Interactions of Biochar with Establishment of Legume-Rhizobia Symbiosis

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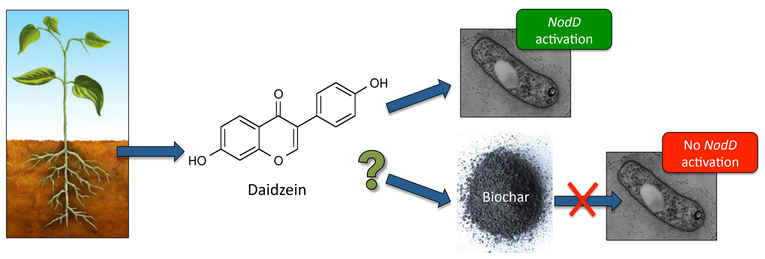
Introduction

- Legume roots exude aromatic flavonoid signaling compounds that induce *nod* genes in *Rhizobium* spp.
- *Nod* gene activation initiates rhizobial infection of legume root hairs causing nodulation and establishment of symbiosis.

Could biochar interfere with this process?

- Biochar can interfere with allelopathic flavonoids.[1,2]
- Biochar can disrupt signaling between bacterial colonies.[3]
- Activated carbon can decrease rhizobial nodulation and mycorrhizal colonization.[4,5,6]
- Biochar is used for remediation of PAH contaminants, which have similar log K_{OW} values to flavonoids (~2.5 – 4).[1,7]
- Biochar can decrease the efficacy of aromatic herbicides by 4 times.[1]

We hypothesize that biochar can disrupt signaling by sorbing flavonoids, reducing establishment of legume-rhizobia symbiosis.



Objectives

- To determine if amendment of walnut shell (WS) biochar to soils will reduce nodulation of legumes by rhizobia.
- To assess whether biochar will affect symbiosis establishment differently based on the texture of the soil.

Materials and Methods

Materials:

- Walnut shell biochar—produced at 900°C, pH of 9.7, SA of 221.7 $m^2\ g^{-1}$
- Yolo silt loam soil—14% clay, 51% silt, 35% sand
- Sterile sand

Experimental Setup:

- Cowpeas (*Vigna unguiculata* L.) grown in a greenhouse experiment; seed inoculated with pea rhizobia inoculum.
- RCBD with 5 blocks, 1 treatment replicate per block
- 5 x 2 factorial treatments:

Silt loam soil	Control - No amendment
	10 g kg^{-1} WS biochar
	25 g kg^{-1} WS biochar
Sand	Lime equiv. for 10 g kg^{-1} biochar*
	Lime equiv. for 25 g kg^{-1} biochar*

*determined by Sikora soil buffer capacity method and biochar liming potential

- Pots kept at a moisture content of 60% WHC
- Watered daily—soil with DI H_2O only, sand with DI H_2O and with Hoagland solution 2x per week

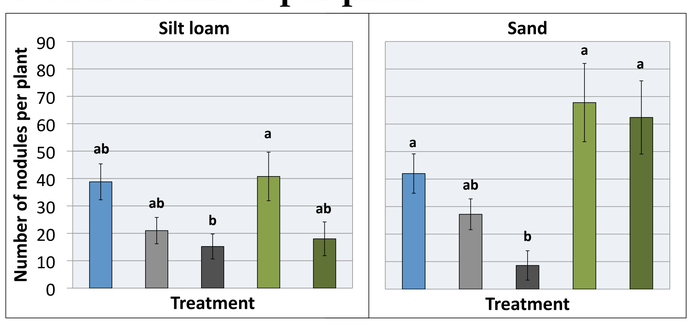
Analyses:

- Plants harvested 33 days after seeding
- Plant health assessments and soil samples collected at harvest
- Roots washed and nodules per plant counted
- Nodules removed; nodule, shoot, and root dry weights determined

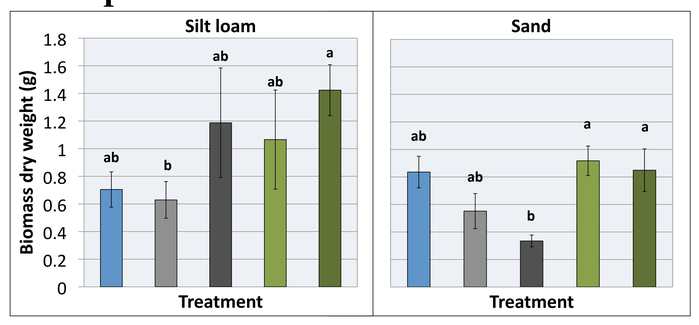
Results

There were significant soil-treatment interactions for nodule number (p=0.02) and total plant biomass (p=0.03). Therefore, treatment effects were analyzed separately for each soil.

Nodule number per plant



Total plant biomass



Figures 1-4. Bars represent the mean ± SE (n = 5, except for silt loam lime 10 g kg^{-1} equivalent and sand control where n=4). Treatments not sharing a letter are significantly different (Tukey Means Comparison tests, $p < 0.05$).

Change in pH over growth period

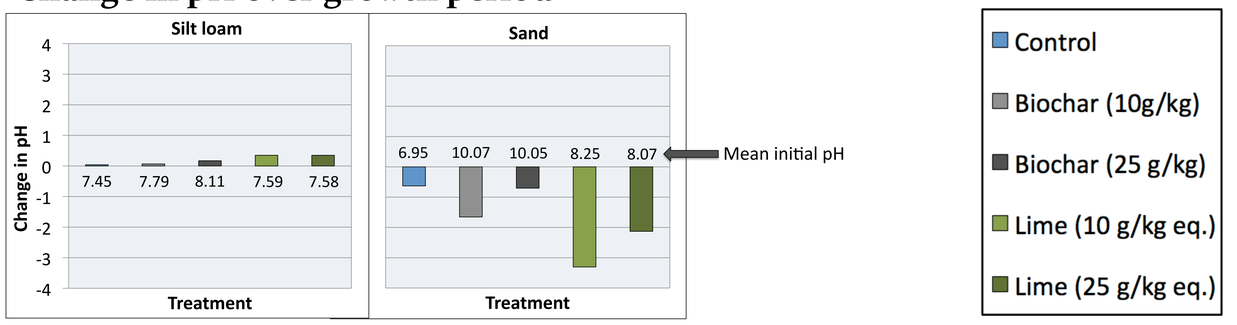


Figure. 5. Final pH – Initial pH. Initial pH values based on incubations (n=3). Final pH values determined from samples collected at harvest (n=5).

Soil texture and biochar effects

Paired t-tests showed non-significant effects of soil texture on nodule number at both the 10 g kg^{-1} biochar rate (p= 0.4251) and 25 g kg^{-1} biochar rate (p= 0.3747).

Conclusions

- Biochar significantly reduced nodulation when applied to the sand at a rate of 25 g kg^{-1}.
- Nodule numbers were reduced in 10 g kg^{-1} biochar treatments in both soils and in the 25 g kg^{-1} in silt loam soil, but these results were not significant.
- The texture of the soil medium did not impact the effects of biochar at either rate.
- pH decreased over time in the highest nodulating treatments.

Next Steps

- Batch sorption experiments using synthetic daidzein and genistein, flavonoids exuded by cowpea, to determine the relative affinities for soils and biochar for these compounds.
- Repeat of greenhouse experiment with pH monitoring throughout growing period.

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