Arbuscular mycorrhizal fungi combined with biosolids and biochar sustained phytostabilization and enhanced soil properties of mine tailings

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Mine Tailings Revegetation Challenges

- Heterogeneity of mine waste
- No universal revegetation strategy exists
- Field scale failure scenarios

- Revegetation plan must be site specific depending on the waste characteristics and extent of toxicity

Phytomanagement Strategies Investigated

- **Revegetation strategies to support sustainable growth:**
  - Amendment strategies
  - Candidate plant species
- **Strategies to prolong amendments benefits by co-applying with carbon-rich materials:**
  - Preserve excess nutrients (N and P)
  - Lower the decomposition rate
- **Revegetation strategies targeting stimulation of soil-functioning processes**
  - Amendment strategies to effectively employ soil key-microorganisms such as mycorrhizal fungi

Long-term Revegetation Study: Role of AMF and Waste Byproducts

Objective 1: Investigate the role of Native AMF strains collected from prairie lands to enhance sustainable growth of prairie species in amended tailings.

Objective 2: Investigate the role of BC and BS to stimulate a beneficial and sustainable AMF symbiotic relationship with prairie plants in amended tailings.

Objective 3: Assess the potential of AMF to stimulate soil formation processes within and below the amended top layer.

Role of Soil Amendments: Potential of Biosolids (BS)

- **Up to 50% organic matter (OM) which serve as:**
  - Carbon and nutrients source for microorganism
  - Binding agents for aggregate formation and stabilization
  - Increasing soil Cation Exchange Capacity (CEC)
  - Increasing water holding capacity (WHC)
  - Metal complexation
  - Reservoir for essential nutrients

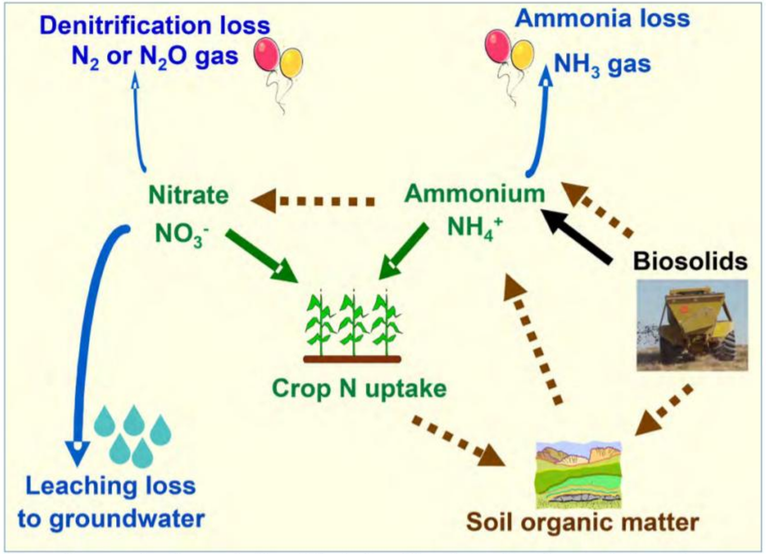
- **A full range of readily available essential nutrients**

❖ **Issues associated with BS application**

- **High application rates required**
- **Limited local BS supplies: many tailings are remote**

Potential Revegetation Failure Mechanisms with BS

➢ Nutrients lability especially N



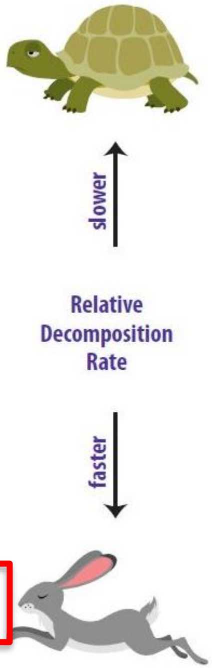
Revegetation requires high BS application rates:

- Excessive nutrient leaching
- Rapid nutrient depletion

➢ Rapid decomposition:

- **Low C:N ratio**

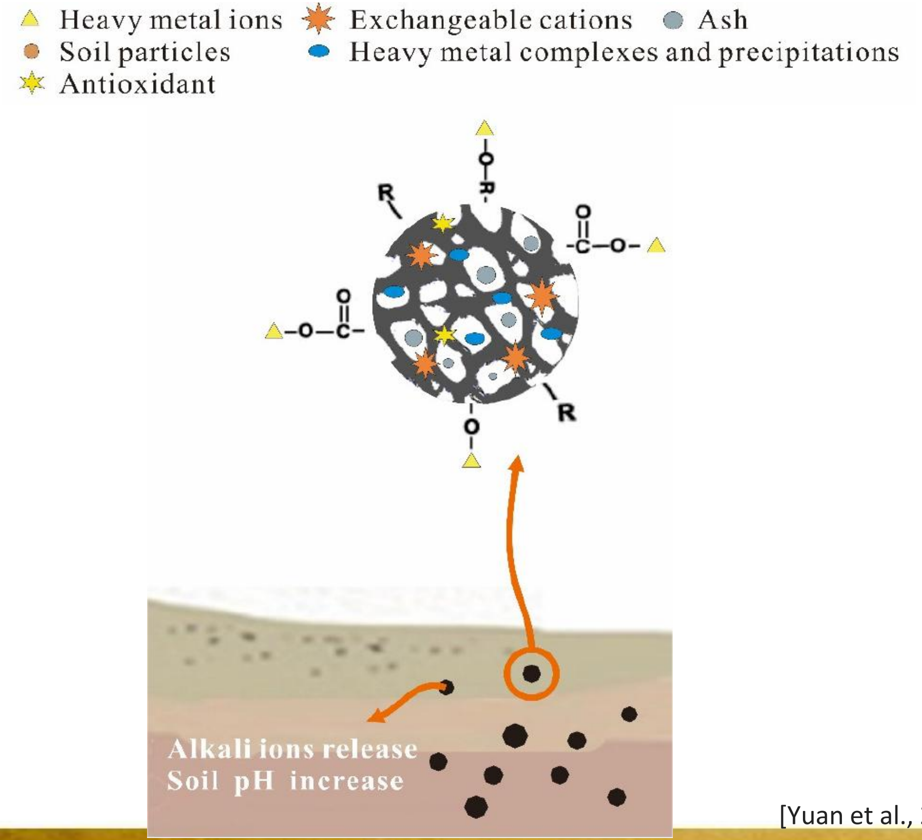
Material	C:N Ratio
rye straw	82:1
wheat straw	80:1
oat straw	70:1
corn stover	57:1
rye cover crop (anthesis)	37:1
pea straw	29:1
rye cover crop (vegetative)	26:1
mature alfalfa hay	25:1
Ideal Microbial Diet	**24:1**
...tted barnyard manure	20:1
leg... hay	17:1
beef manu...	17:1
young alfalfa hay	13:1
hairy vetch cover crop	11:1
soil microbes (average)	8:1



https://catalog.extension.oregonstate.edu/sites/catalog/files/project/pdf/pnw508_0.pdf
https://farmtable.com.au/build/integrating-livestock-into-a-continuous-cropping-system-in-a-high-rainfall-environment/

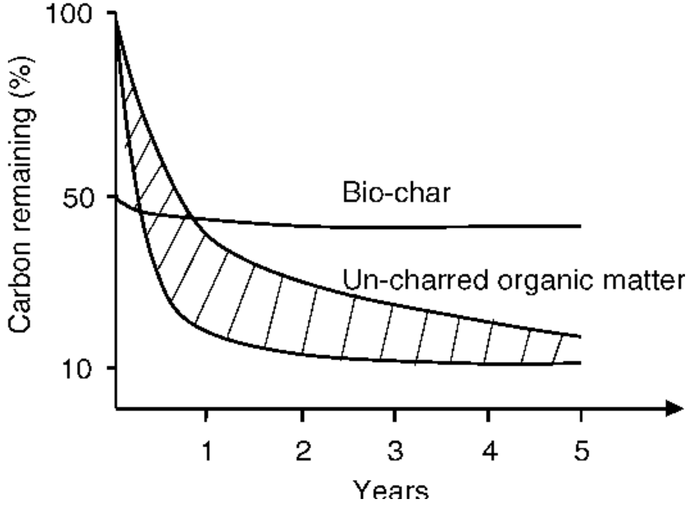
Role of Soil Amendments: Potential of Biochar (BC)

- **High sorption characteristics**



[Yuan et al., 2020]

- **High C:N**
- **Recalcitrant**



[Lehmann et al., 2006]

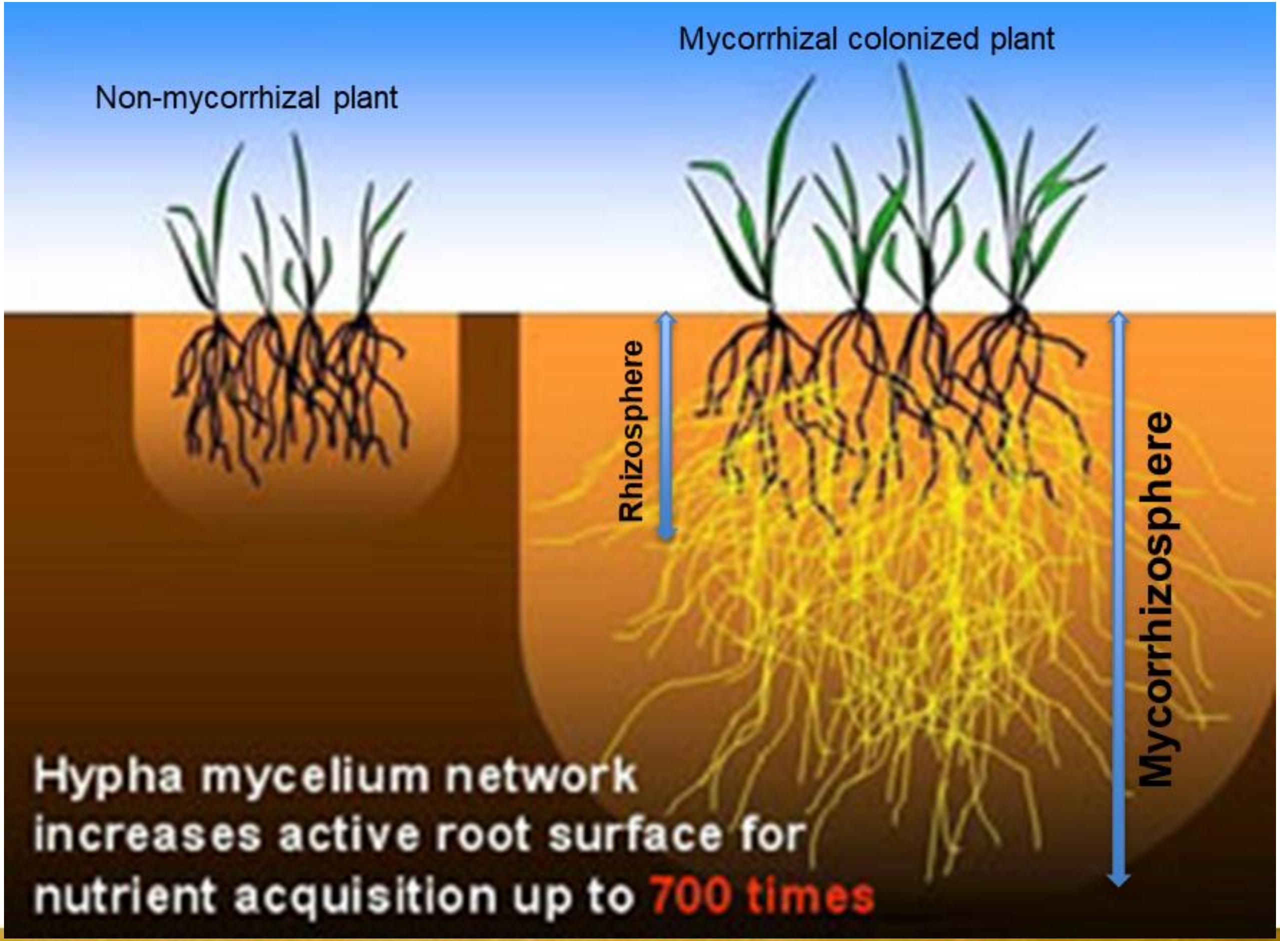
Role of Soil Amendments: Potential of Arbuscular Mycorrhizal Fungi (AMF)

- **Exploring larger soil volume**

- **Water absorption** ↑

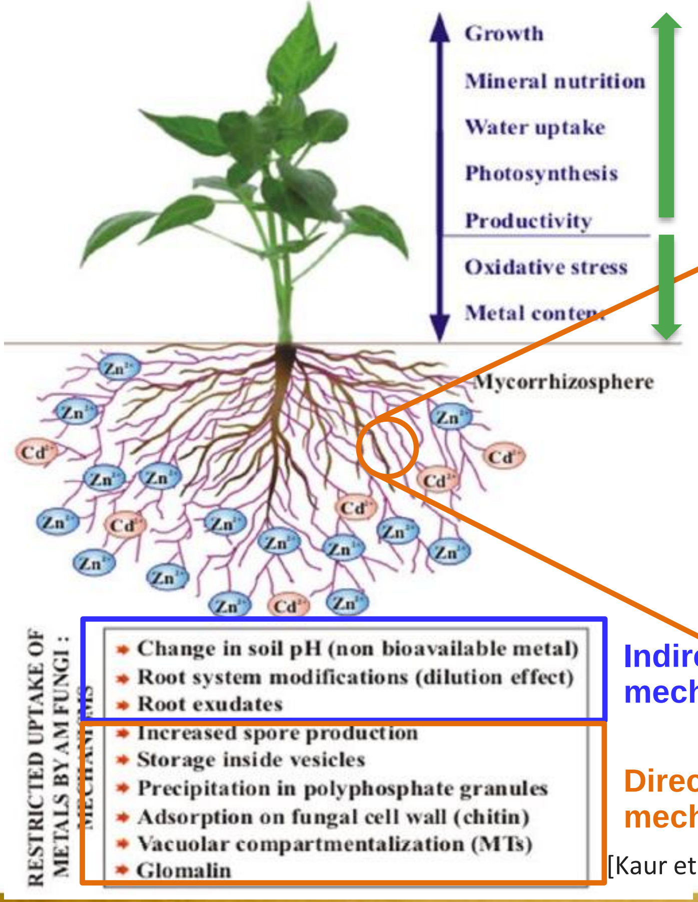
- **Nutrient Acquisition** ↑

https://greenbeanconnection.wordpress.com/2013/11/16/mycorrhizal-fungi-the-proof-is-in-the-roots/



Role of Soil Amendments: Potential of Mycorrhizal Fungi (MF)

➢ Plant Growth Enhancement and Metal Tolerance Mechanisms

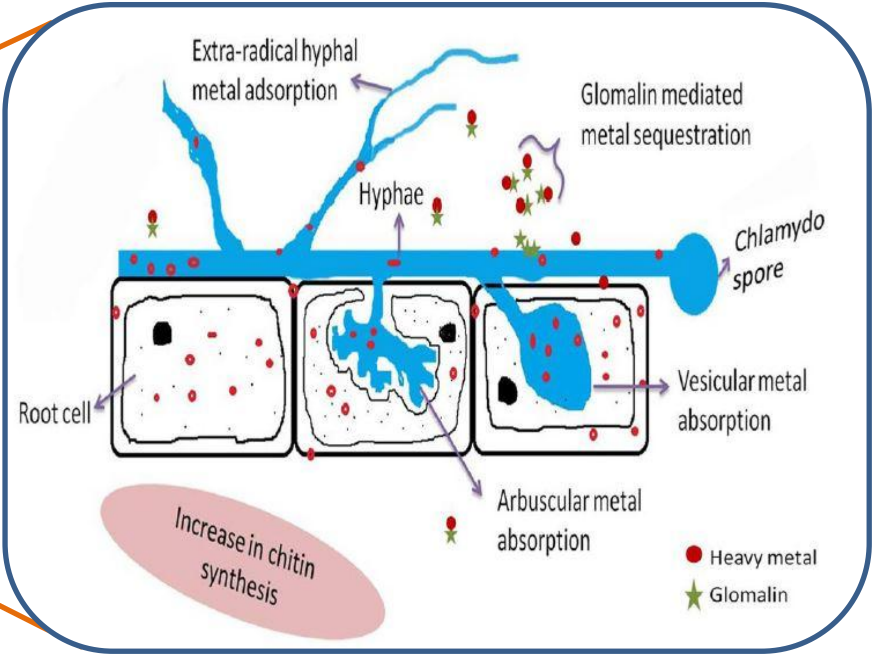


Indirect mechanisms

Direct mechanisms

[Kaur et al., 2018]

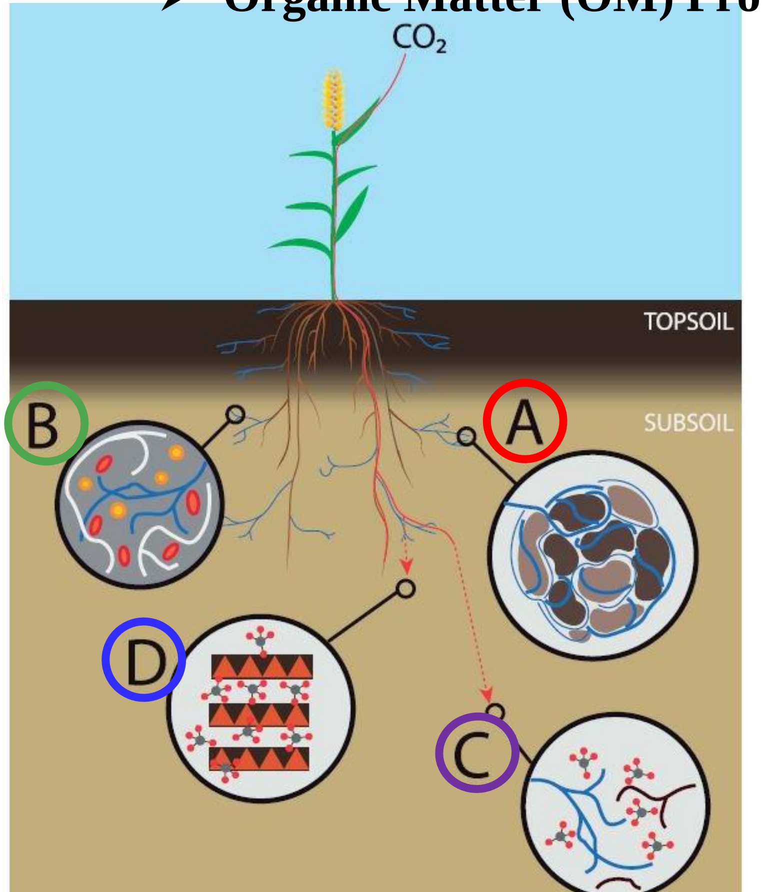
Direct metal tolerance mechanisms in MF



[Janeeshma et al., 2020]

Role of Soil Amendments: Potential of Mycorrhizal Fungi (MF)

➢ **Organic Matter (OM) Protection and C-Sequestration**



[Sosa-Hernández et al., 2019]

A: Aggregate-protected OM

B: Reduced decomposition rates by competing with bacteria and fungi

C: Increased C-input via mycelium exudates and turnover

D: Formation of highly stable mineral-associated OM fractions

➢ **In addition: Soil aggregate formation and stabilization**

Stimulate a Beneficial AMF Symbiosis in Extremely Degraded Tailings

- AMF highly sensitive to **P** which could be the case under **BS** application
- AMF can be sensitive to **metal toxicity**

Experimental Design:

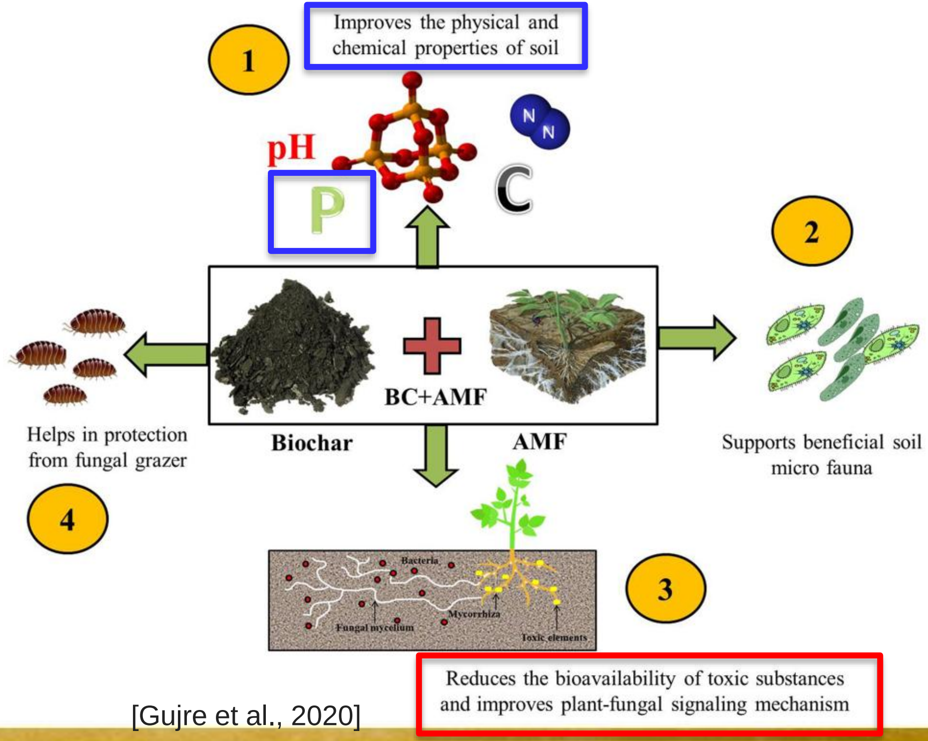
AMF

+

BS + BC

- **<u>BC may have multiplicative supporting benefits</u>**

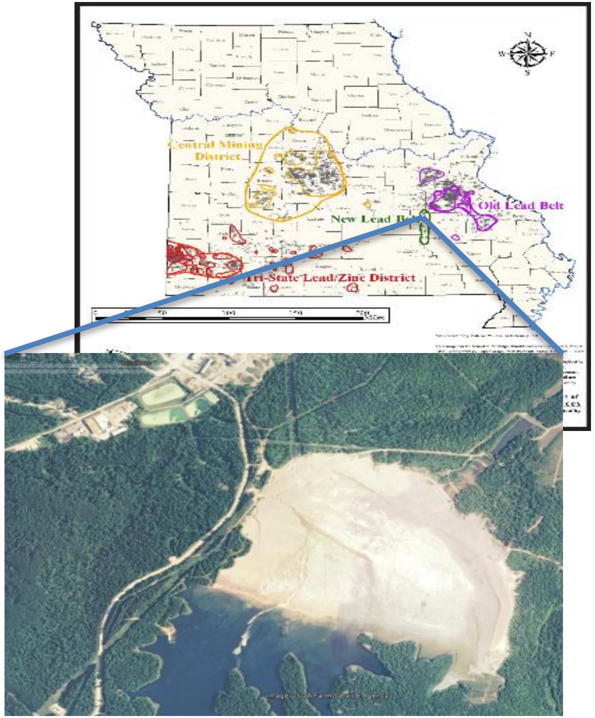
- Biochar facilitative effects on AMF abundance and functioning



[Gujre et al., 2020]

Site Description

Mine 28 impoundment
(lead and zinc mine)



"New Lead Belt" Viburnum MO, Iron County
Years active (1960-2004)

- **Physicochemical properties of tailings**

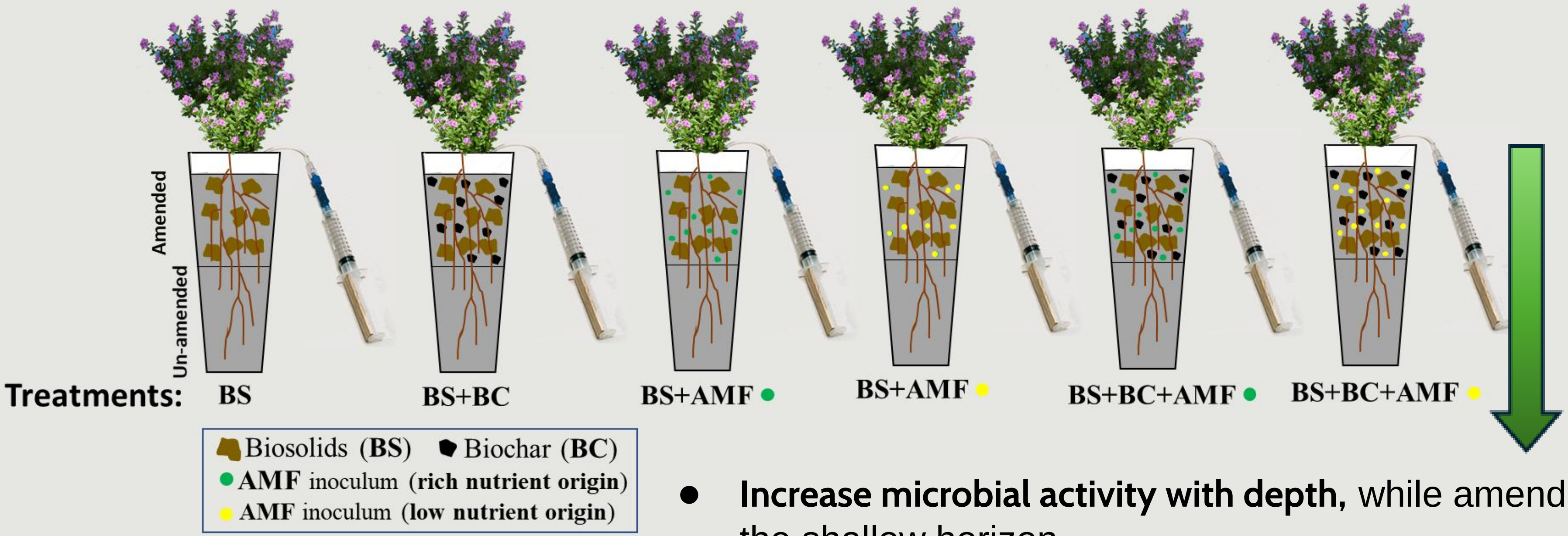
Sand %	Silt %	clay %
40	50	10

- High pH
- Lack of organic matter (**OM**)
- Lack of essential nutrients
- High **Pb**, **Zn**, and **Cu**

Properties	Tailings
pH	7.6
CEC meq/100g	3.6
Organic matter %	0.1
Nitrate (mg/kg)	0.5
Ammonium (mg/kg)	1.7
Olsen P $_{Available}$ (mg/kg)	2.8
K $_{Extractable}$ (mg/kg)	40
Ca $_{Extractable}$ (mg/kg)	659.5
Mg $_{Extractable}$ (mg/kg)	295
Zn $_{Extractable}$ (mg/kg)	99.9
Cu $_{Extractable}$ (mg/kg)	0.999
Pb (mg/kg)	3553
As (mg/kg)	52.3
Cd (mg/kg)	13.67
Co (mg/kg)	39.25
Cr (mg/kg)	11.49
Ni (mg/kg)	70.67
Mo (mg/kg)	2.536

Role of Arbuscular Mycorrhizae Fungi Symbiosis with Prairie Native Species:

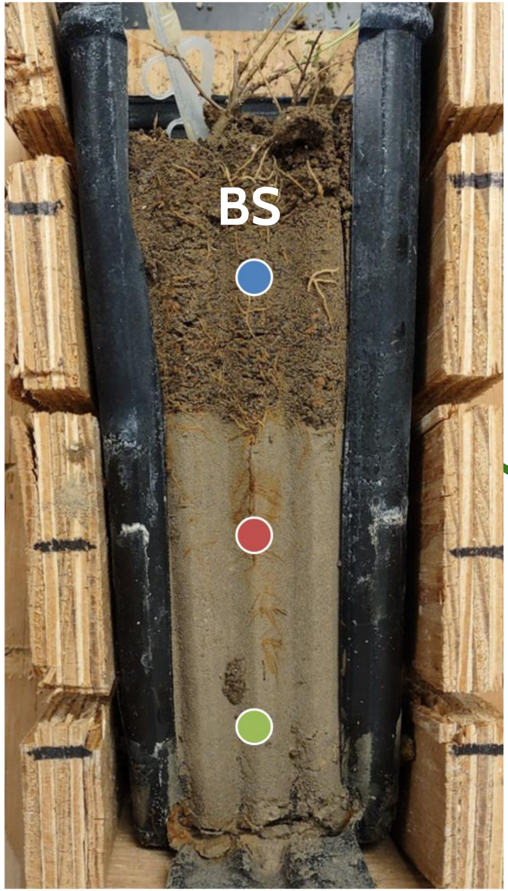
➢ **Long-term Experiment:** Goal - Build soil and increase soil function at depth



- **Increase microbial activity with depth,** while amending the shallow horizon
- Sustained and increasing fertility and ecosystem services

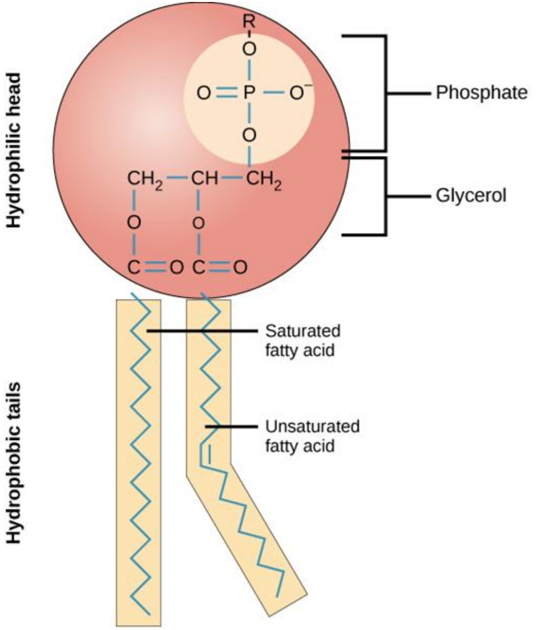
AMF Promoted Vigorous Roots Growth: 7-year Growth

- AMF promoted vigorous root growth compared to non-AMF
- AMF impact extended to deeper unamended tailings layers
- Soil cores were sampled at 3 depths for microbial and OC analysis



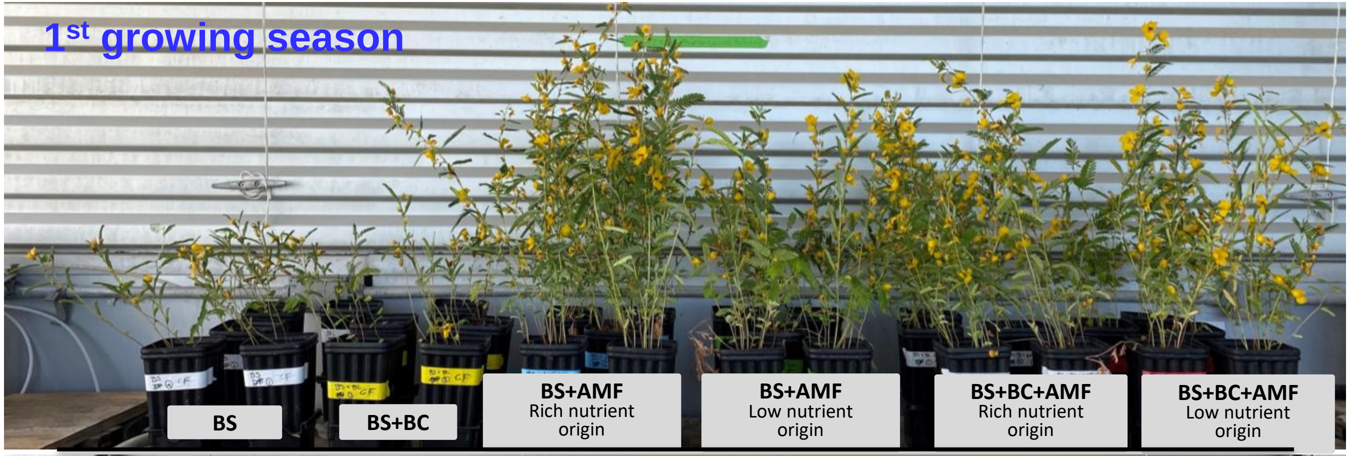
Methods: Phospholipid Fatty Acid Analysis (PLFA)

- Measure of microbial quantification and compositional analysis
- Closely linked to a functional view of soils
- Offers tangible and timely data[1]
- Performed by the MU Soil Health Assessment Center (SHAC)



Role of AMF: Plant Growth

C. fasciculata



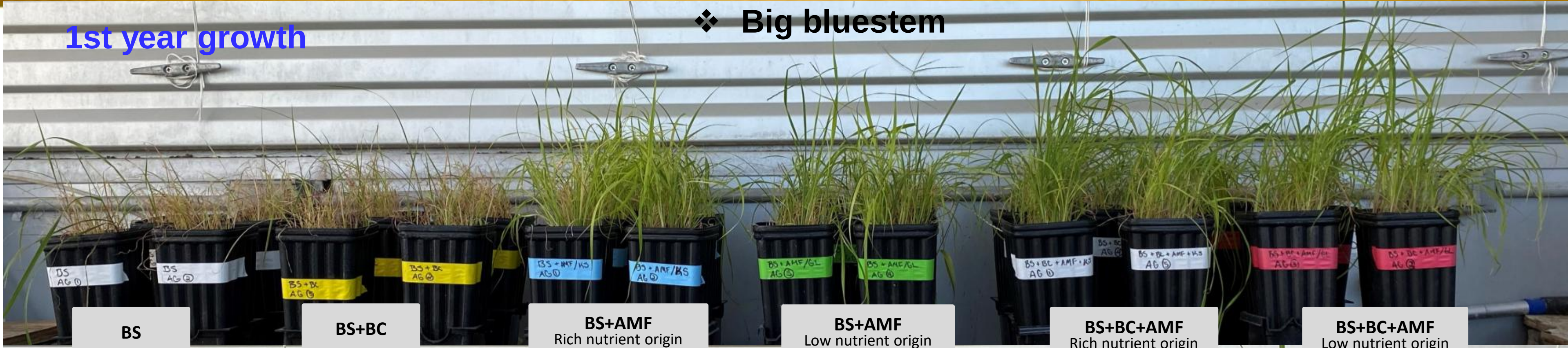
Role of AMF: Plant Growth

Amorpha fruticose

Role of AMF: Plant Growth

❖ **Purple prairie clover**

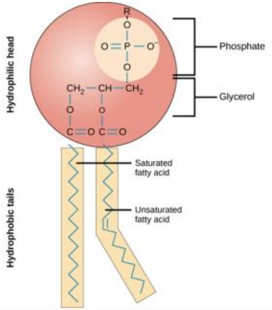
❖ Big bluestem



Role of AMF: Plant Growth

- Illinois bundleflower

Results: Roots act as significant microbial vectors



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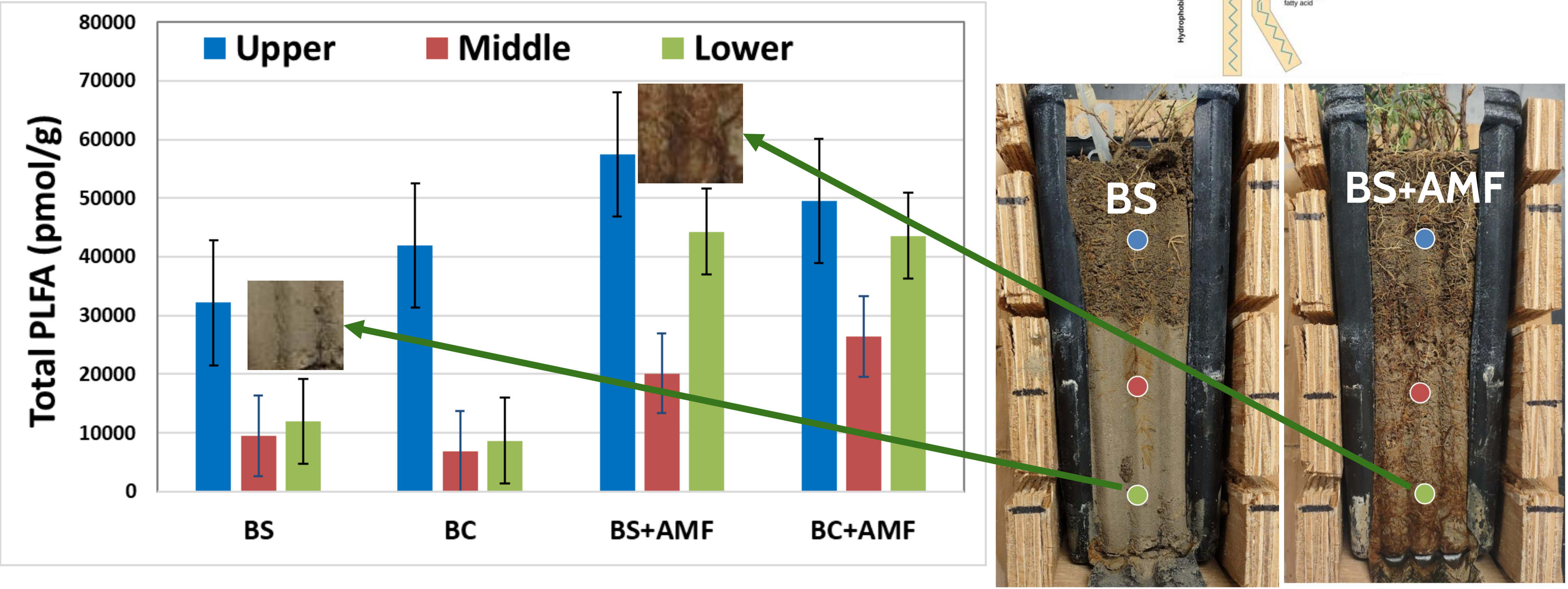
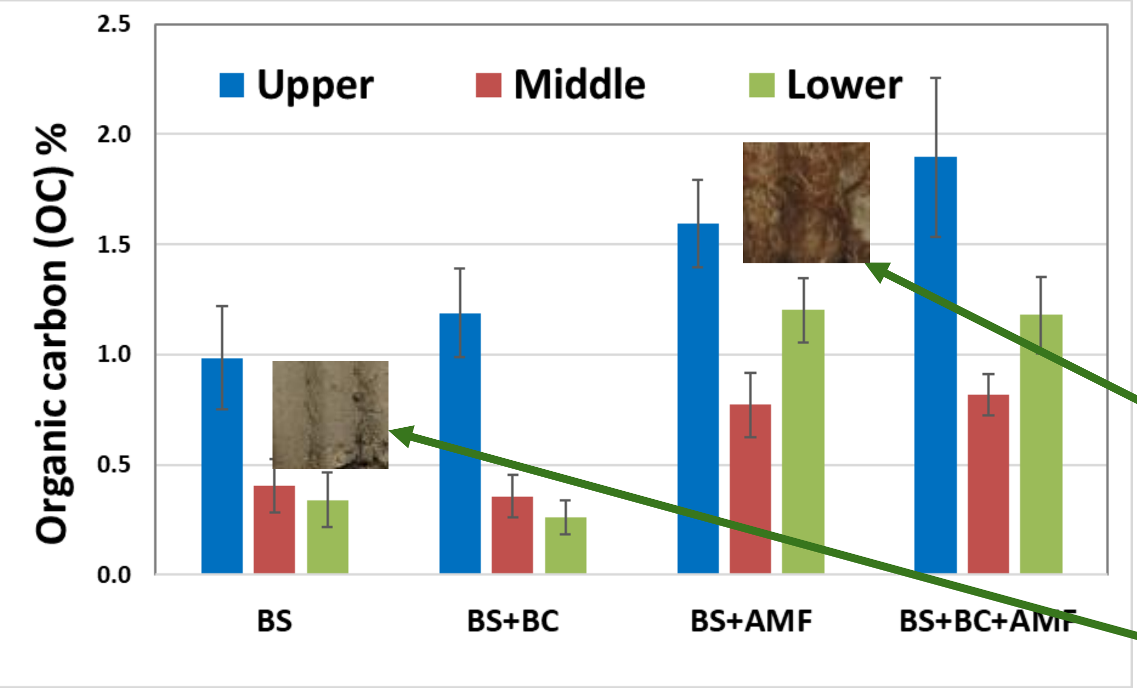
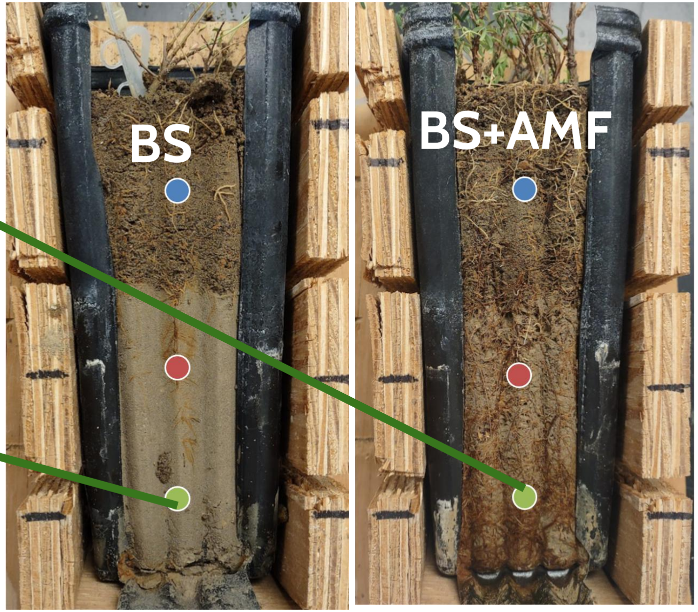


Organic Carbon Input (OC)

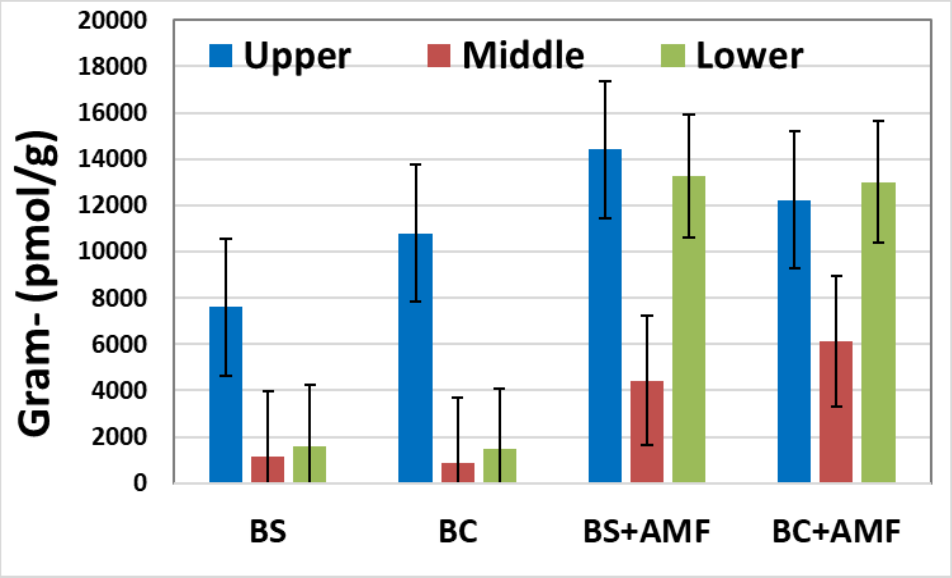


Root stimulated OC input into untreated lower tailings layer

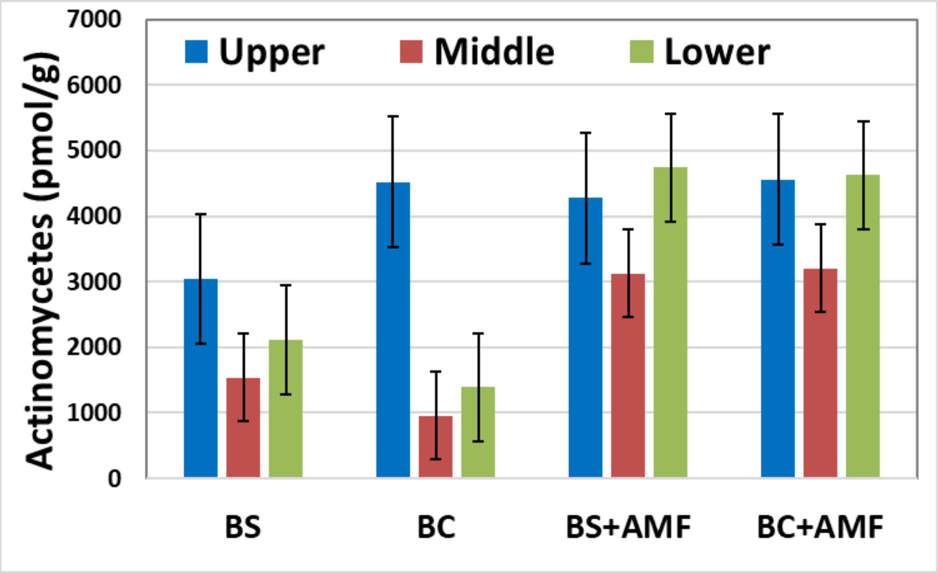


AMF critical to microbial establishment at depth, increasing activity and diversity at depth

Gram- bacteria

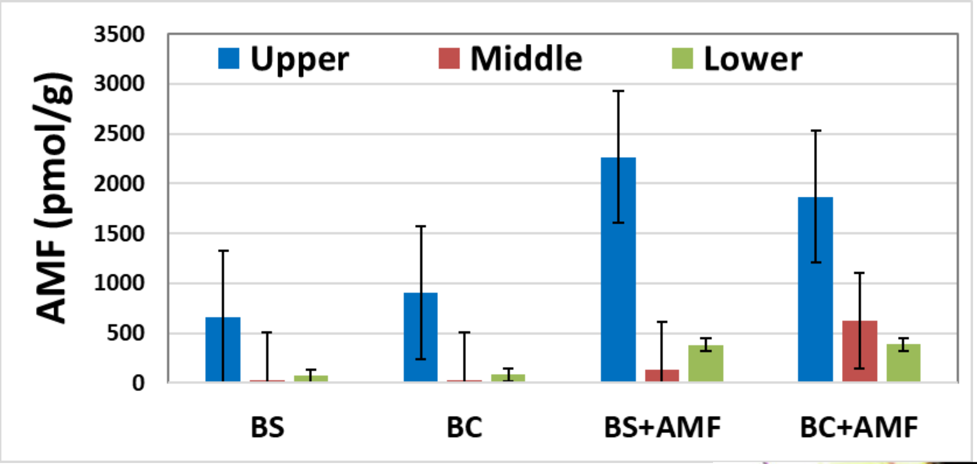


Actinomycetes



- AMF promoted beneficial microorganisms linked to organic matter decomposition and nutrient cycling in the untreated lower tailings layer

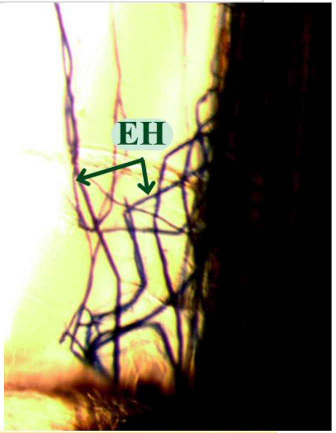
AMF proliferates itself at depth, suggesting continued benefits

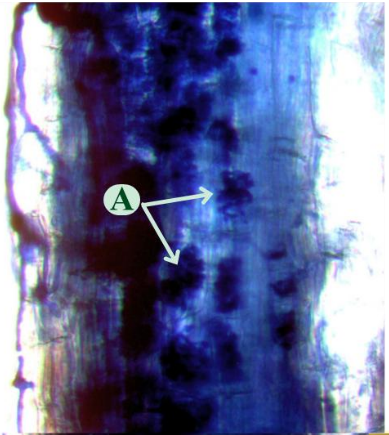


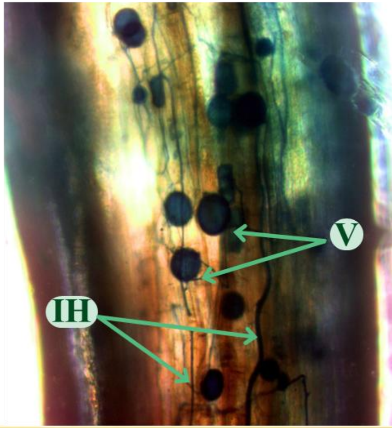
AMF structures include:

- Intraradical hyphae (IH)
- Extraradical hyphae (EH)
- Vesicles (V)
- Arbuscules (A)

➢ AMF structures within plant root cells and its extended hyphae





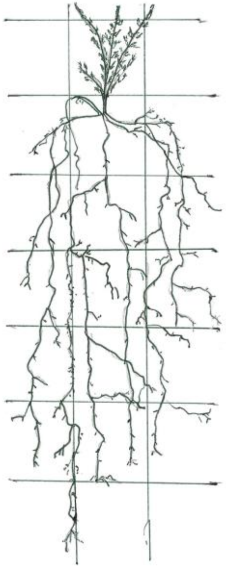


Expected benefits different in open-field studies

- Pot study influenced root development
  - Root grouping at lower layers
- Full depth development of *D. purpurea*
  - Mature depth of 2.0m
  - Some prairie species could reach 4.5 m rooting depth







Plans of future study

- Further PLFA studies
  - Additional plant species
  - Additional AMF cultures
- Physicochemical correlations
- Comparison to 16S rRNA data
- Implications for soil building

Acknowledgements

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Undergraduates: Bennett Scott, Emma Kettler

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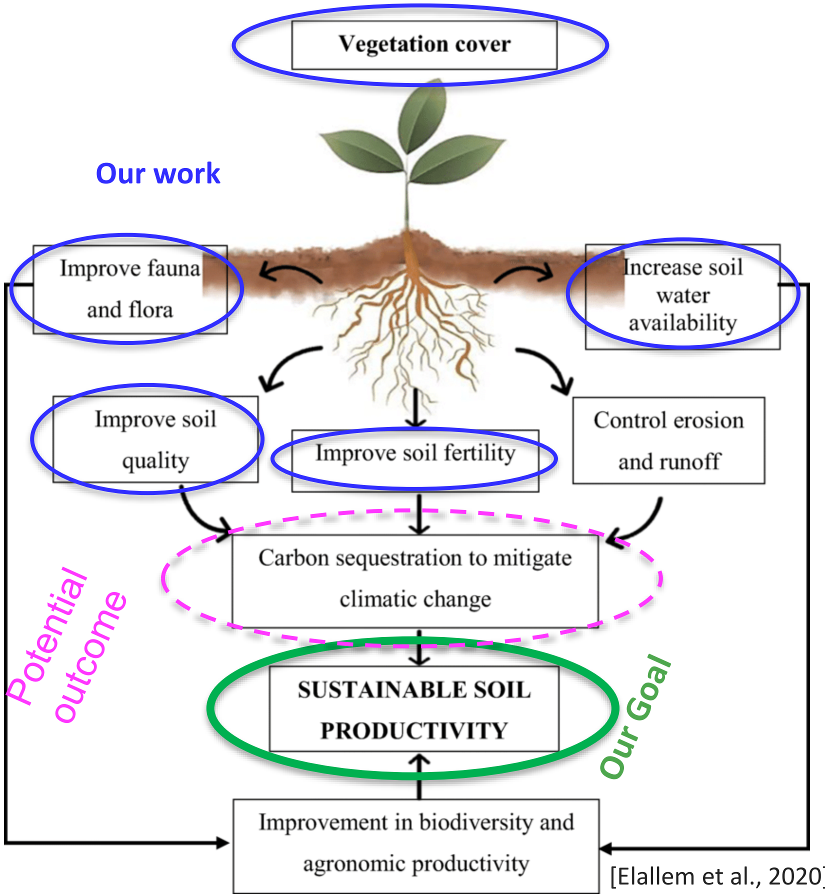
Thank you

Questions??

Revegetation Potential for Mine Tailings Ecorestoration:

- **Tailings revegetation:**
  - Cost effective
  - Long-term phytostabilization:
    - Wind, water, chemical immobilization
  - Pollution control
  - Threats removal to public health
  - Ecological revitalization

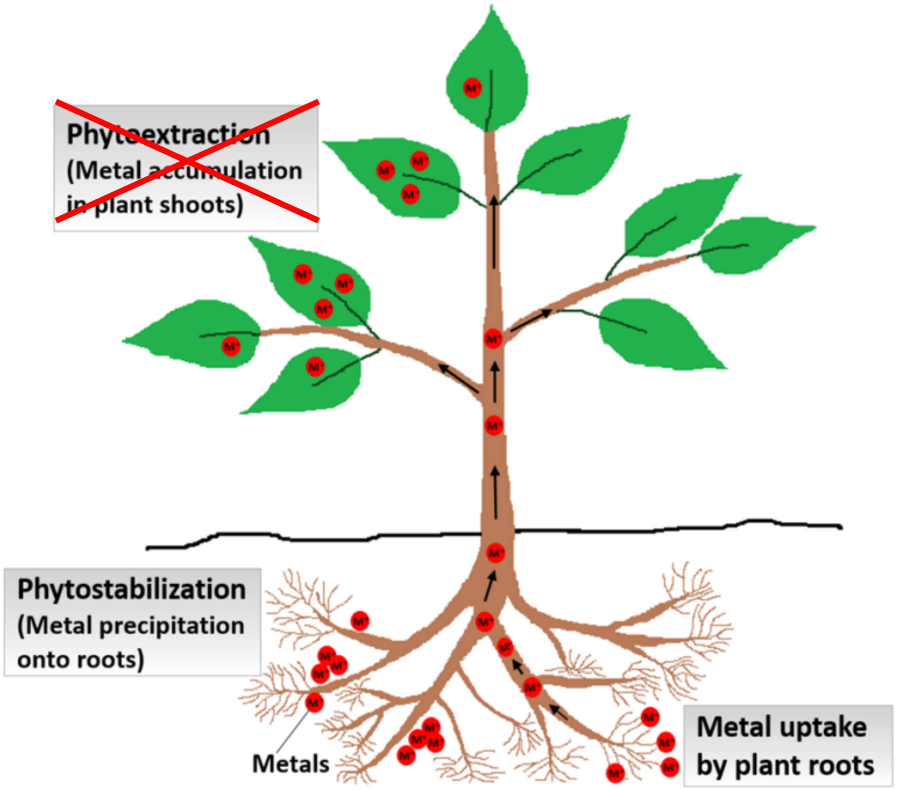


[Elallem et al., 2020]

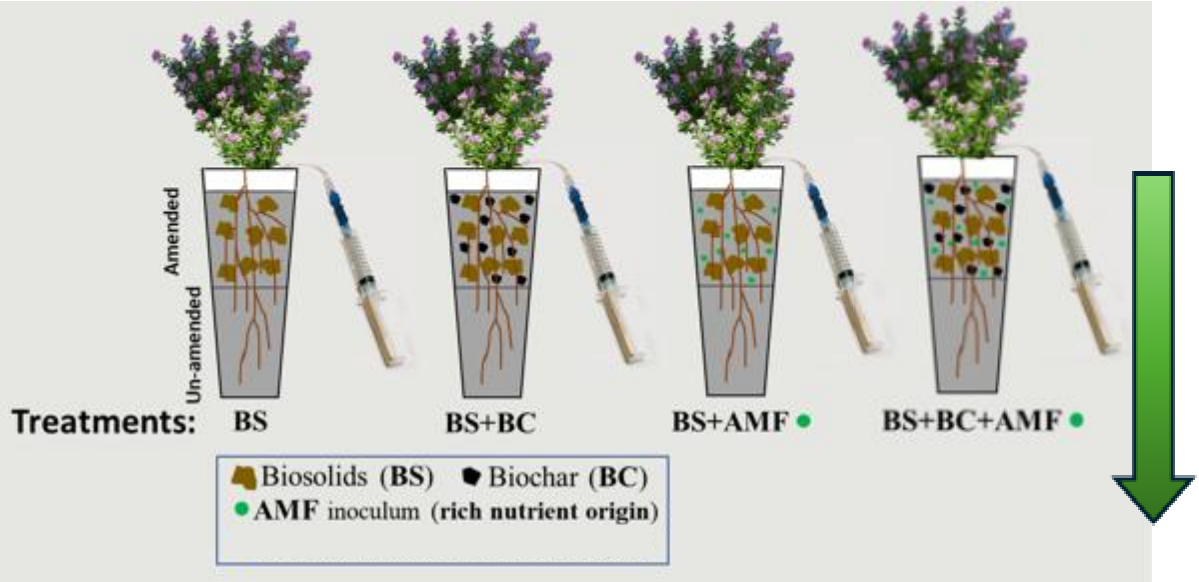
Two Main Phytoremediation Approaches:

Long-term Phytostabilization

- Vegetation cover
- Physical/chemical immobilization
- Dust/leaching control
- Land reclamation and concurrent restoration of ecosystem services



Methods: Amended phytostabilization pot experiment



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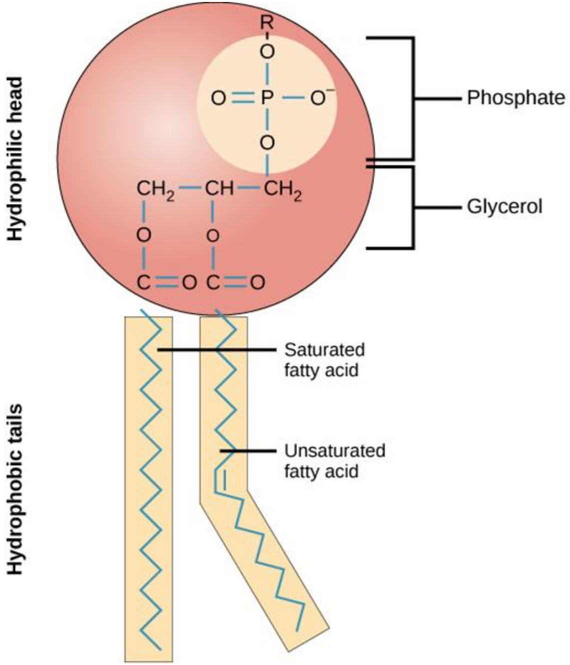
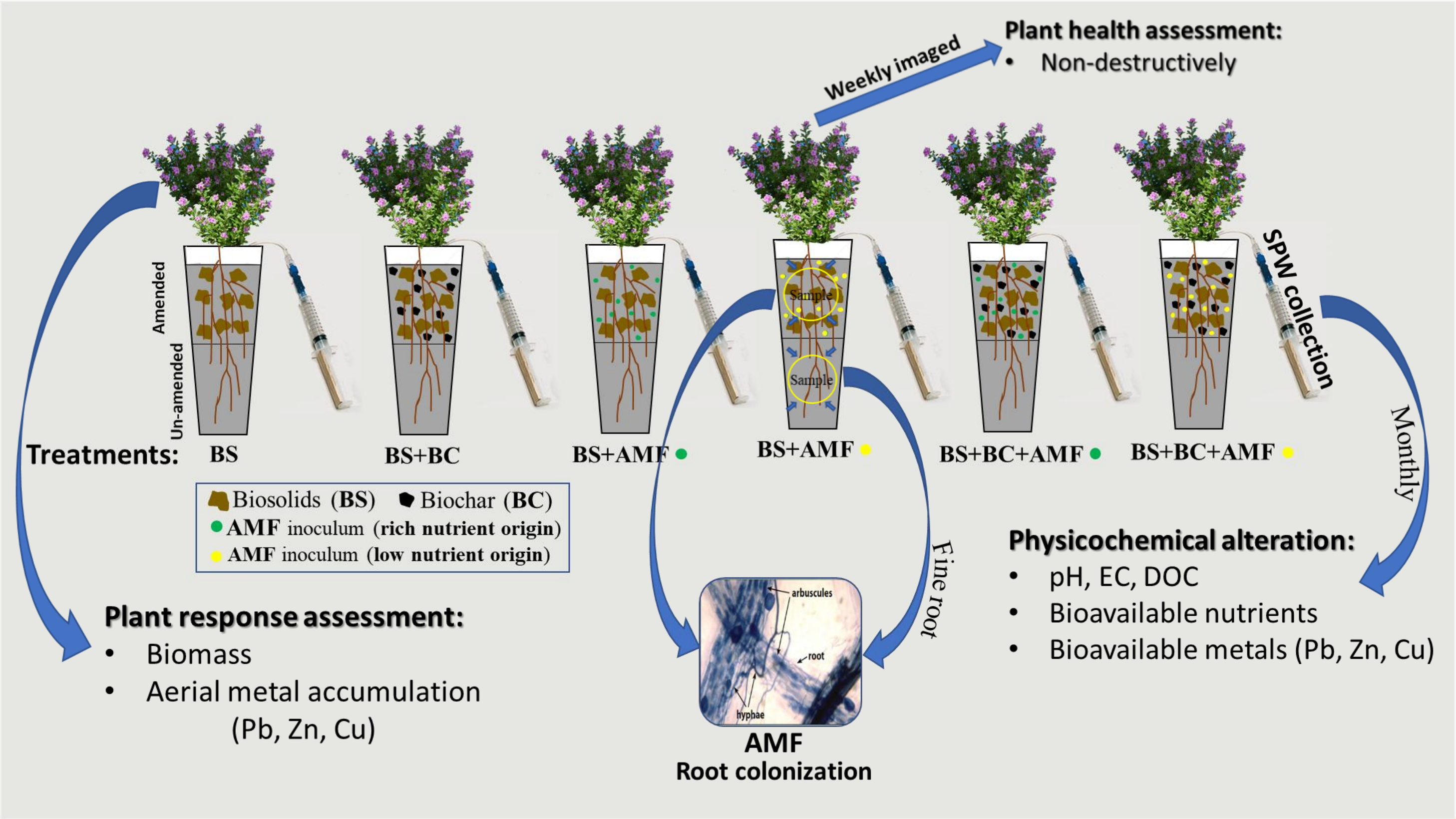


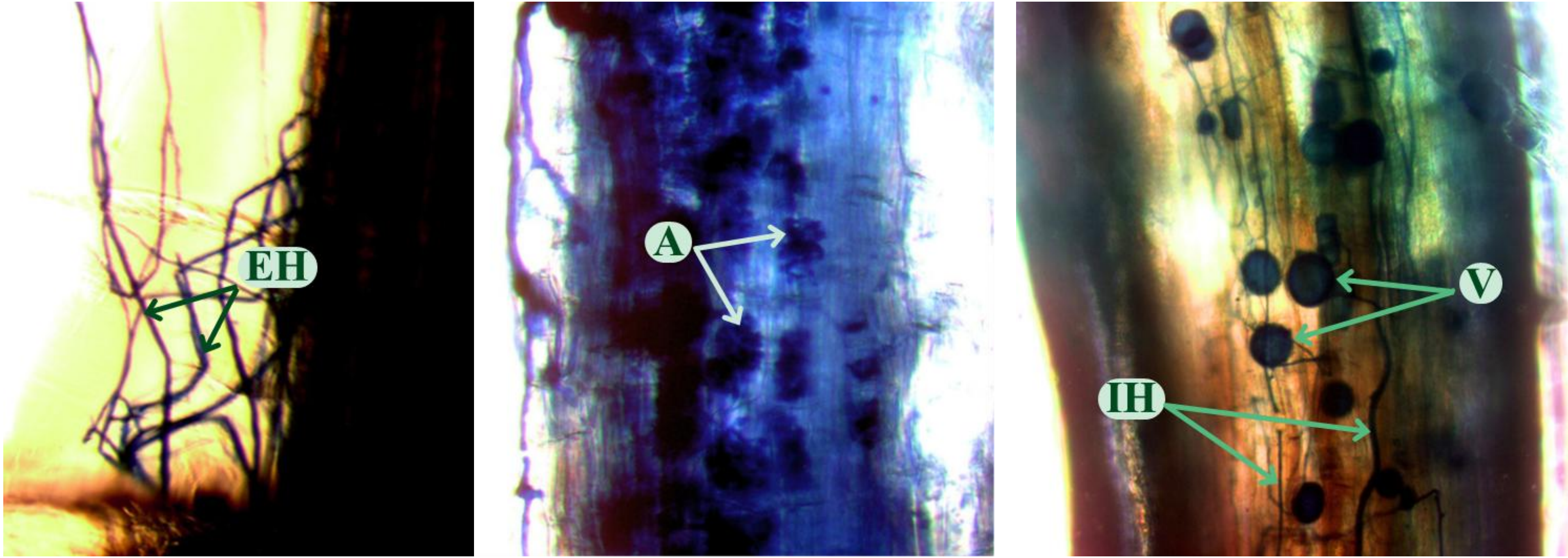
Role of Arbuscular Mycorrhizae Fungi Symbiosis with Prairie Native Species:

➢ Experimental Design

❖ **5 Native prairie species: 1 grass and 4 legumes**



Arbuscular mycorrhizal fungi (AMF) colonizing plant root



Extraradical hyphae (EH), Arbuscules (A), Intraradical hyphae (IH), and Vesicles (V)