Combining Nitrogen and Rhizobia to Improve Nursery Growth of Nitrogen-fixing Plants

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Introduction – Nitrogen and Plant Growth

- Nitrogen usually the most critical element for plant growth.
- Present in soil in organic and inorganic forms.
- Ammonium and nitrate predominant inorganic forms.
- Organic forms (amino acids) can be primary N source.

Introduction – Nitrogen and Plant Growth

- N form can affect plant growth.
- Much work with conifers, at least with inorganic forms.
- Organic forms with northern latitude conifers and others.
- Sometimes better growth; sometimes not.
- Few studies with organic forms and broadleaves and forbs.
- Organics having lower leaching potential.

Introduction – Nitrogen Runoff

Introduction – Nitrogen, Plant Growth, & Rhizobia

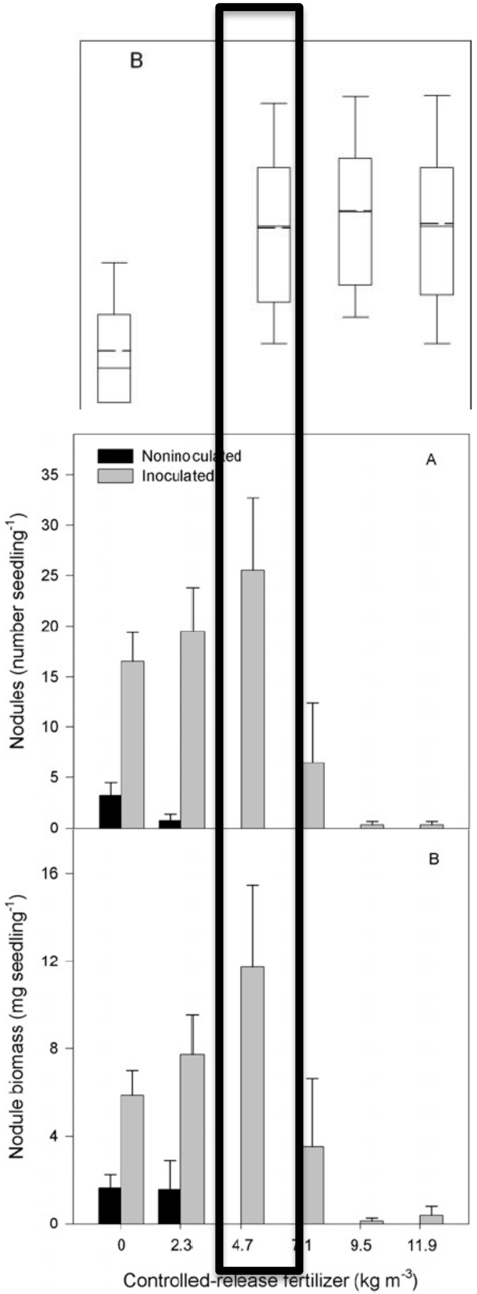
- Rhizobia bacteria colonize some plant roots, produce nodules, and convert atmospheric N into ammonia in exchange for plant carbohydrates.
- Harsh site implications.
- Conventional wisdom: applications of high N suppress nodule development. Low N can improve nodulation.
- Data with *Acacia koa* indicates good nodule development despite high N applications.

Previous studies framed the questions

Height

Nodule number

Nodule biomass



Dumroese et al. (2011a)

Dumroese et al. (2009)

Potential for Using Biochar in Container Media Used to Grow Native Plants

Converting forest biomass to bioenergy creates biochar waste, which might be used to help grow native plants for restoration.

Nursery costs may be reduced, plant quality improved, and carbon sequestered below ground at minimal cost. Variety of species.

Dumroese et al. (2011b; 2018)

Introduction – Biochar

- Quality / characteristics vary by feed stock and process.
- Conversion of organic matter in the absence of oxygen.
  - Pyrolysis – 300 to 700 ^{o}C, less energy, more biochar, no ash.
  - Gasification – 700 ^{o}C, more energy, less biochar, some ash.
- Gasification may have improved cation exchange capacity.
- Nearly all plant response research done with pyrolysis.

Methods – Two Studies

N Source

- Black locust and lupine
- 5 nitrogen treatments
  - Organic vs inorganic
- 2 and 1 mg N per week
- 75% container capacity
- Morphological assessment
- ^{15}N assessment
- Commercial inoculants

Biochar Source

- Black locust
- 3 biochar treatments
  - Pyrolysis vs. gasification
- 2 mg N per week
- 75% container capacity
- Morphological assessment
- ^{15}N assessment
- Commercial inoculants

Methods – ^{15}N Theory

- Most N is ^{14}N
- "Tag" fertilizer by adding minute ^{15}N
- Determine ^{15}N:^{14}N for plant only using fertilizer
- If plant has active rhizobia that add ^{14}N to the system from the atmosphere, ratio will be lower
- Expressed as $\delta\ ^{15}N$
- Higher $\delta\ ^{15}N$ means less N supplied by rhizobia

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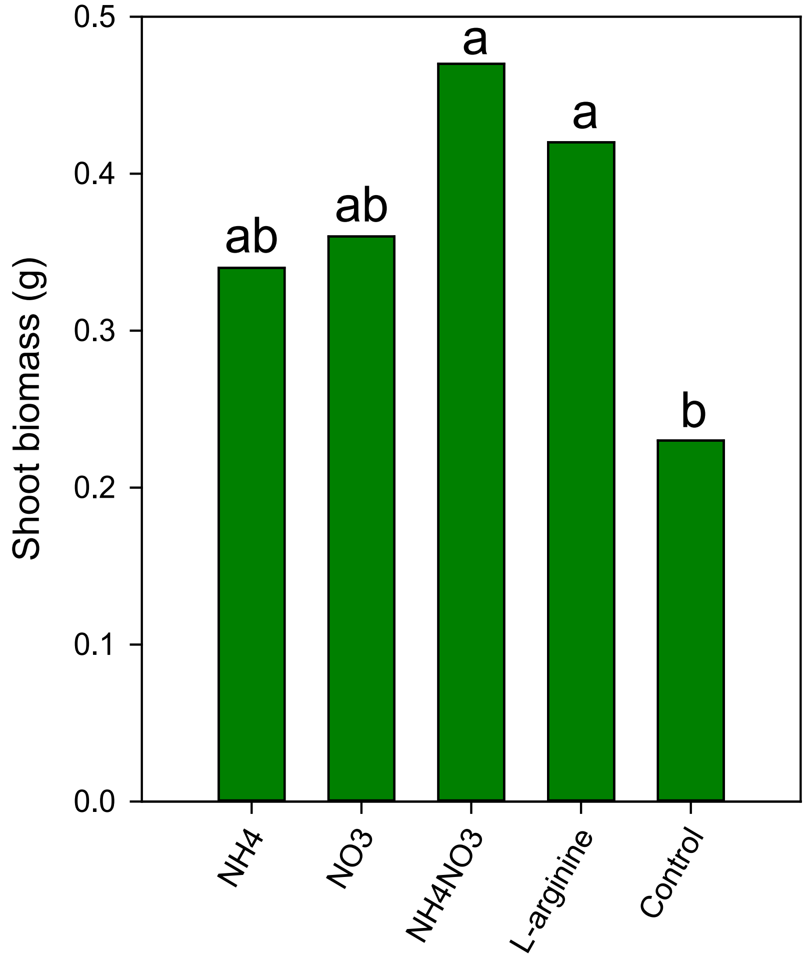
Results – Lupine

- N form and inoculation significant
- N fertilization increased morphological traits
- Inoculation improved growth



NH_4NO_3 – Without inoculation

NH_4NO_3 – With inoculation

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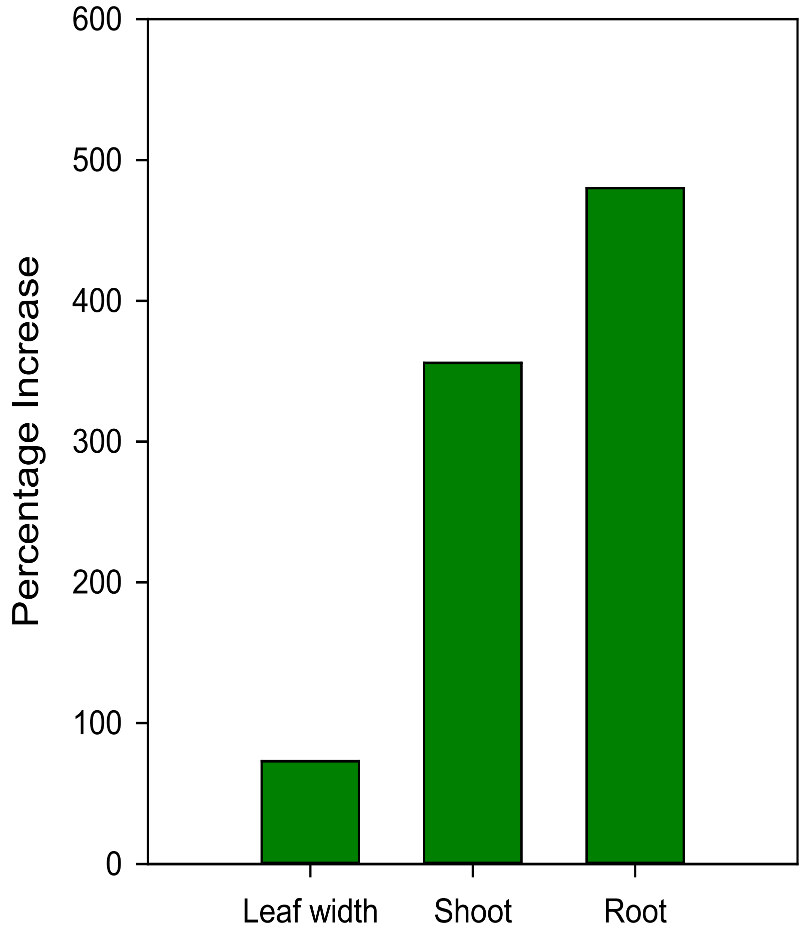
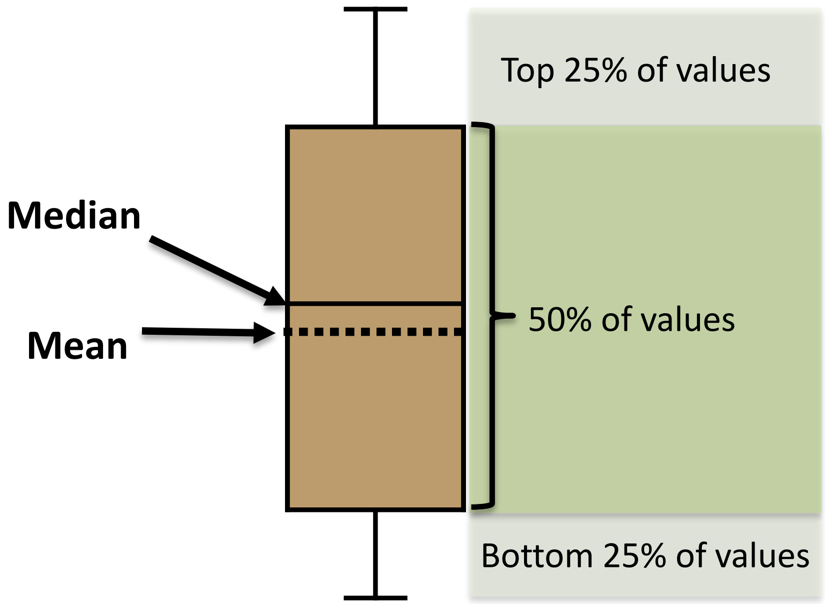


Control – No inoculation, no N

Control – Inoculation, no N

Results – Box Plots?

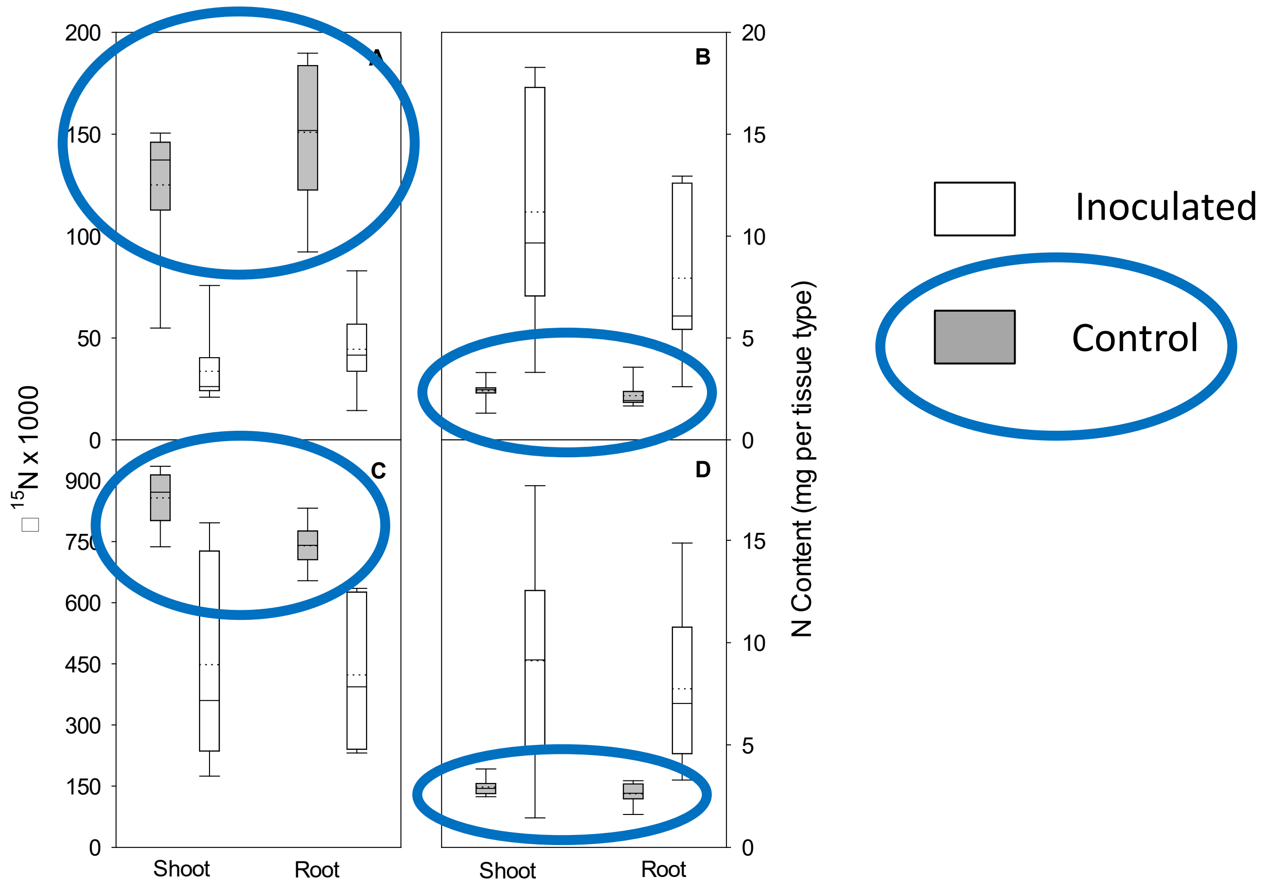


Results – Interpretation

Results – Lupine – N Form

Arginine (organic)

NH_4NO_3 (inorganic)



Zhang et al. (2019)

Results – Black Locust

- N form, inoculation, and interaction significant
  - *Exceptions: Shoot-to-root ratio unaffected; *RCD and nodule biomass unaffected by interaction*
- Inoculation + N fertilization yielded biggest seedlings
- N fertilization improved growth; no differences among N forms
  - *Exception: Nodule biomass affected by N form*
- Inoculation improved growth

Variables

- Height
- RCD*
- Leaf biomass
- Stem biomass
- Shoot biomass
- Root biomass
- Nodule biomass*
- S:R

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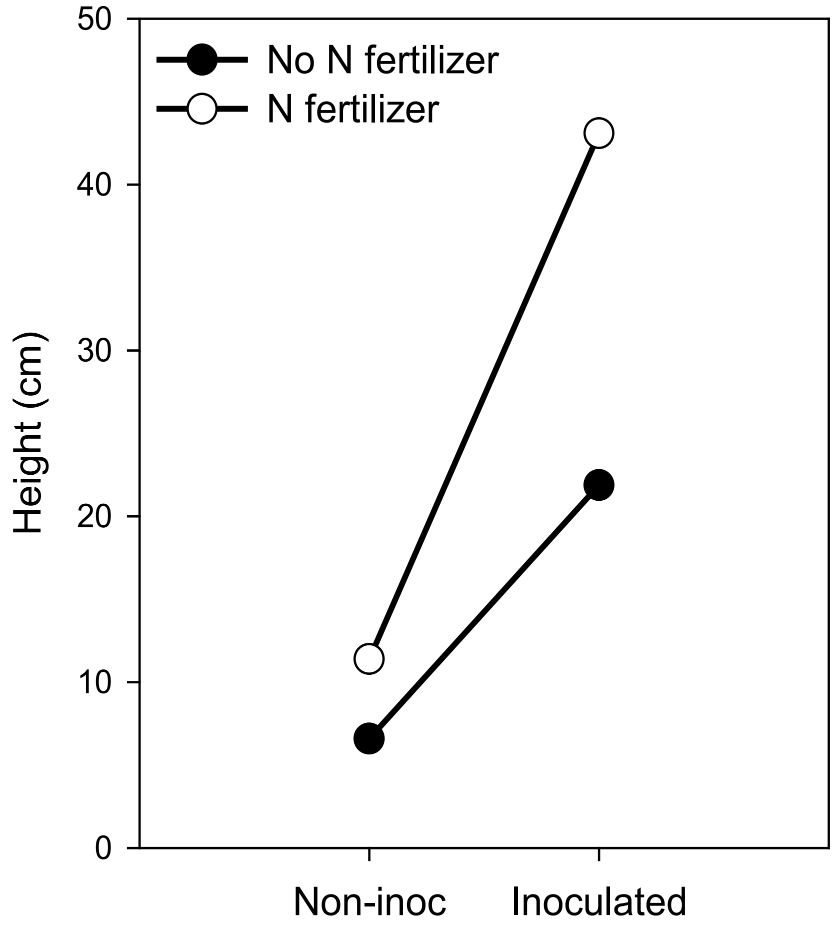

Controls

Left: inoculation
Right: no inoculation

Ammonium sulfate

Left: inoculation + N
Right: ammonium sulfate

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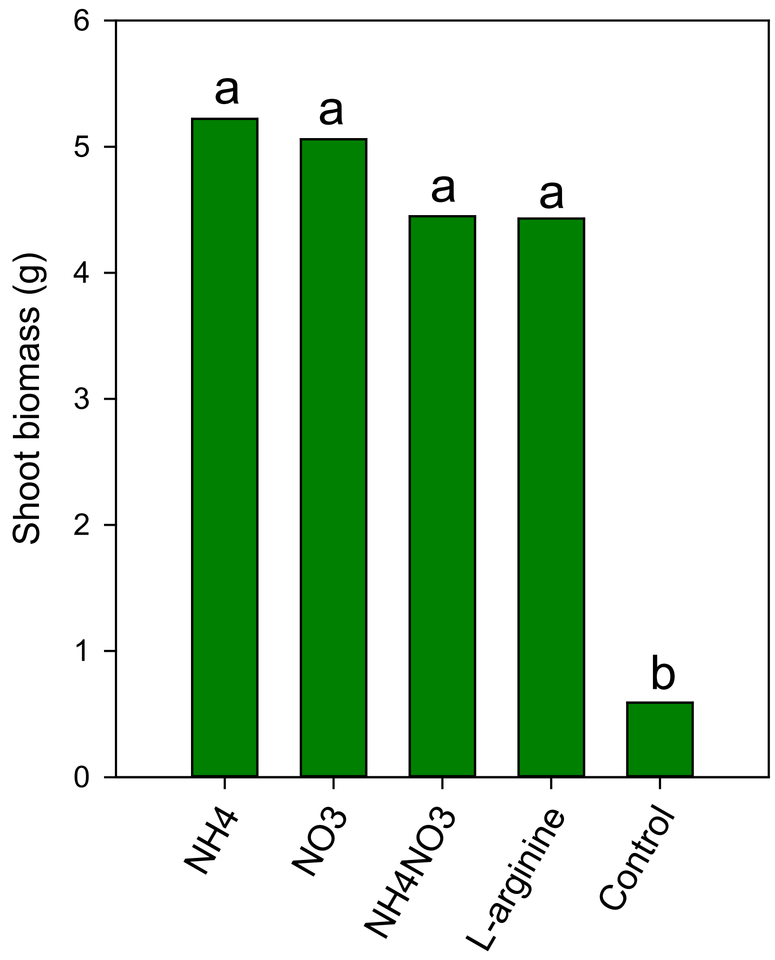

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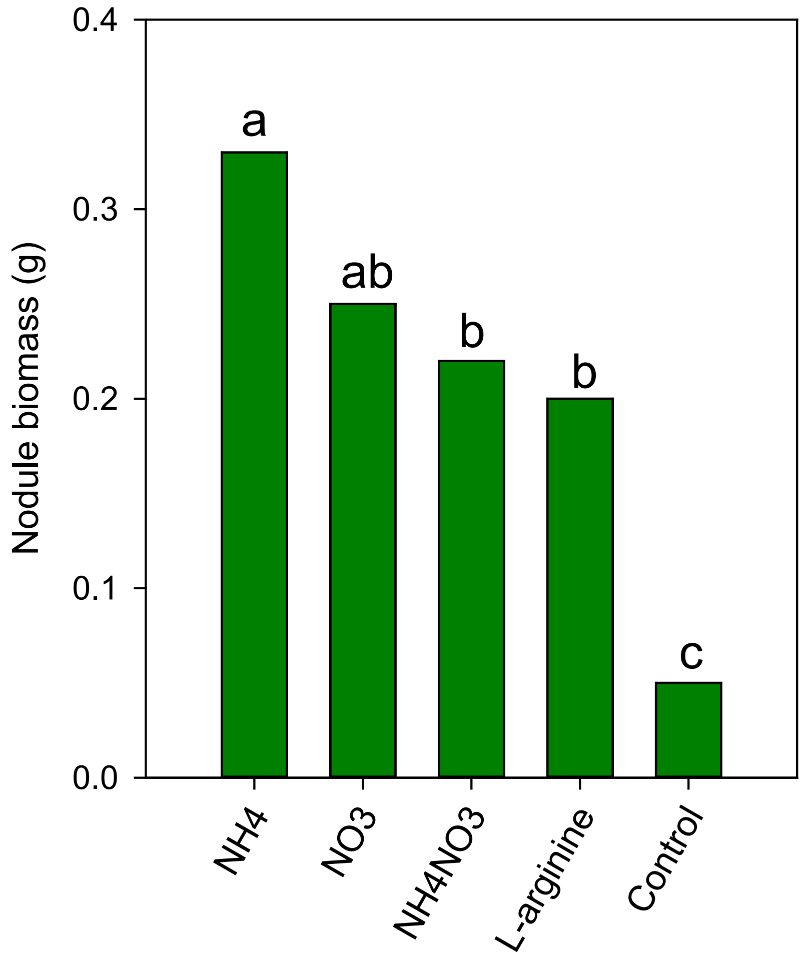

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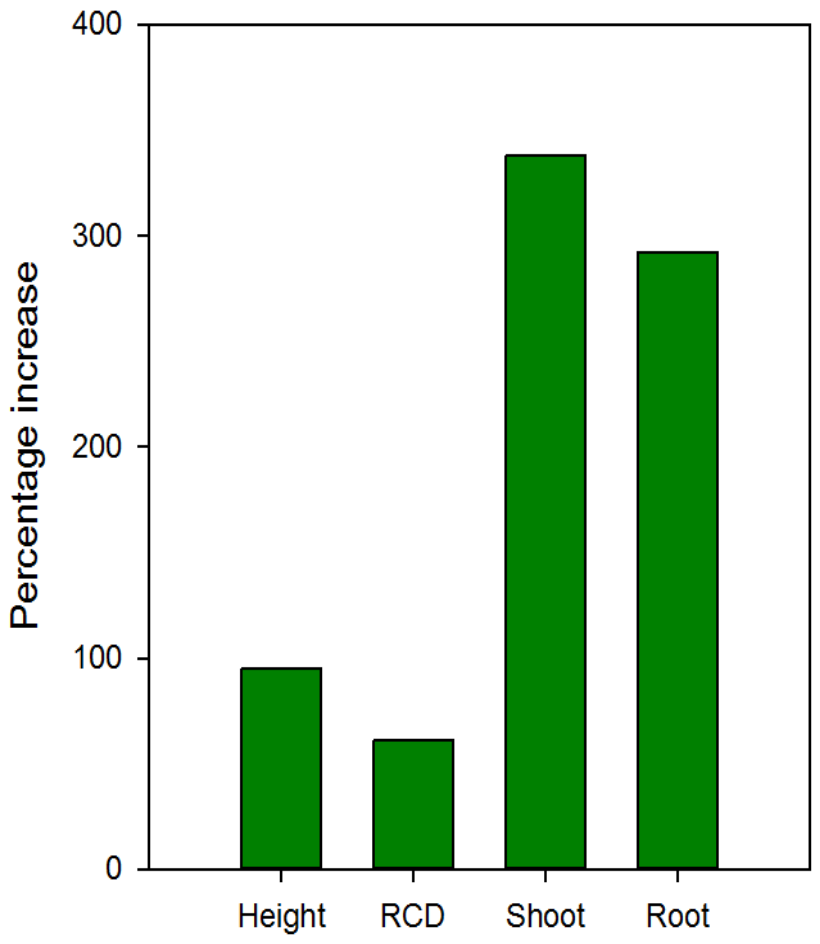


N fertilizer

(far left is the no N fertilizer control)

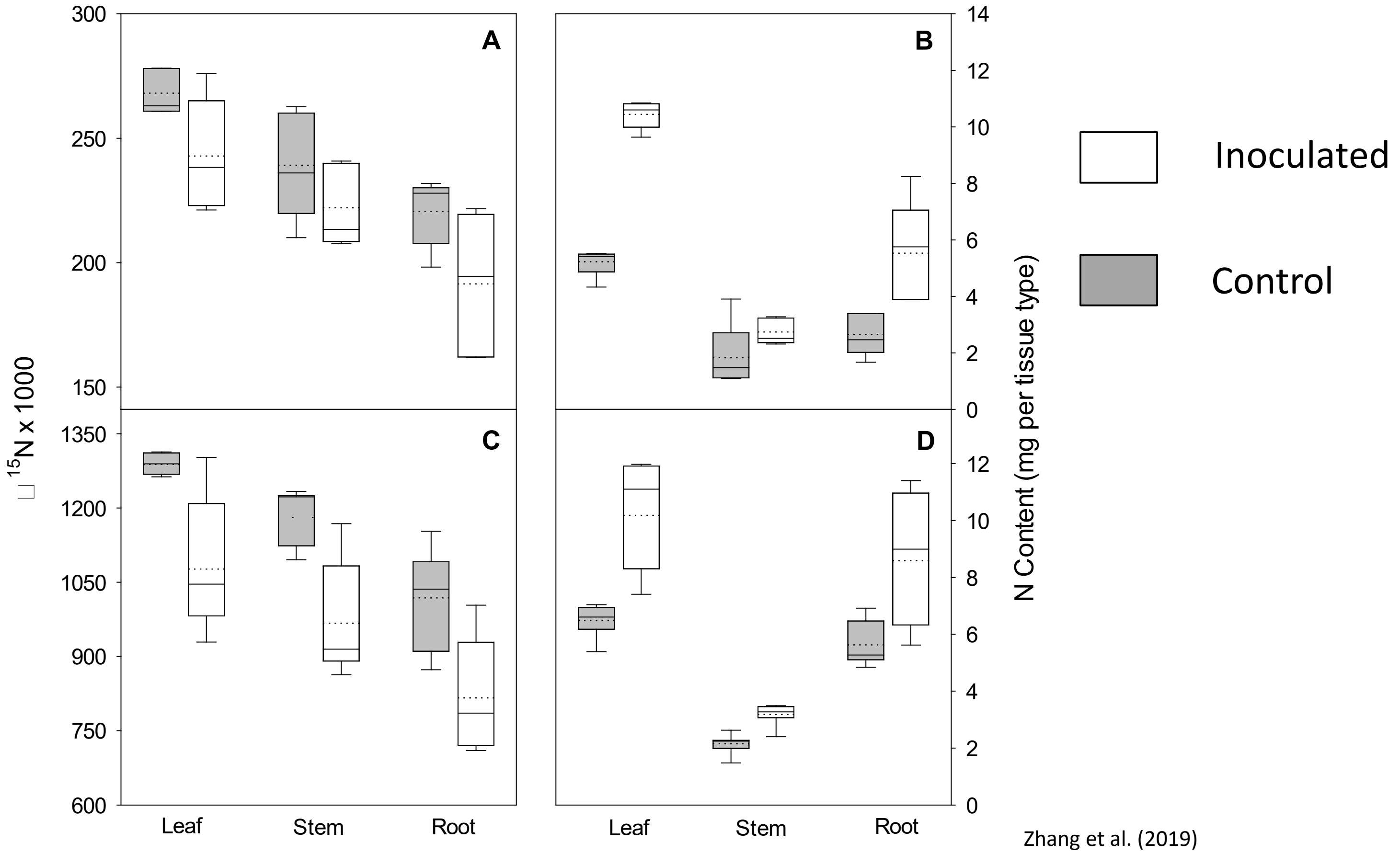
Inoculated + N fertilizer

(far left is the no N fertilizer control)

Results – Black Locust – N Form

Arginine (organic)

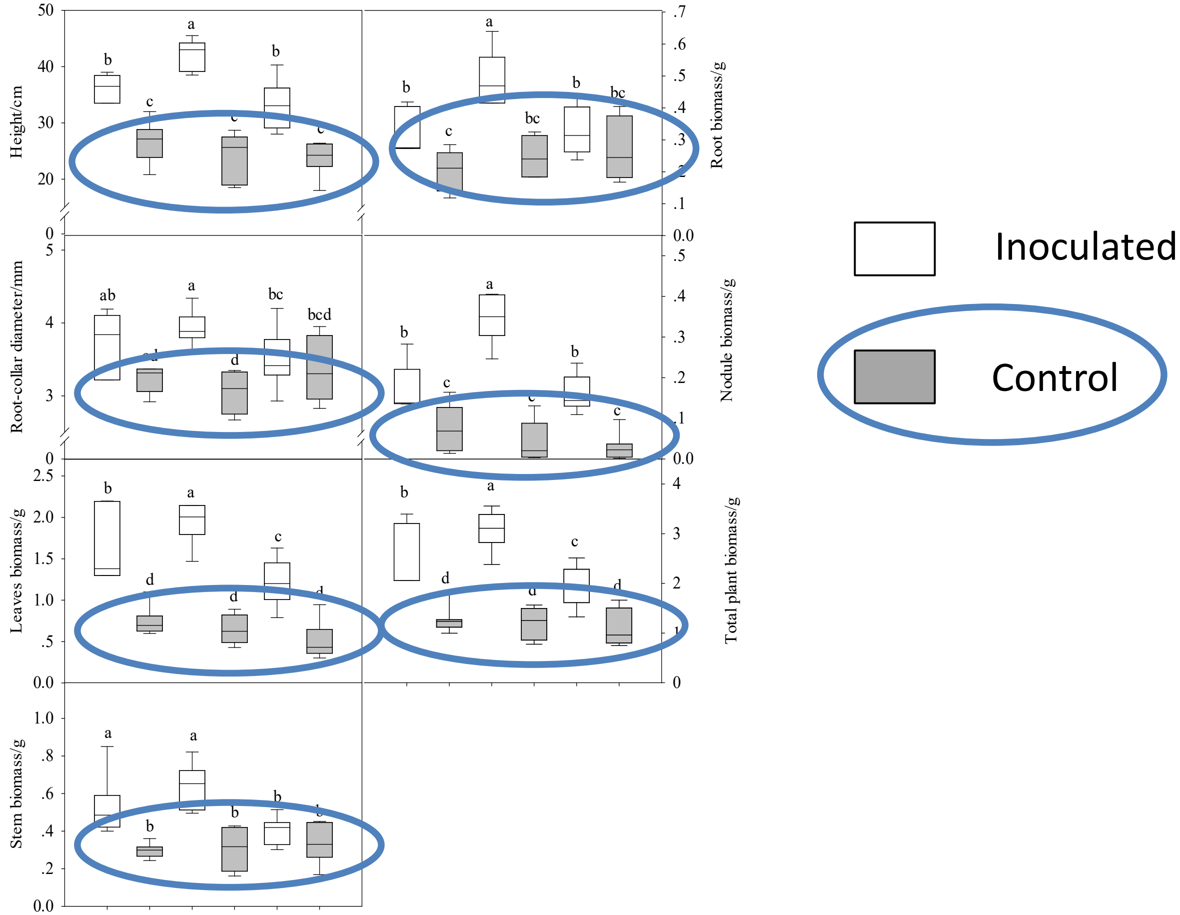
NH_4NO_3 (inorganic)



Zhang et al. (2019)

Results – Black Locust – Biochar

Height
RCD
Biomass
Nodule
Root
Shoot
Leaf
Total



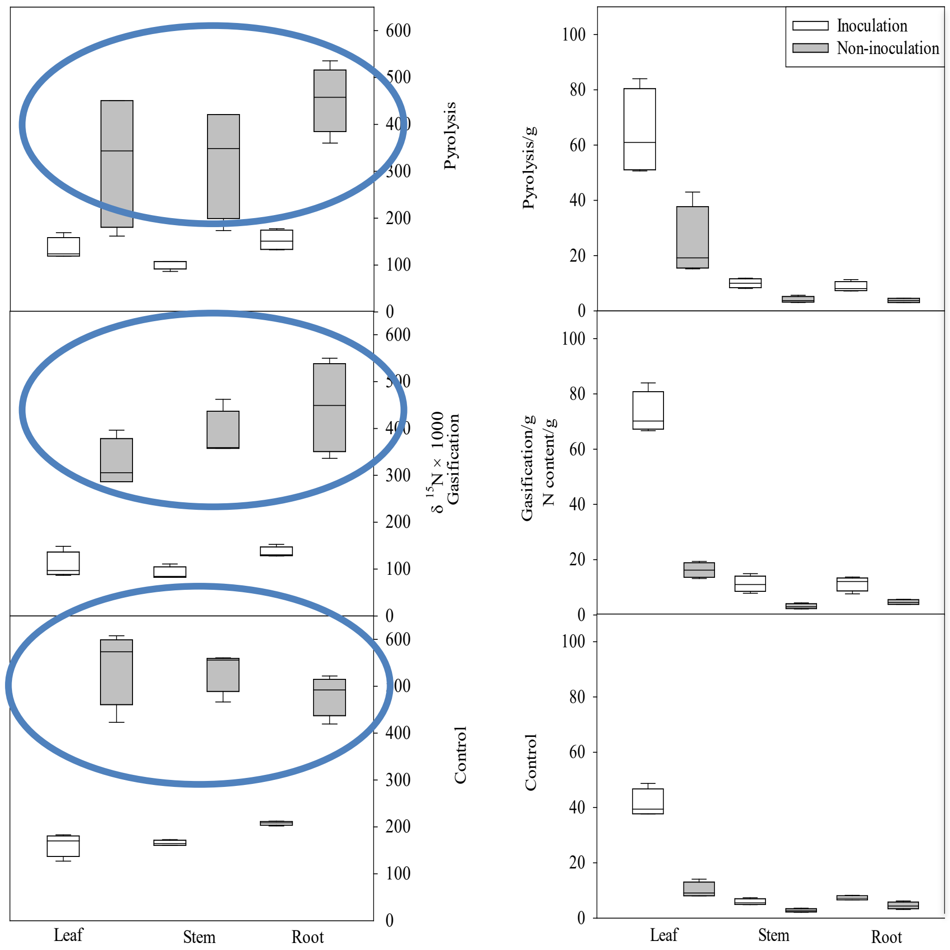
Sun et al. (in prep)

Results – Black Locust – Biochar

Pyrolysis

Gasification

No biochar



Inoculated

Control

Sun et al. (in prep)

Results – Black Locust – Biochar

Conclusions

- **N fertilization without inoculation improved growth; N form did not matter.**
- **Inoculation without N fertilization improved growth.**
- **For black locust, N fertilization + inoculation improved growth more than either factor alone.**
- **N-fixing *Rhizobium* colonize and fix N well despite nursery conditions of robust N fertilization.**
- **Biochar from gasification can further stimulate N-fixation.**

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Inoculation – DIY

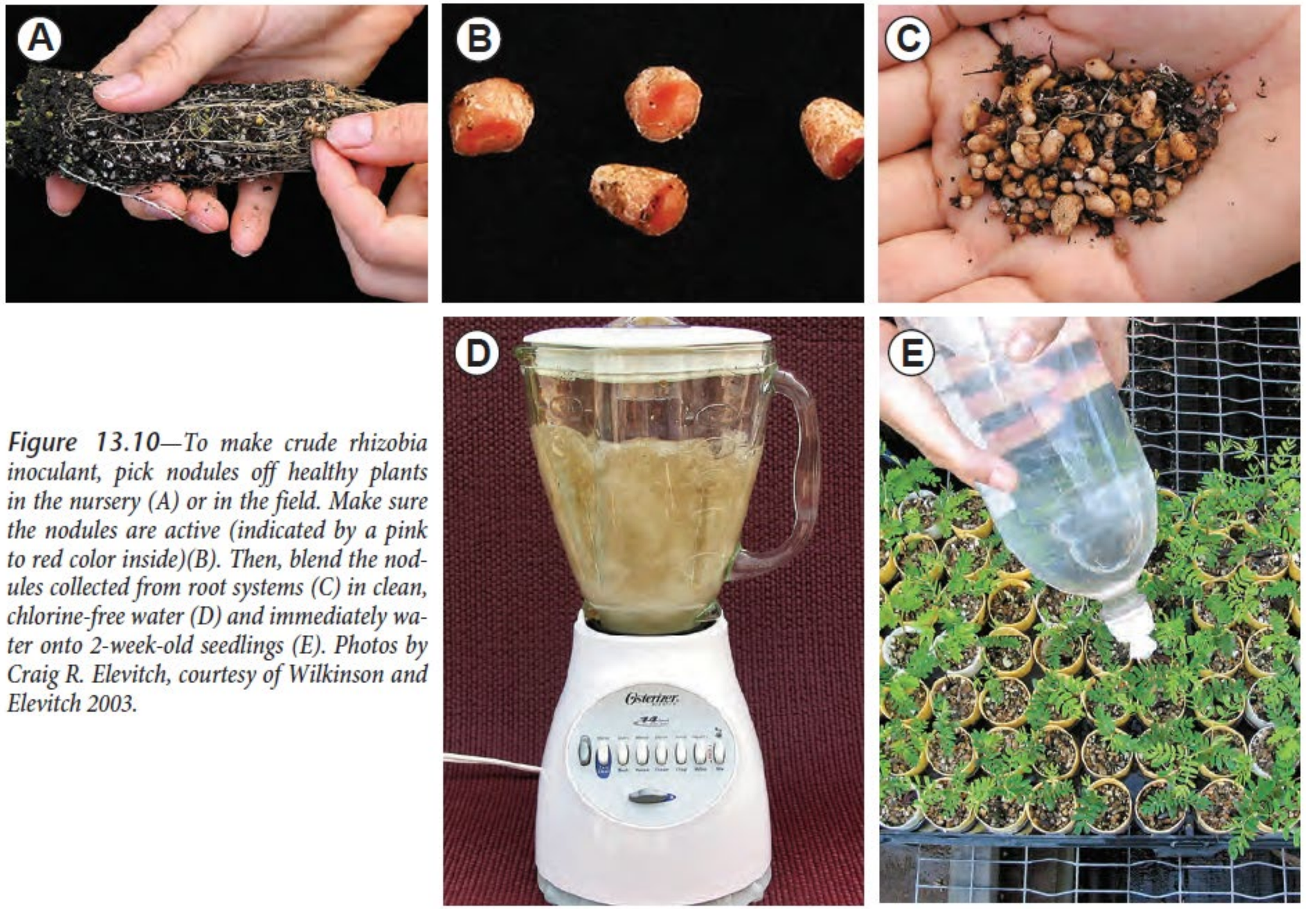

Figure 13.10—To make crude rhizobia inoculant, pick nodules off healthy plants in the nursery (A) or in the field. Make sure the nodules are active (indicated by a pink to red color inside)(B). Then, blend the nodules collected from root systems (C) in clean, chlorine-free water (D) and immediately water onto 2-week-old seedlings (E). Photos by Craig R. Elevitch, courtesy of Wilkinson and Elevitch 2003.

Wilkinson and Janos (2014)

Thank you!

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References (Note: these are included in the conference handout)

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