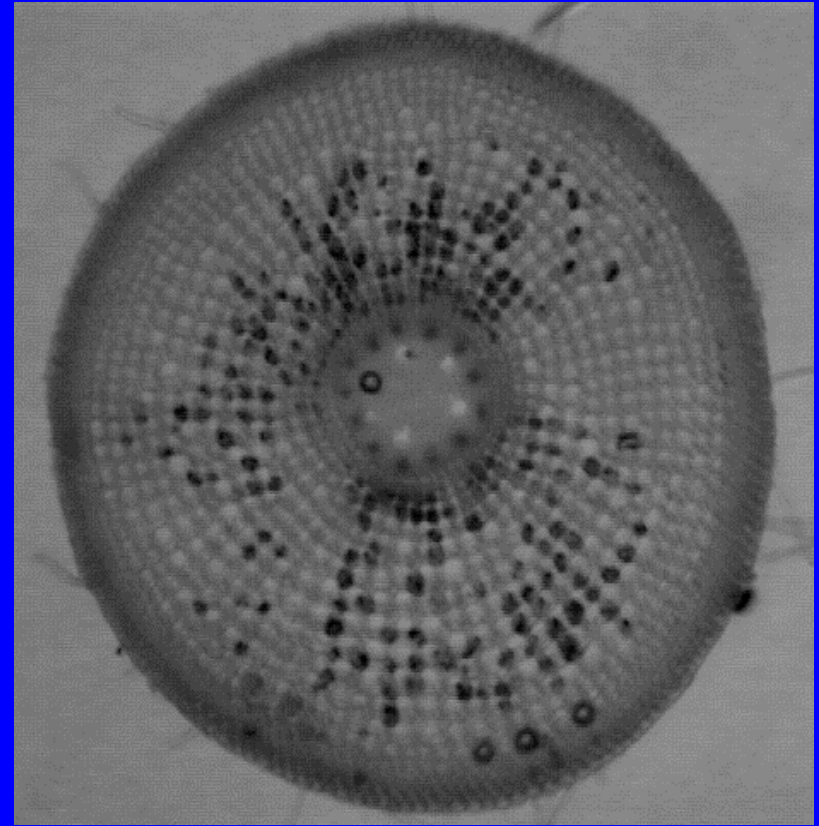
- During flooding roots are deprived of oxygen
- An aerenchyma system is developed to overcome deficiencies
- Root samples were taken from various treatments of plants, and fixed for microscopy
- Roots were sectioned serially, and examined by light microscopy

# *Spartina anglica* Root Structure

1 cm from root tip



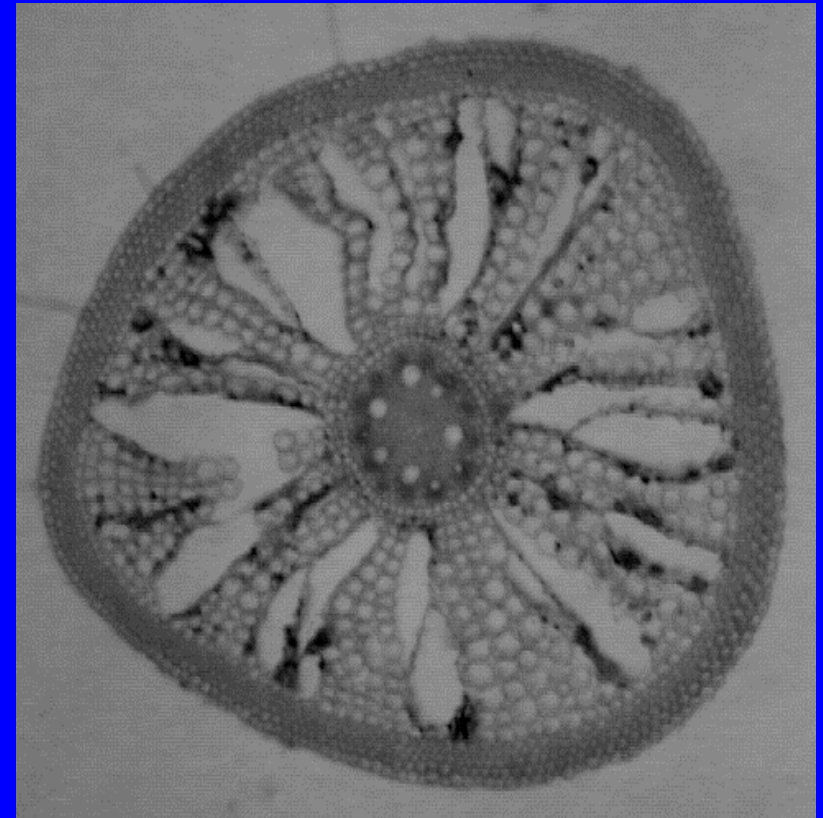
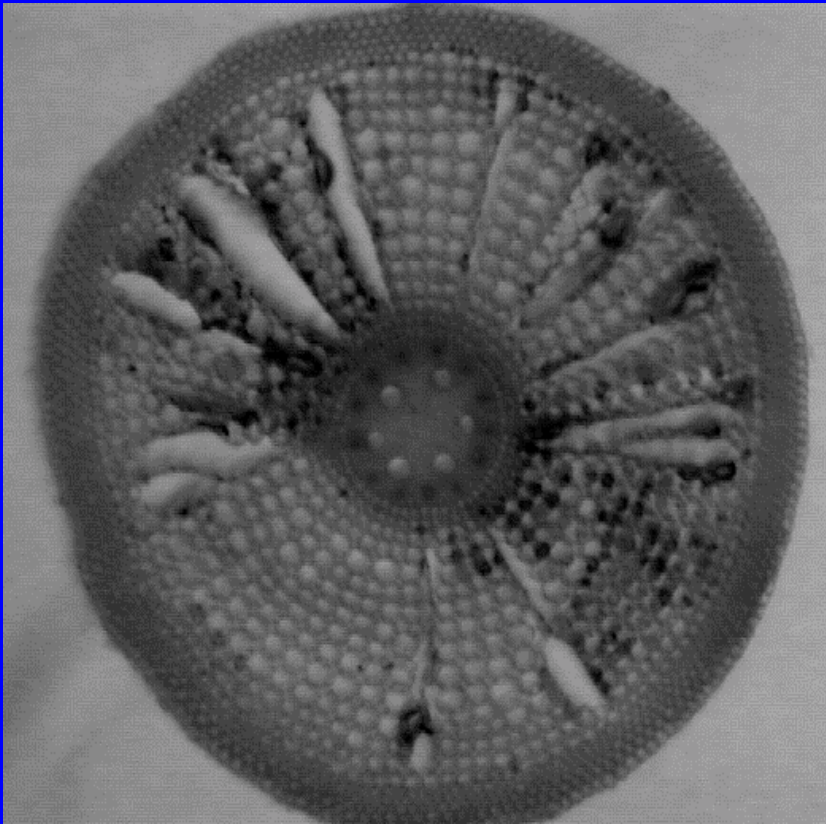
2 cm from root tip



# *Spartina anglica* Root Structure

4 cm from root tip

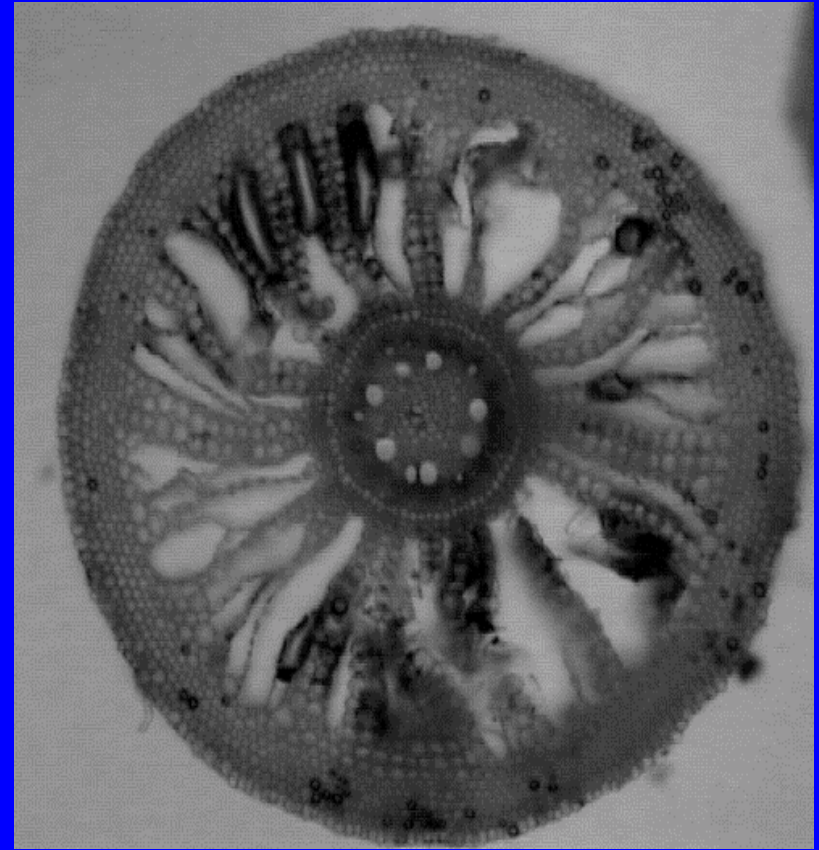
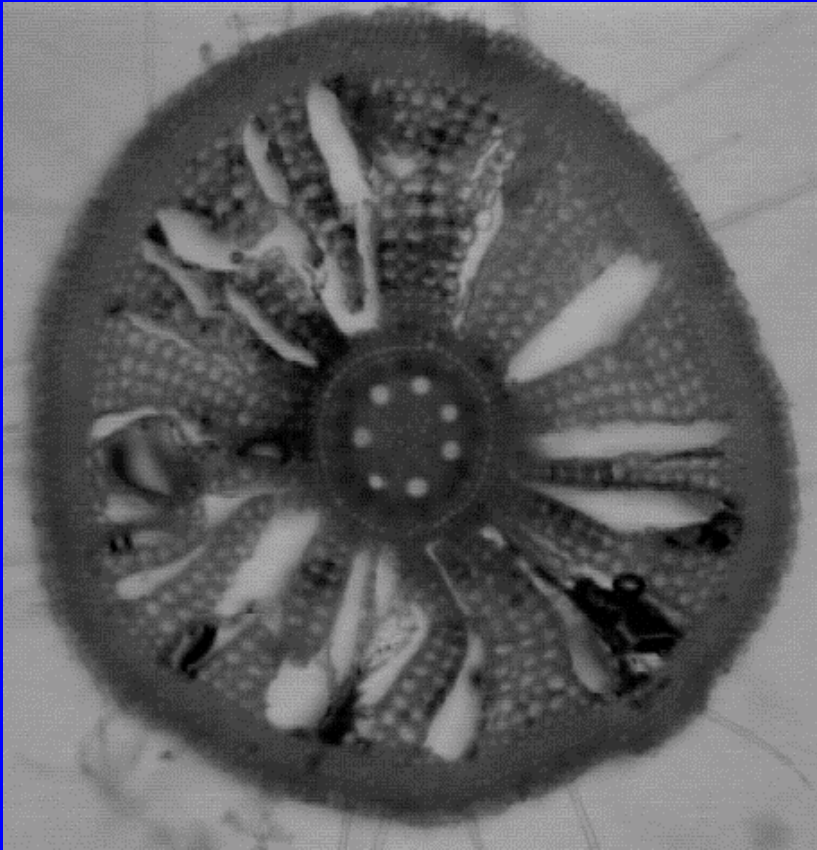
6 cm from root tip



# *Spartina anglica* Root Structure

8 cm from root tip

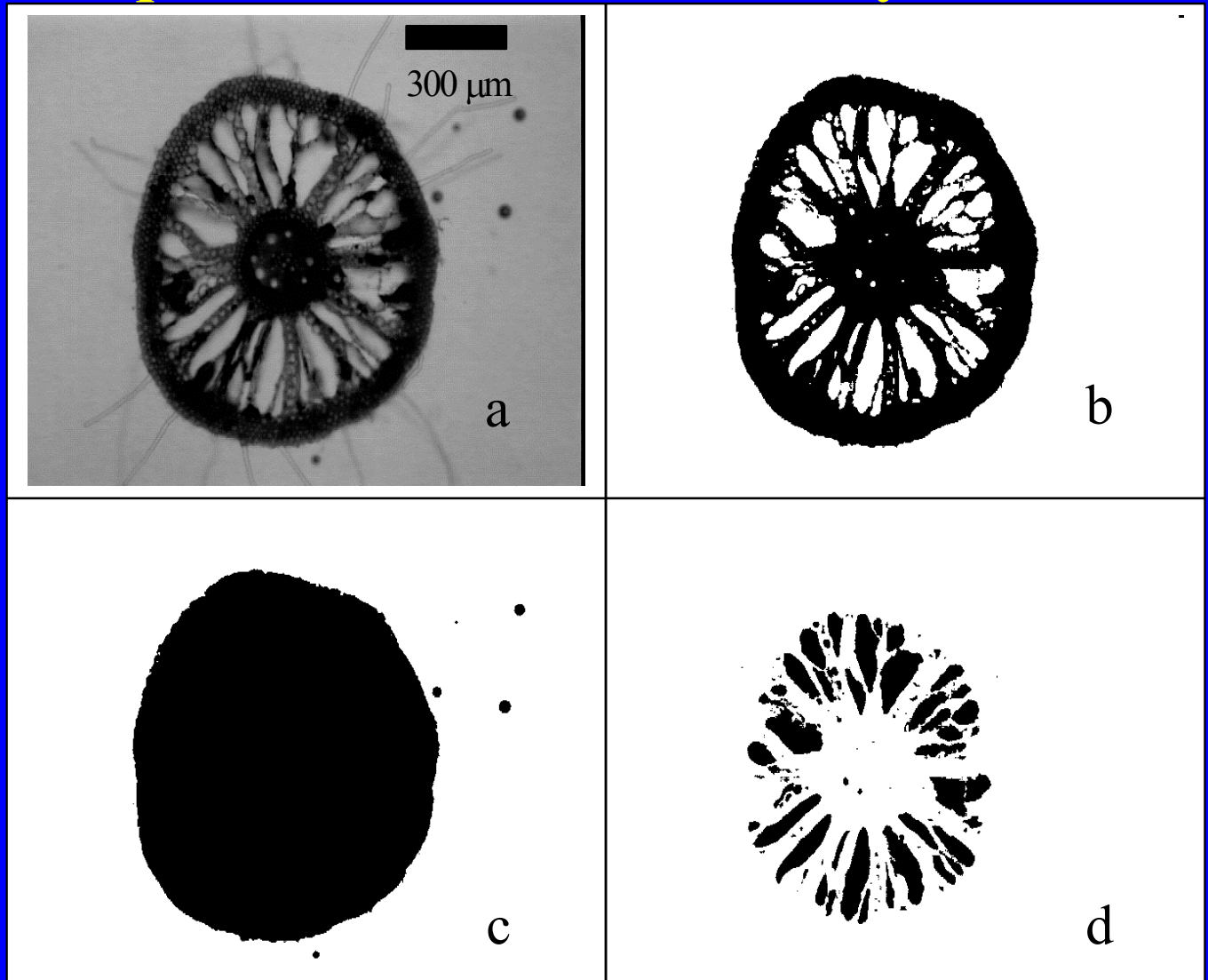
10 cm from root tip



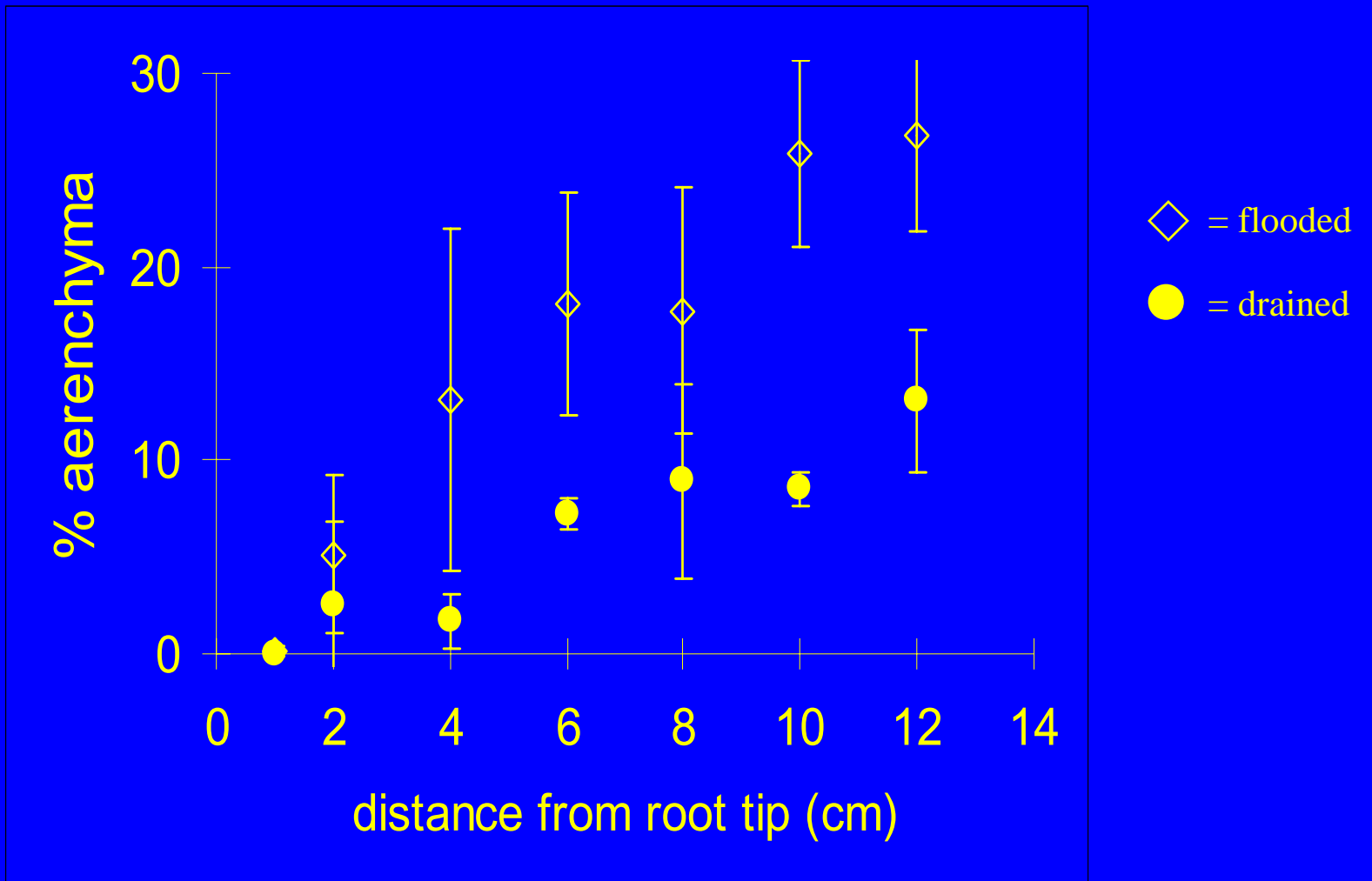


# Digital quantification of aerenchyma

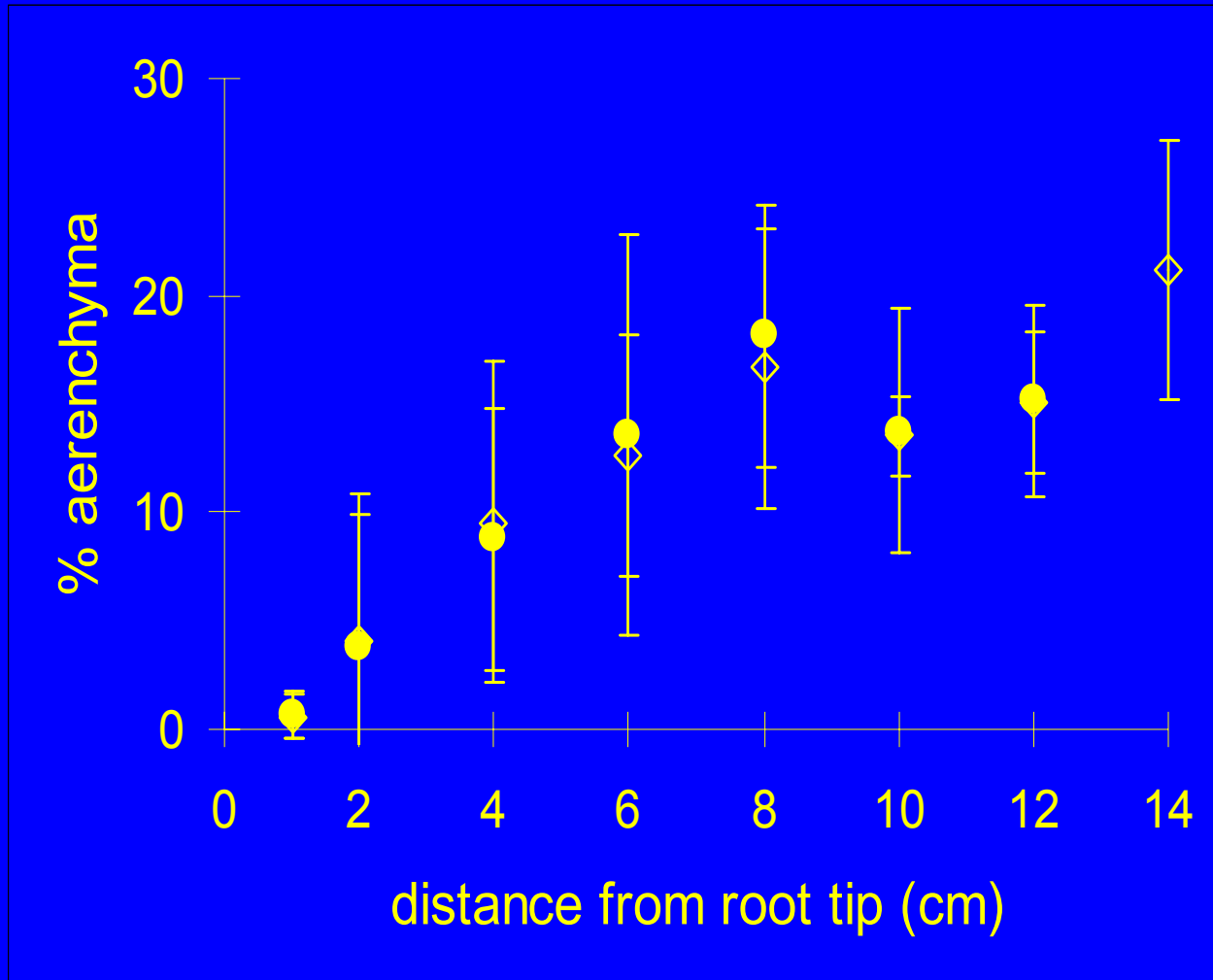
Percent aerenchyma equals the number of pixels in “d” divided by the number of pixels in “c.”



# *S. alterniflora* develops more aerenchyma when flooded



# *S. anglica* does not develop more aerenchyma when flooded



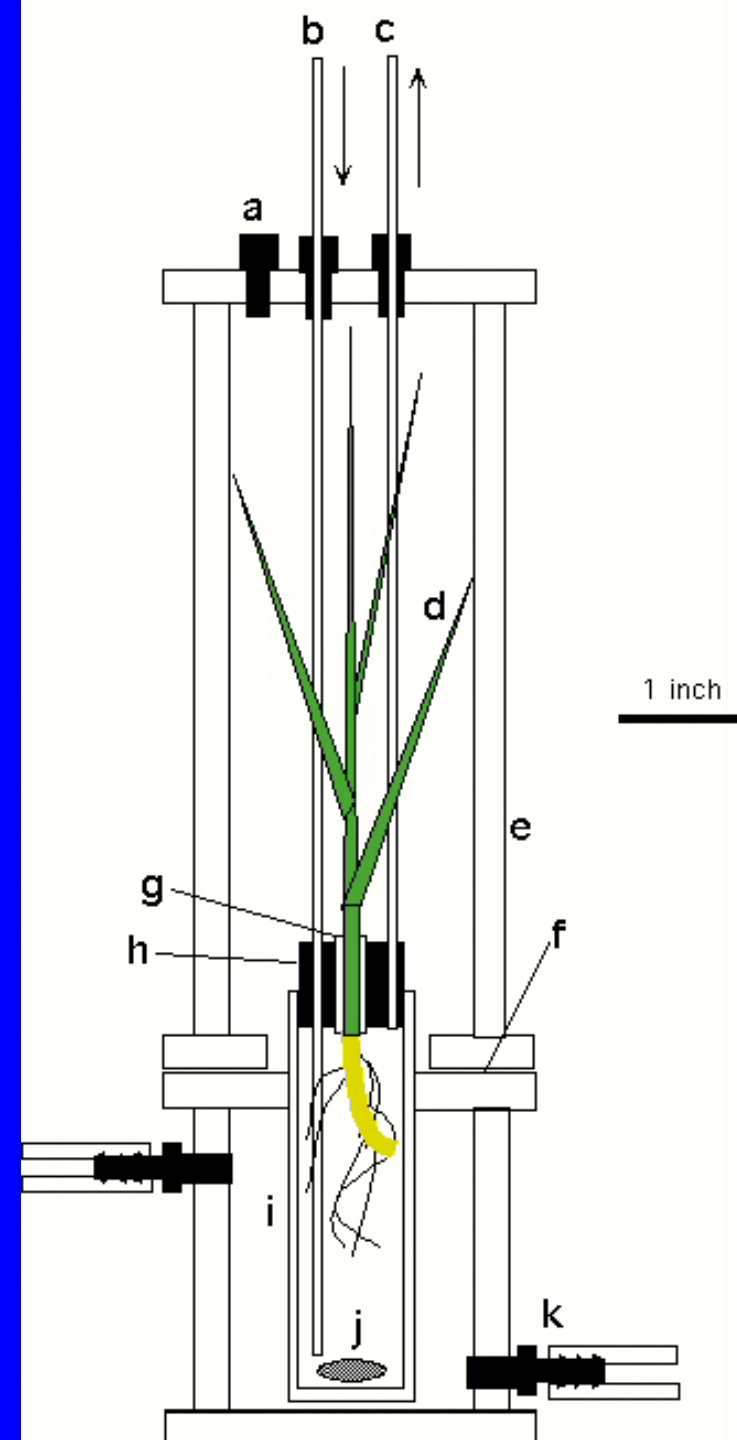
# Root Structure Results

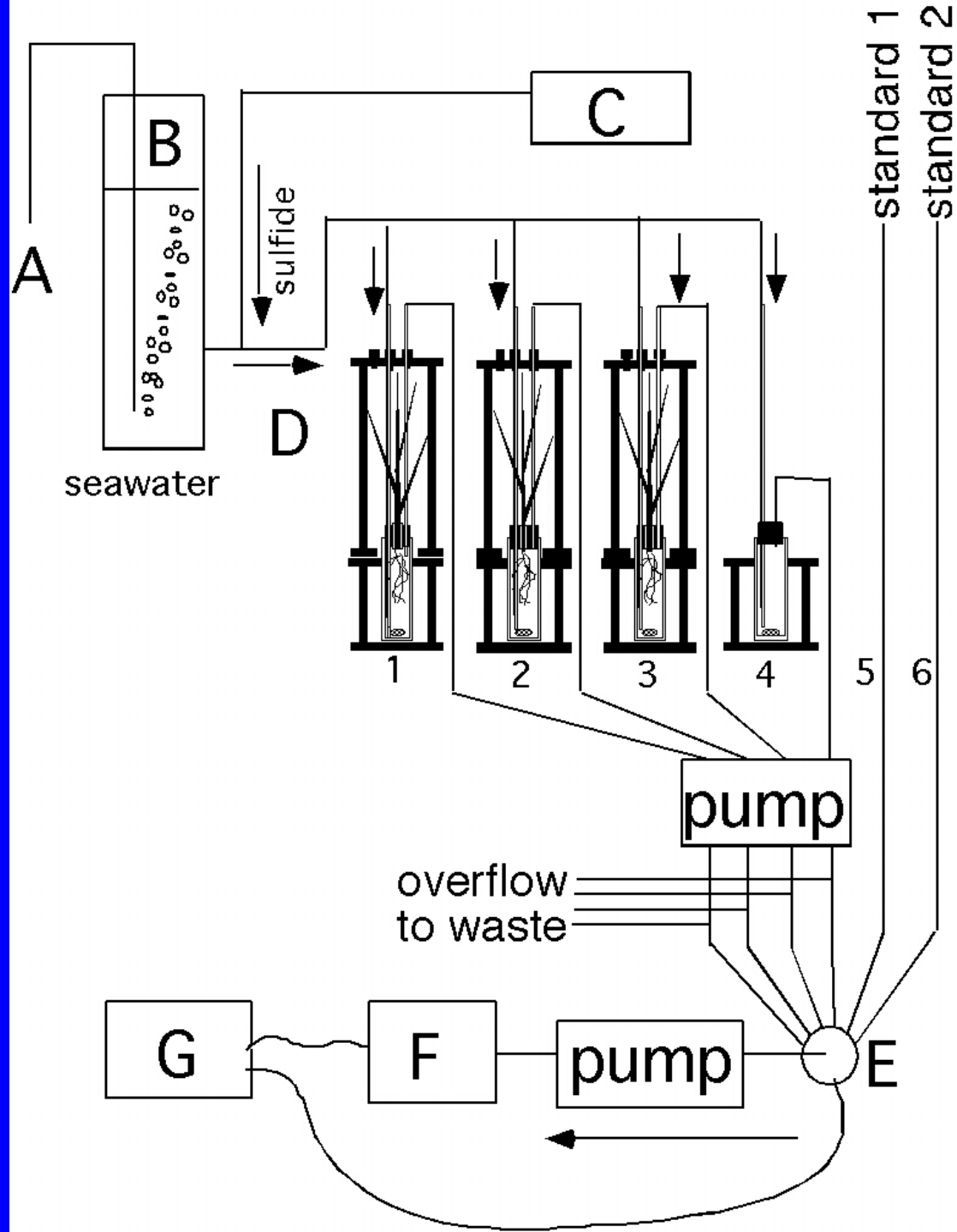
- *Spartina alterniflora* roots become stressed with the onset of flooding and form more aerenchyma.
- *Spartina anglica* plants were not stressed by flooding! They do not form aerenchyma even when confronted with flooding stress.

# Quantifying oxygen transport

# *S. anglica* respirometry experiments

- Use automated flow-through respirometry system
- Quantify root  $O_2$ ,  $H_2S$ ,  $CO_2$ ,  $NH_3$  fluxes
- Aerenchyma gas transport
- Sulfide detoxification



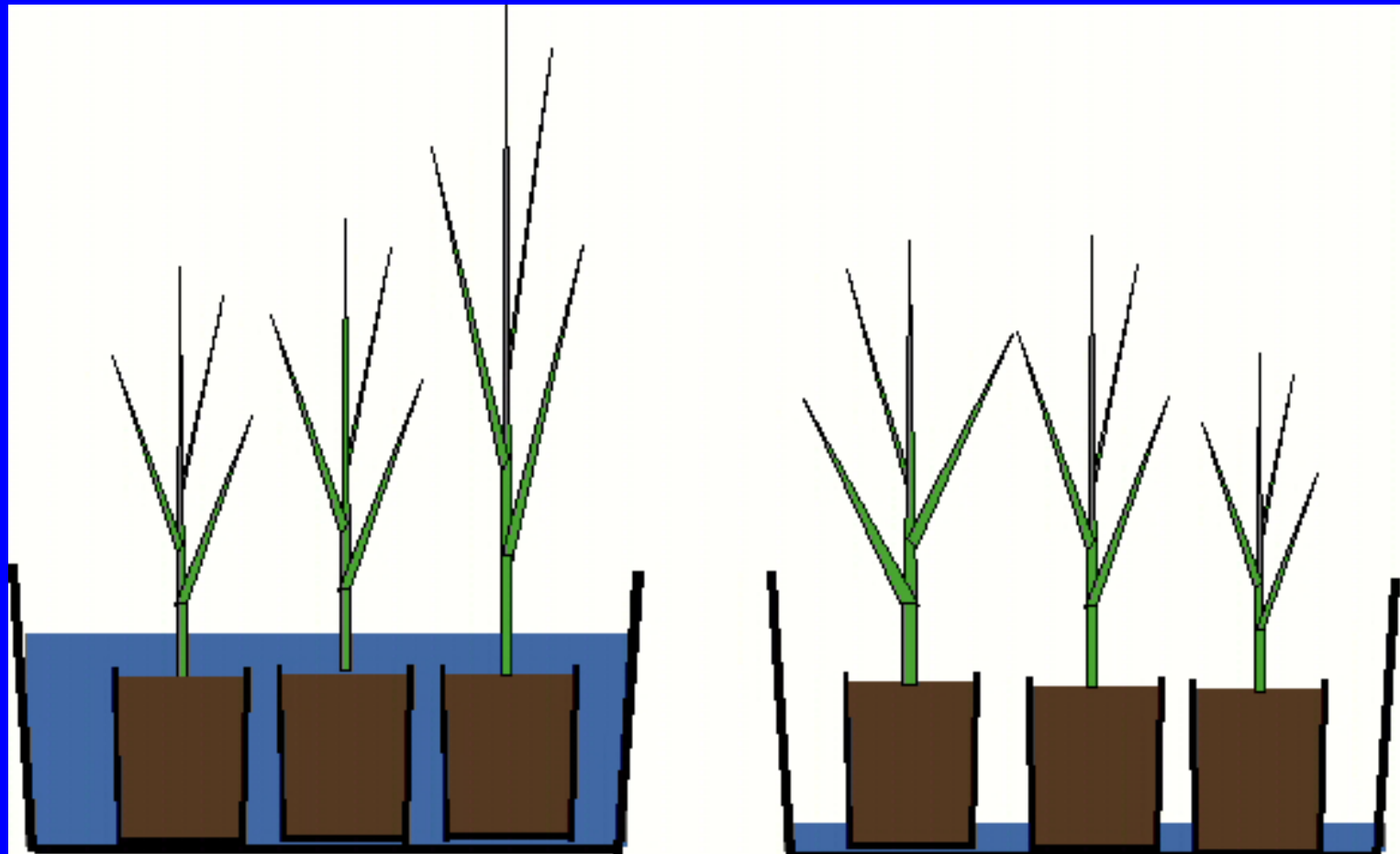


# Flow-through respirometry





# Compare plants grown under different conditions



flooded

drained

# Flooded plants consume less oxygen from the medium

Rates of root oxygen flux in *Spartina anglica* grown under drained and flooded conditions. Negative fluxes denote uptake from the medium. Values given are mean  $\pm$  SD (n).

O<sub>2</sub> flux ( $\mu\text{mol g}^{-1} \text{h}^{-1}$ )

Flooded

Drained

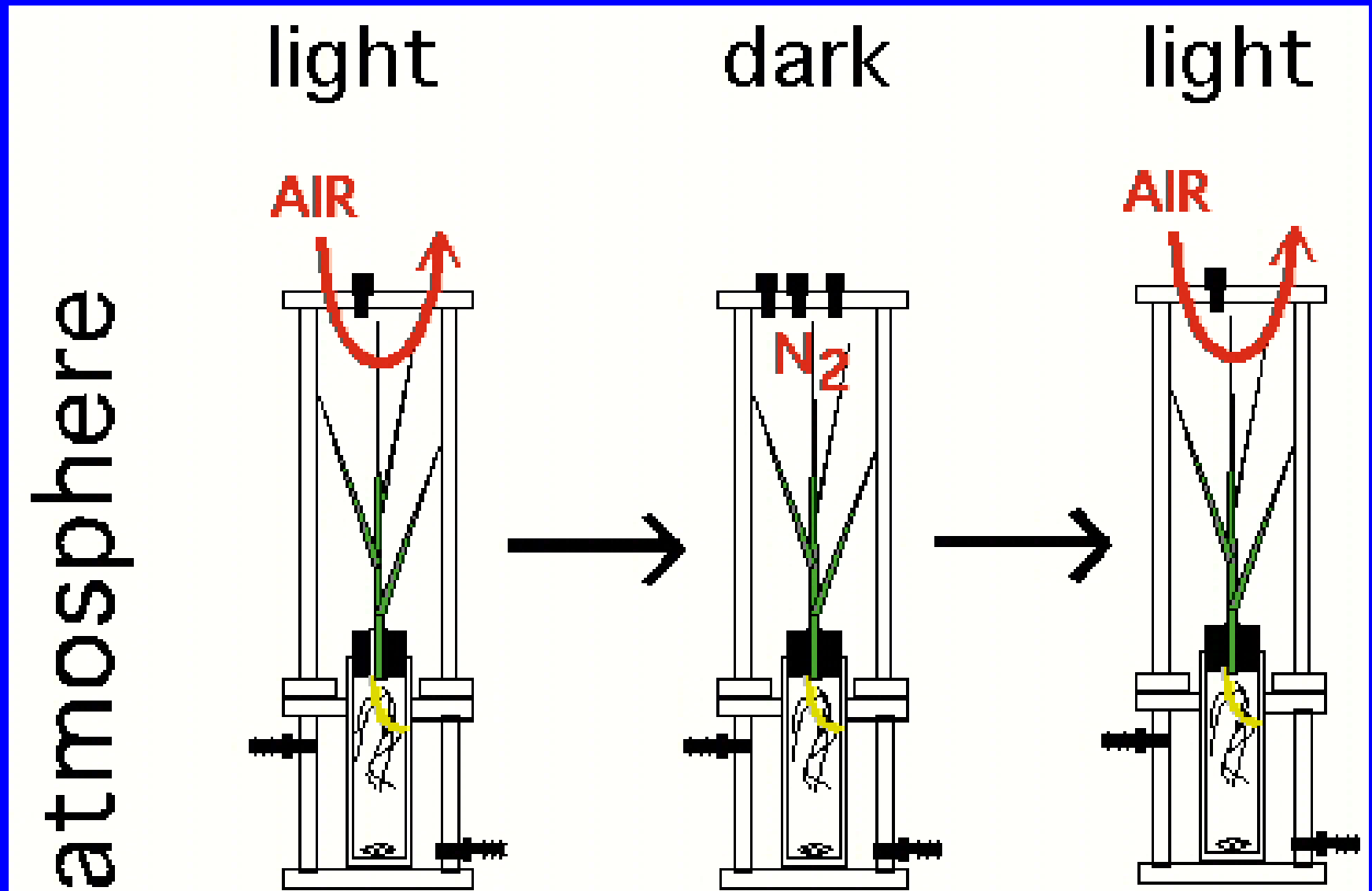
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**-0.02  $\pm$  0.49 (16)**

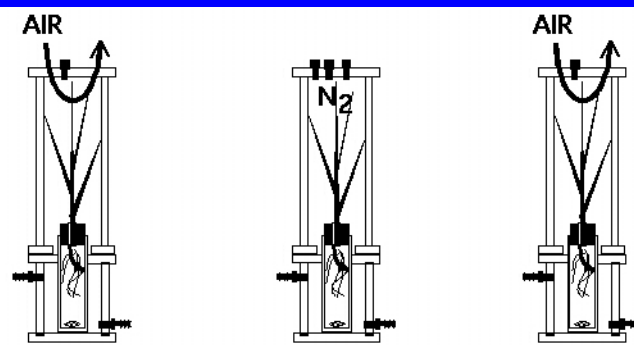
**-0.70  $\pm$  0.50 (8)**

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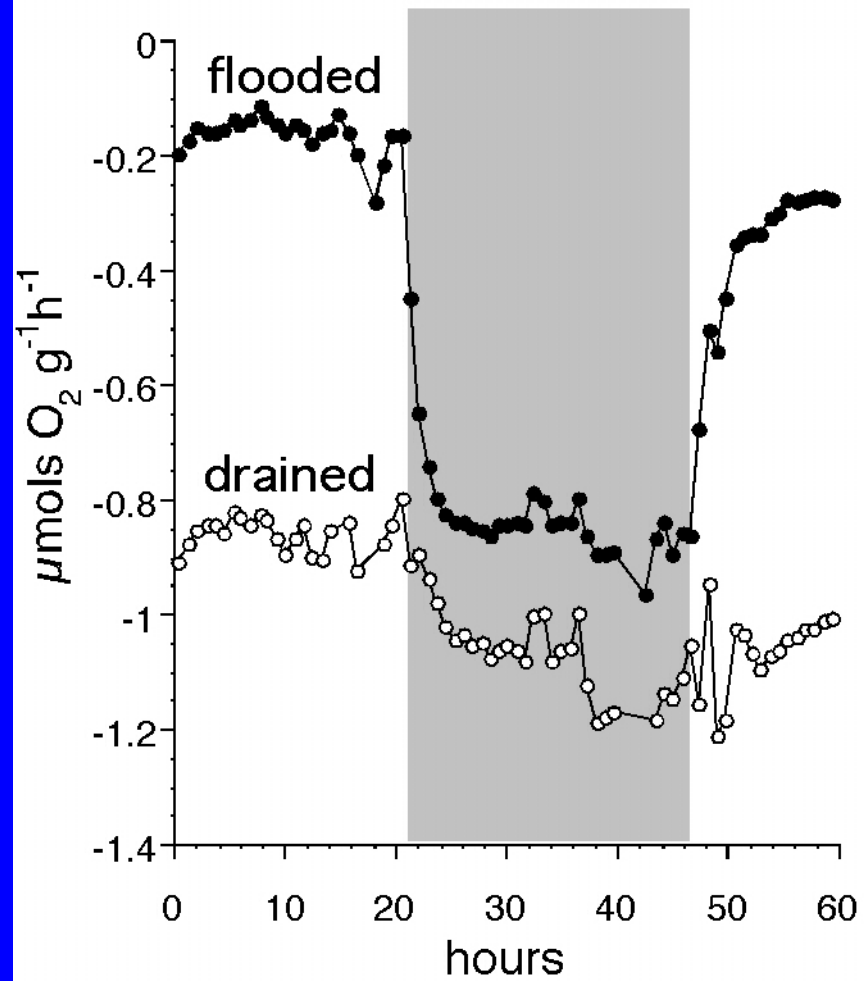
# Quantifying aerenchyma O<sub>2</sub> transport

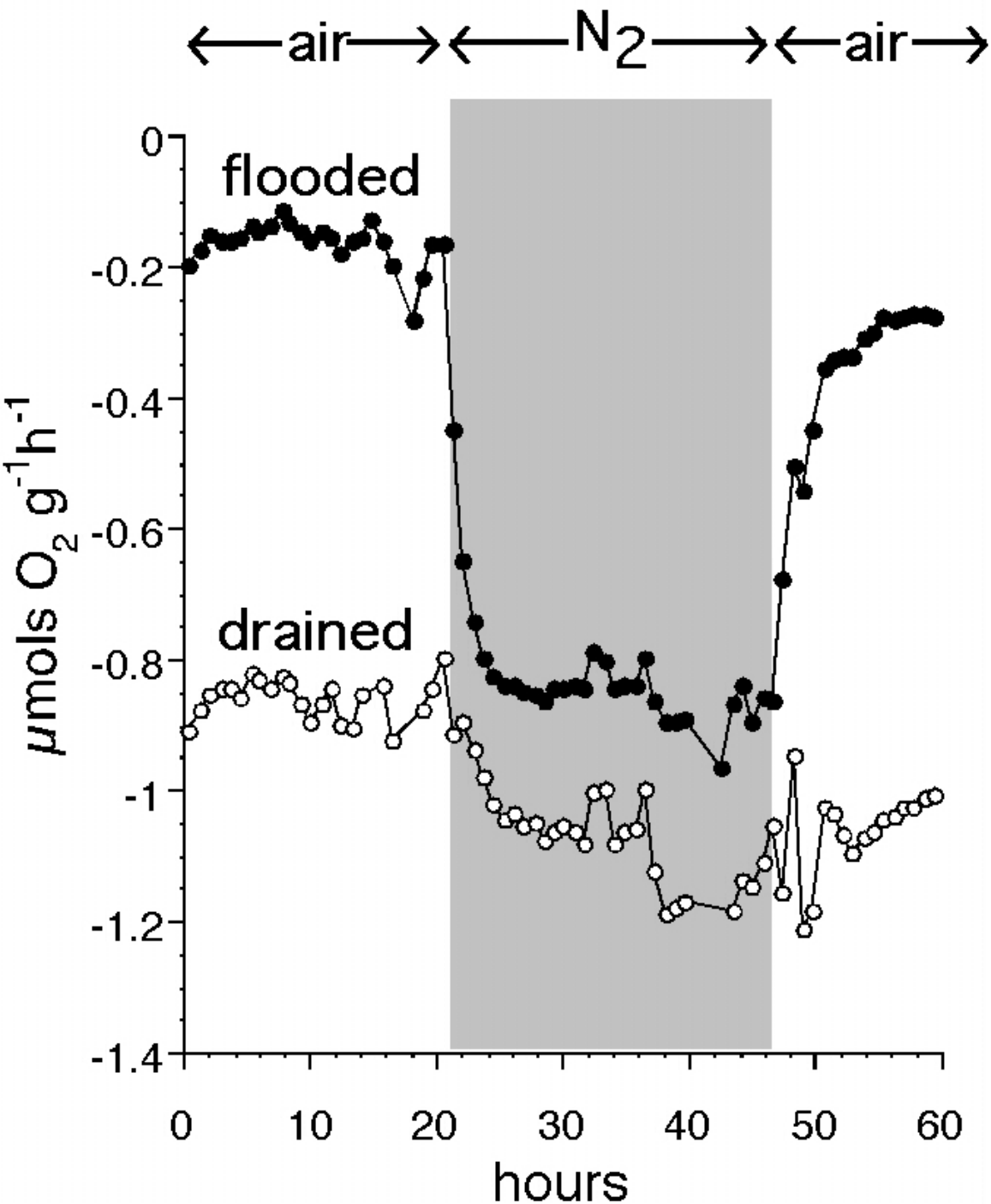


atmosphere



← air → ← N<sub>2</sub> → ← air →





- Aerenchyma O<sub>2</sub> transport is high (0.6  $\mu\text{mols g}^{-1} \text{ h}^{-1}$ ) in flooded plants and negligible in plants grown under drained conditions

# Summary of respirometry results

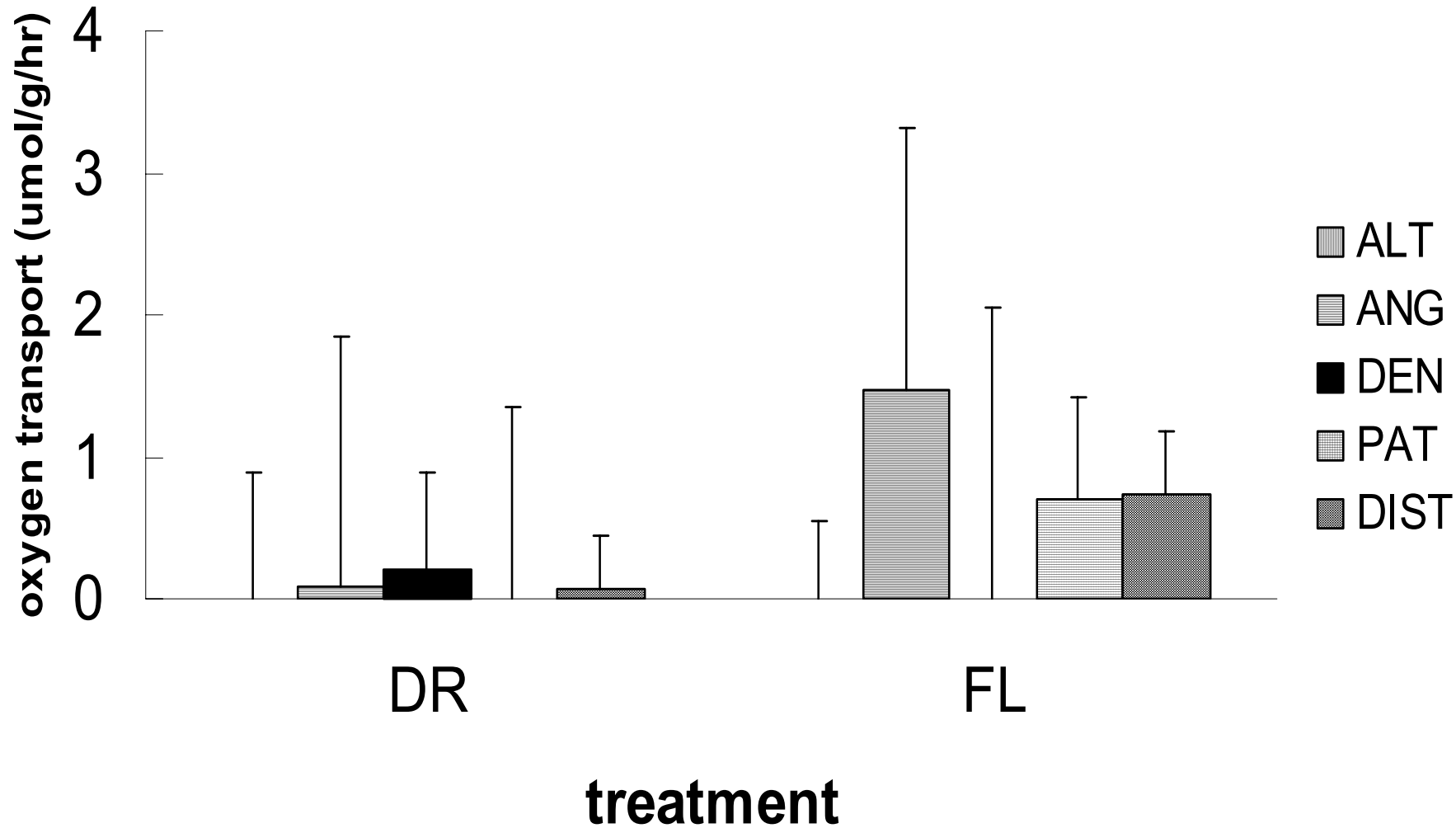
## Gas Fluxes to the Rhizosphere\*

Species/Treatment	Air atmosphere ( $\mu\text{mols g}^{-1} \text{h}^{-1}$ )	sealed N <sub>2</sub> atmosphere ( $\mu\text{mols g}^{-1} \text{h}^{-1}$ )
O <sub>2</sub> flux		
<i>Spartina anglica</i> flooded	<b>0.110 ± 0.324 (13)</b>	<b>-0.830 ± 0.083 (3)</b>
<i>Spartina anglica</i> drained	<b>-0.523 ± 0.355 (5)</b>	<b>-0.826 ± 0.302 (4)</b>
<i>Spartina alterniflora</i> flooded	<b>-2.689 ± 1.512 (6)</b>	<b>-2.562 ± 1.612 (6)</b>

Oxygen transport appears to be  
more effective in  
*S. anglica* compared with *S.*  
*alterniflora*

- *S. alterniflora* do not exhibit net release of oxygen to the medium (Howes and Teal, 1994)
- Possible implications for success of *S. anglica* as an invasive species?

# Additional species vs. *S. anglica*

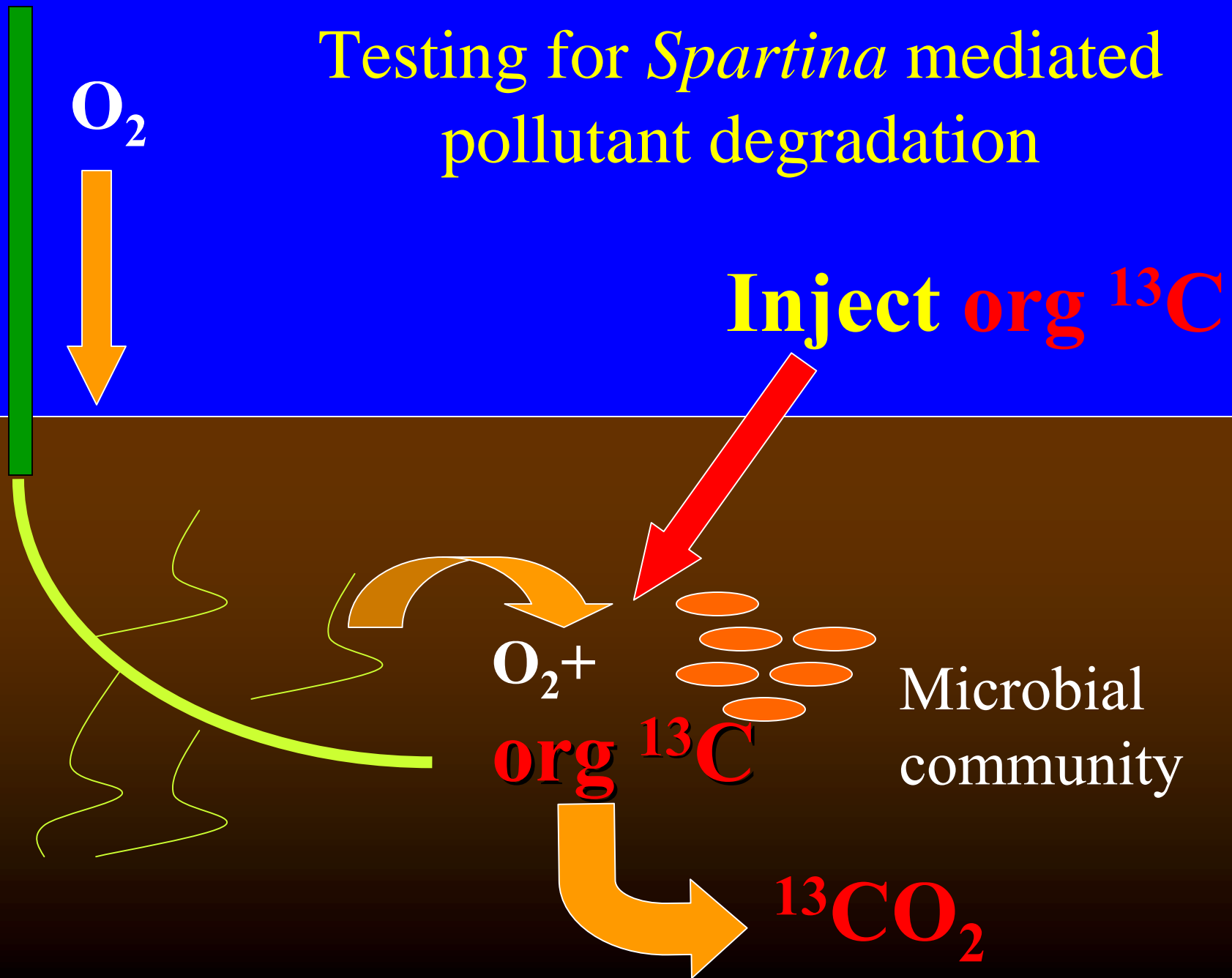




# *S. anglica* sediment oxidation

Treatment/Habitat	Sediment Redox Potential (mV)	Porewater Sulfide ( $\mu\text{M}$ )
Intact <i>S. anglica</i>	$-183.38 \pm 170.82$ (8)	$643.55 \pm 539.61$ (10)
Bare Mudflat	$-291.67 \pm 56.61$ (3)	$737.14 \pm 1296.50$ (6)
Mowed to Mud	$-326.00 \pm 68.07$ (5)	$747.58 \pm 473.03$ (8)
Mowed to Stubble	$-316.60 \pm 27.63$ (5)	$2205.80 \pm 975.20$ (8)
Mowed Previous Year	$-193.33 \pm 232.41$ (3)	$1114.37 \pm 766.46$ (3)

# Testing for *Spartina* mediated pollutant degradation



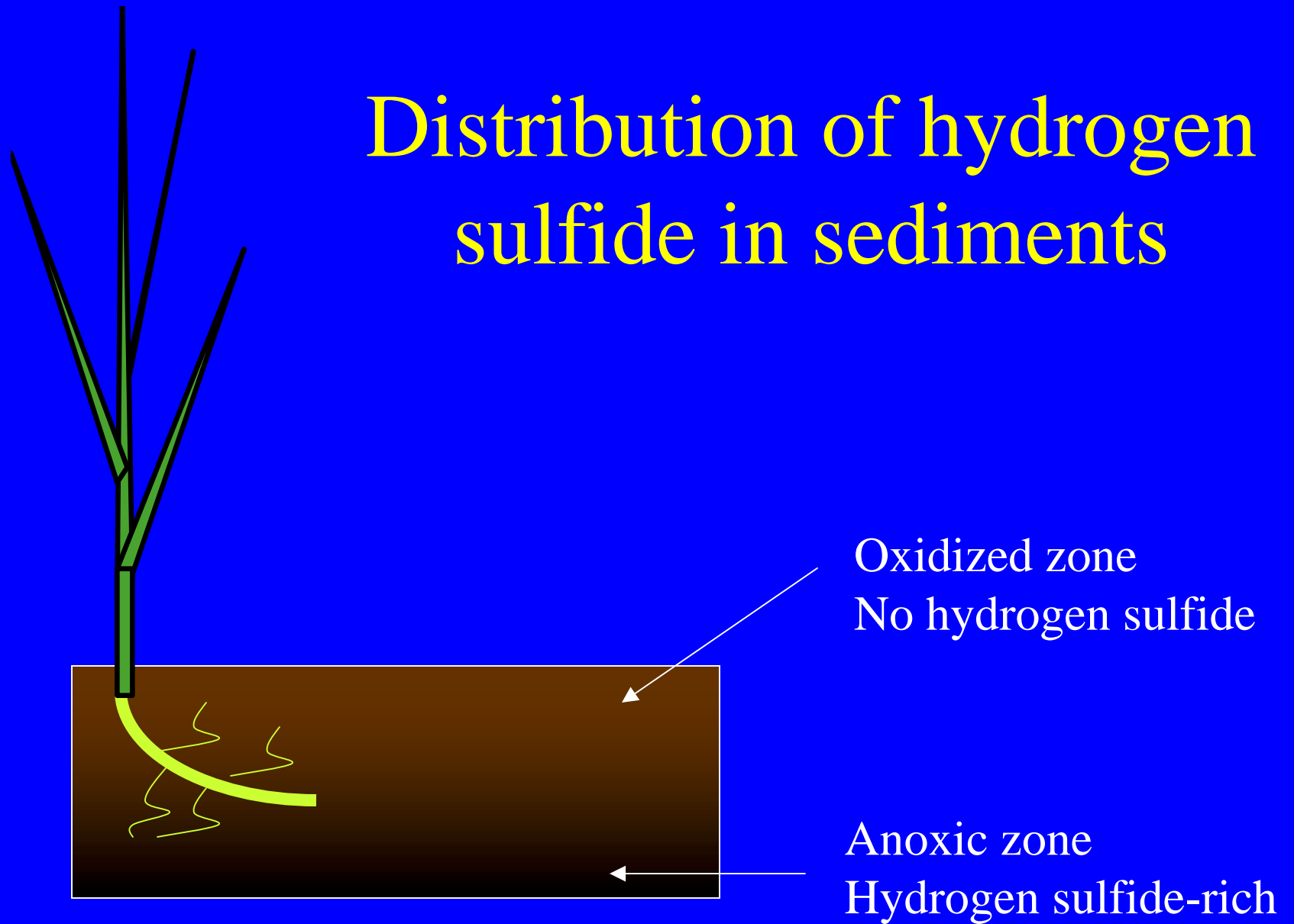
# What substrate to use?

- Commercial availability of labeled compound
- Oxygen required for degradation
- low vapor pressure
- oxidation to carbon dioxide

# Acknowledgments

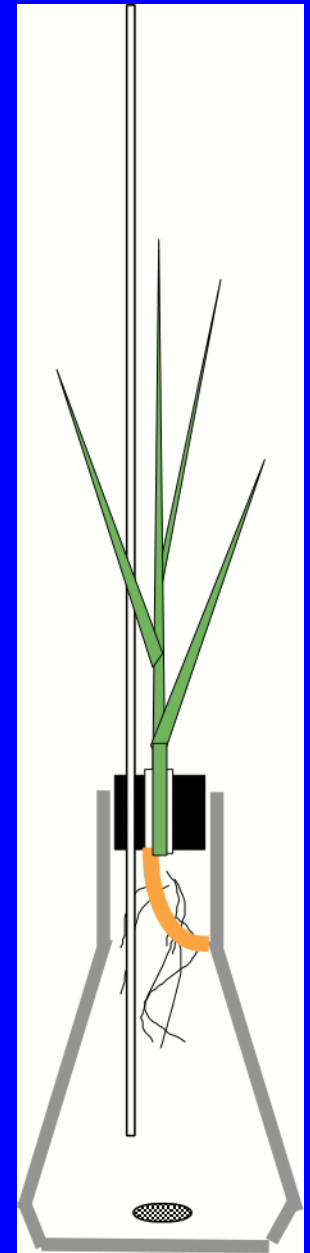
- Brian Maricle - Ph. D. student
  - aerenchyma measurements, some respirometry, environmental measurements
- EPA joint program in phytoremediation/Mitch Lasat
- NSF - development of respirometry system

# Distribution of hydrogen sulfide in sediments



# Checking for oxygen transport

- A plant can be sealed into a flask of  $N_2$ -flushed water
- An oxygen-sensing probe can be used to monitor the water--any increase in  $O_2$  must have come through the plant





## Flow-through chambers

- Quantify  $O_2$  consumption and transport
- Upper enclosure allows plant to be subjected to numerous conditions:
  - Normal air ( $\sim 21\% O_2$ )
  - $N_2$  gas ( $0\% O_2$ )
  - Under water

