# GARLIC MUSTARD (*ALLIARIA PETIOLATA*) INVASION & IMPACTS:
## IMPLICATIONS FOR MANAGEMENT & RESTORATION

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# Outline

- Intro: garlic mustard invasion & impacts
- Research questions
- Garlic mustard's impacts on native herbs & AMF
  - Field Studies
  - Greenhouse Study
- Native herb impacts on garlic mustard
  - Field Study
- Conclusions: implications for restoration

Photo: Garlic mustard
From www.genevalakeconservancy.org

# Garlic Mustard (*Alliaria petiolata*)

- Introduced from Europe ~1860
- Invasive in hardwood forests
- Prolific reproduction
- Few "enemies"
- Displaces native plants & decreases diversity (Nuzzo1999, McCarthy 1997)


Top: Garlic mustard's distribution in North America (www.plants.usda.gov)
Bottom: Garlic mustard in seed (left); seedlings (right)

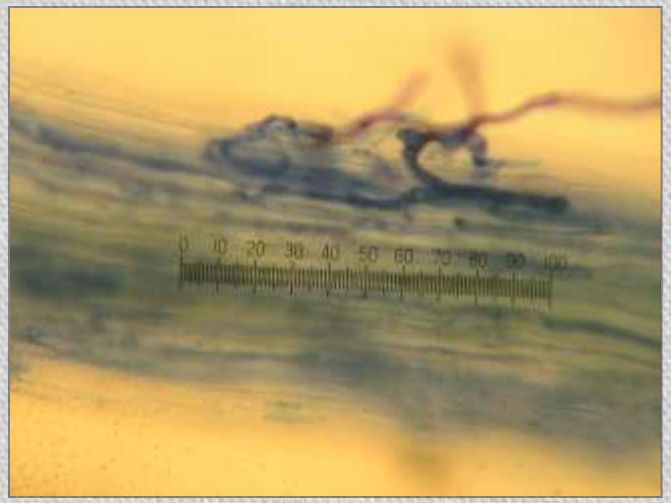
# Garlic mustard – life history

**1st year:**
Seedling &
Rosette

**2nd year:**
Adults,
Flowers,
Siliques (fruit)

# Garlic mustard - impacts

- Competition
  (Myers & Anderson 2003; Meekins & McCarthy 2005
- Alters nutrient levels
  (Rodgers et al. 2008)
- Chemical "weapons" (e.g. glucosinolates)
  (Cipollini & Gruner 2007; Roberts & Anderson 2001)



- Toxic to arbuscular mycorrhizal fungi (AMF)
  (Callaway et al. 2008; Roberts & Anderson 2001)
  - AMF important for nutrient/water acquisition
  - Reduced tree seedling growth in invaded soils (Stinson et al. 2006)

AMF hyphae infecting root of *Thalictrum dioicum;* stained with analine blue. 200x

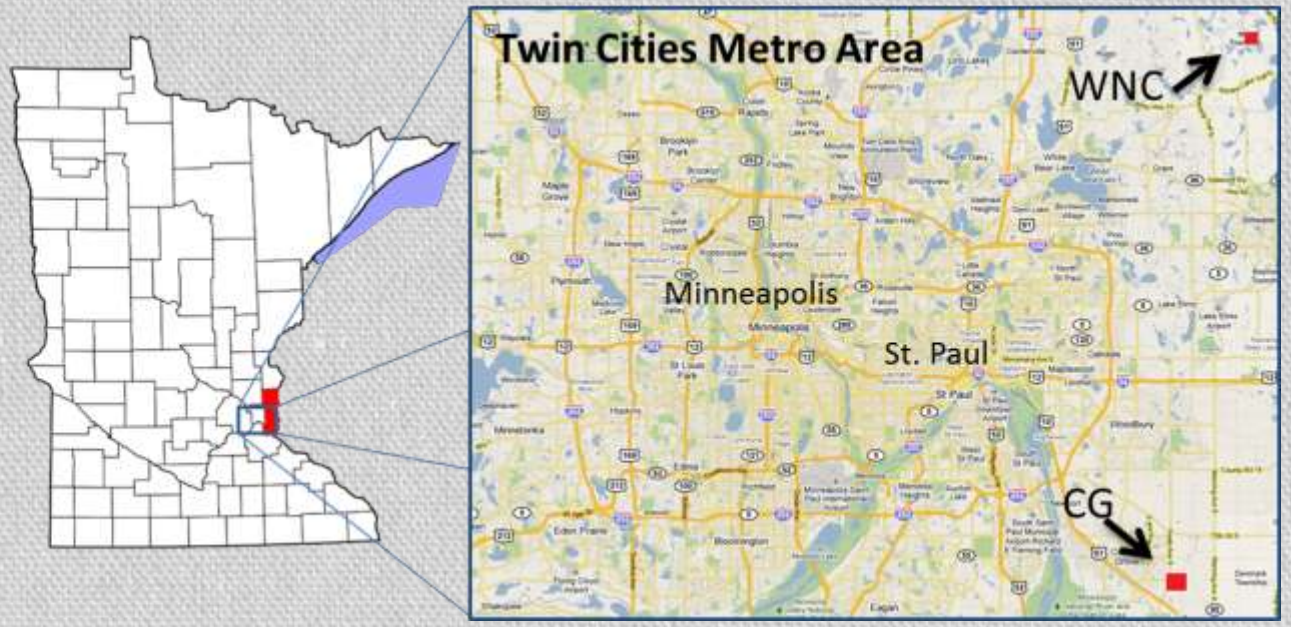
# Research Questions – native herbs & garlic mustard

- Broad questions:
- Is garlic mustard driving or responding to declines in native herb abundance/diversity?
- Do soil effects persist following garlic mustard removal ("legacy effects")
  - Implications for regeneration after biocontrol?

- Today's focus:
  - **Does garlic mustard reduce AMF colonization of herb roots?**
  - **Does garlic mustard negatively affect native herb growth?**
  - **Does native plant cover/richness reduce garlic mustard invasion?**

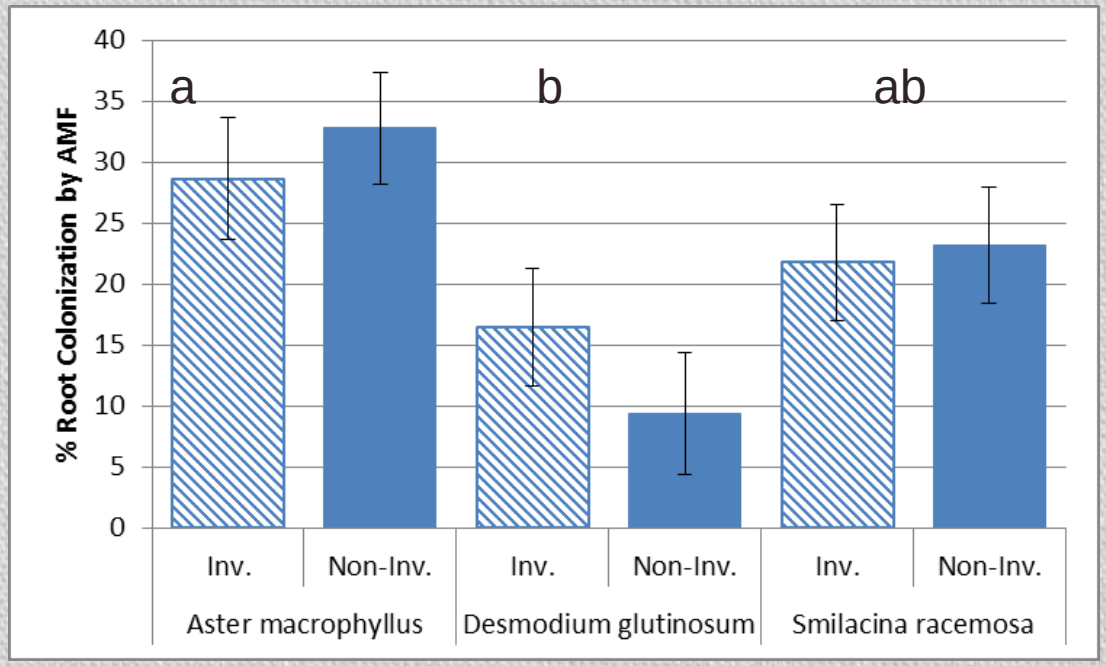
# Study Sites

- Warner Nature Center, Marine-on-St. Croix, MN
- Cottage Grove Ravine Regional Park, Cottage Grove, MN
  - Dry-mesic oak woodlands
- Greenhouse: Univ. of MN, St. Paul
  - Soils from WNC



# Impacts on AMF – observational study

- Site: Warner Nature Center
- 4 Native Spp. + Garlic mustard
- 12 root samples each from invaded & non-invaded area
- Stained with aniline blue; presence/absence of AMF recorded
- ANOVA – SAS 9.2




- **Results:**
  - GM & *Geranium maculatum*: 0% colonization

- Invasion: no significant effect on % root colonization
- Significant differences by species (a, b)

# Impacts on herbs & AMF: restoration study

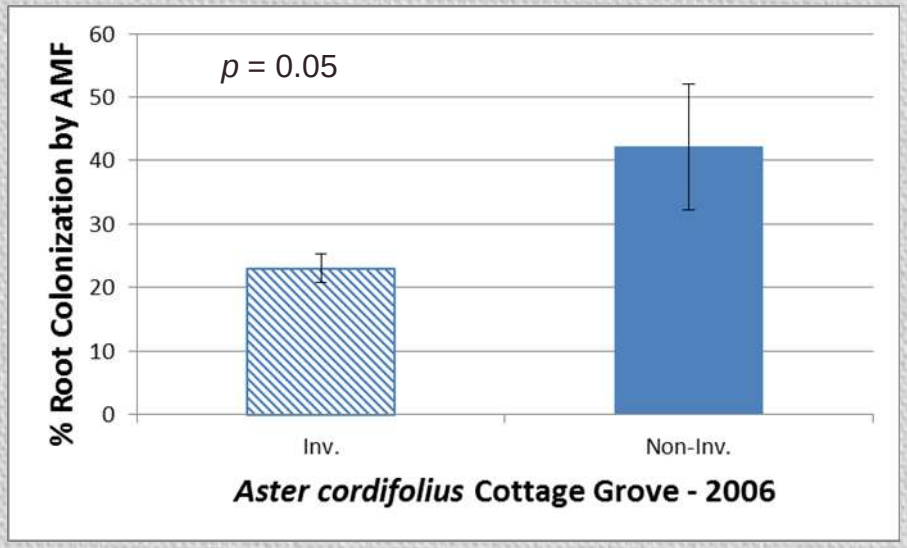
- 2 sites * 2 Invasion "treatments"
* 2 removal treatments * 2 years
- 96 1-$m^2$ plots (6 reps)
- 10 species planted/plot Fall 2006 & 2007
  - 2-3 plants/species/plot
- Response: Plant cover(biomass)/spp./plot
  - Mean % Root colonization/spp./plot


Non-invaded, full removal. Summer 2007.

# Restoration Study – preliminary results

- Plant Cover
  - Monthly summers 2007 & 2008
  - Repeated measures ANCOVA SAS 9.2
    - Site as random block effect
    - Light levels as covariate
- Preliminary results:
  - No significant differences by invasion treatment
  - Trend: higher cover in invaded plots
  - Significantly higher P in invaded areas may counteract loss of AMF or shift toward parasitism
- Next steps: Analysis of biomass

- AMF Colonization
  - Species * Invasion ANOVA SAS 9.2
  - Preliminary results:
    - Only species has significant effect ($p < 0.001$)
    - Within species, only *A. cordifolius* showed significant invasion effects



# Impacts on herbs & AMF – greenhouse study

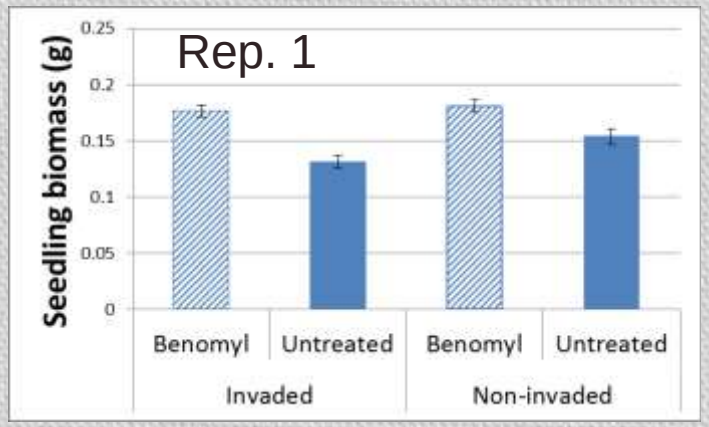
- Treatments
  - **Field soil**: invaded & non-invaded
    - 1:1:2 soil: potting mix: sterilized sand
  - **Benomyl** (fungicide) applications (Benomyl & None)
    - Applied weekly
- Factorial design
- 24 replicates/species
- Two experimental replicates
  - (March – July & July – Sept. 2009)
- 11 species planted by seed
  - Thinned to 1 seedling/cone
  - 7 species included in analysis

- Responses:
  - Plant biomass
  - % Root colonization

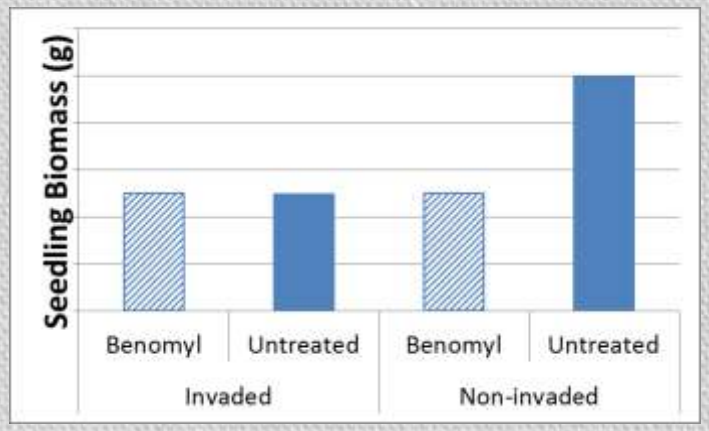
# Greenhouse study – biomass results

- No soil * fungicide interaction
- Rep. 1: Higher biomass in non-invaded ($p$ = 0.03)
- Rep. 2: Higher biomass in invaded for 5 species (Soil * Species $p$ = 0.02)
  - Soil changes or climate differences?
- Higher biomass in benomyl, both reps, all species, except *M. diphylla*
- (Species * Fungicide $p$ = 0.002; p = 0.04)
  - Release from pathogens (AMF or other fungi?)

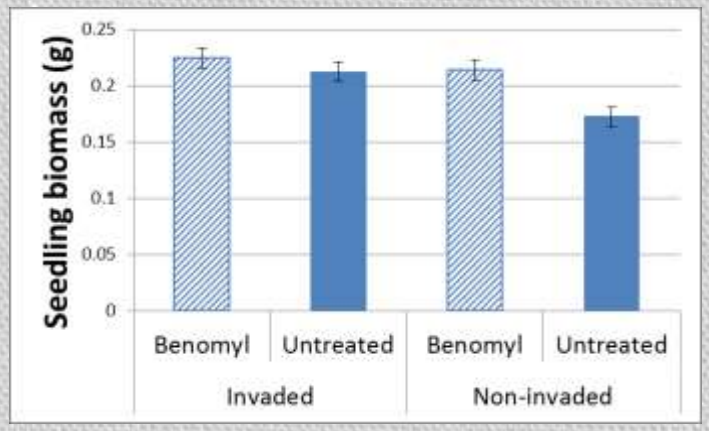
Observed Results




Expected Results




Rep. 2



# Garlic mustard impacts on herbs & AMF - conclusions

- **Does garlic mustard reduce AMF colonization of herb roots?**
  - Observation & Restoration Studies suggest: NO for most species tested
    - possibly species/site-specific responses
    - Possibly "senstive" AMF species already lacking from these sites
  - Greenhouse study – in progress
- **Does garlic mustard negatively affect native herb growth?**
  - Restoration Study suggests NO
    - Effects may be counteracted by increased nutrients?
    - Biomass analysis – in progress
  - Greenhouse study suggests: Sometimes yes, sometimes no?
    - Effects of garlic mustard/reduced AMF only manifest when plants are stressed?

# Native herbs – impacts on garlic mustard

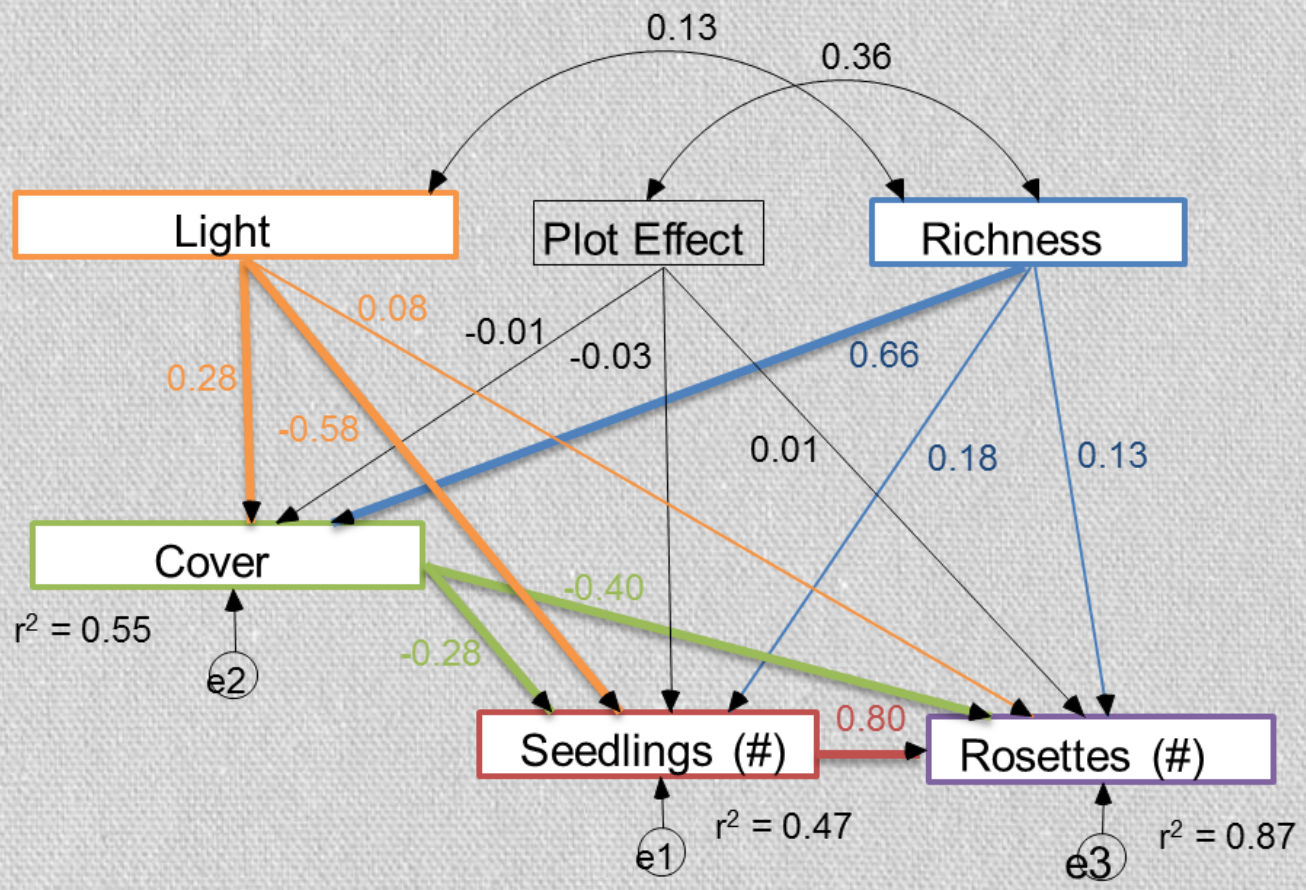
- 15 whole plots
  - 4 subplots – 0, 3, 6, & 10 spp. (2003)
  - 50 GM seeds planted Fall 2005
- Factors:
  - Species richness (0-10 species)
  - Native % cover
  - Light levels (DIFN – LAI 2000; growing season averages)
  - Other environmental variables
- Responses:
  - seedling #
  - rosette #
  - adult #
  - biomass (total/plot & avg./plant)
  - silique # (total/plot & avg./plant)

Study plot, Warner Nature Center

# Native herb impacts - analysis

- Structural Equation Model (SEM) – Amos Graphics, Student Ed.
  - Multivariate regression analysis
  - Test hypothetical interactions (pathways) between multiple variables
  - Standardized estimates – relative strength of pathways
    - Variables: Light, Richness, Cover, "plot effect" & garlic mustard responses
    - Plot effects –
    - Nonmetric Multidimensional Scaling (NMS) – PC Ord 5.32 to spatially distinguish plots based on environmental variables
      - Cumulative $r^2$ of selected axis 0.98
      - Defining variables: Nitrate ($No_3^-$) $r^2 = 0.94$ Phosphorus (P) $r^2 = 0.22$

# Native herb impacts – garlic mustard (1st year)



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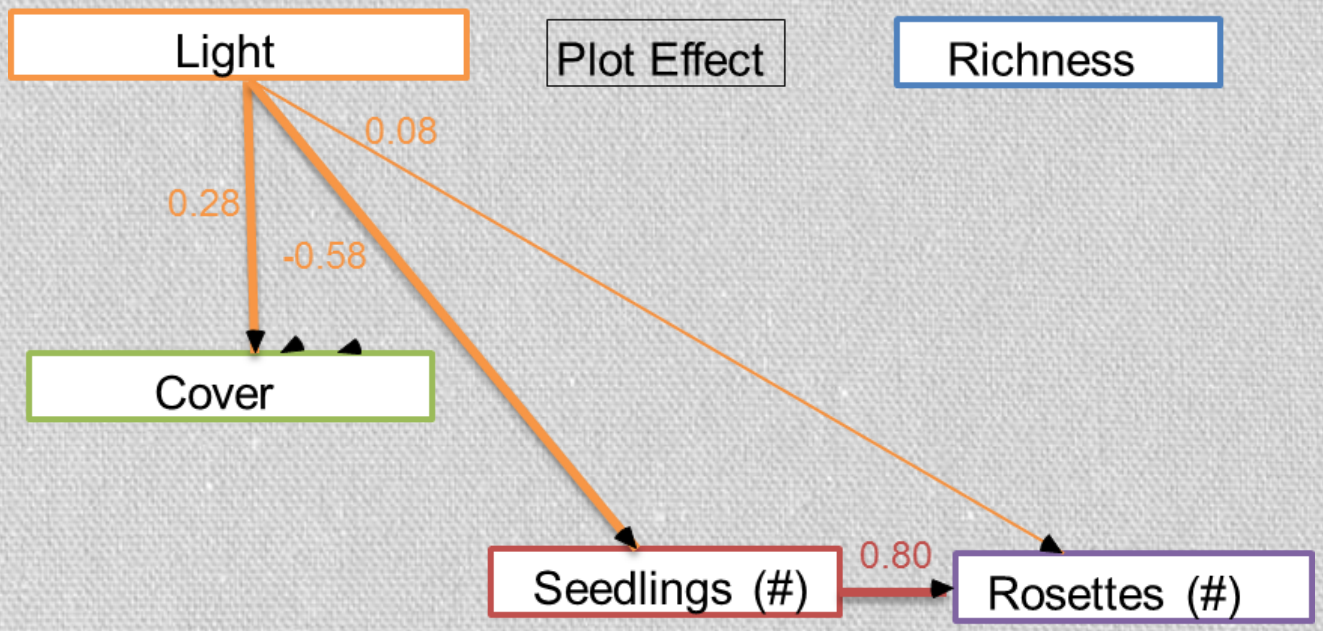

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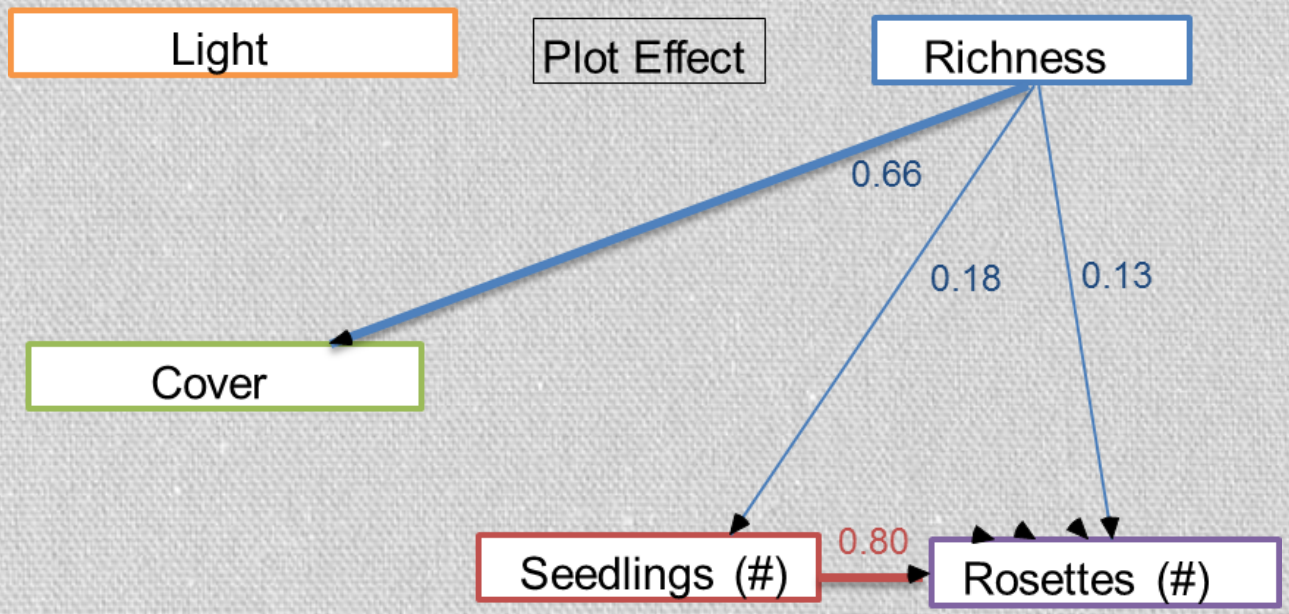

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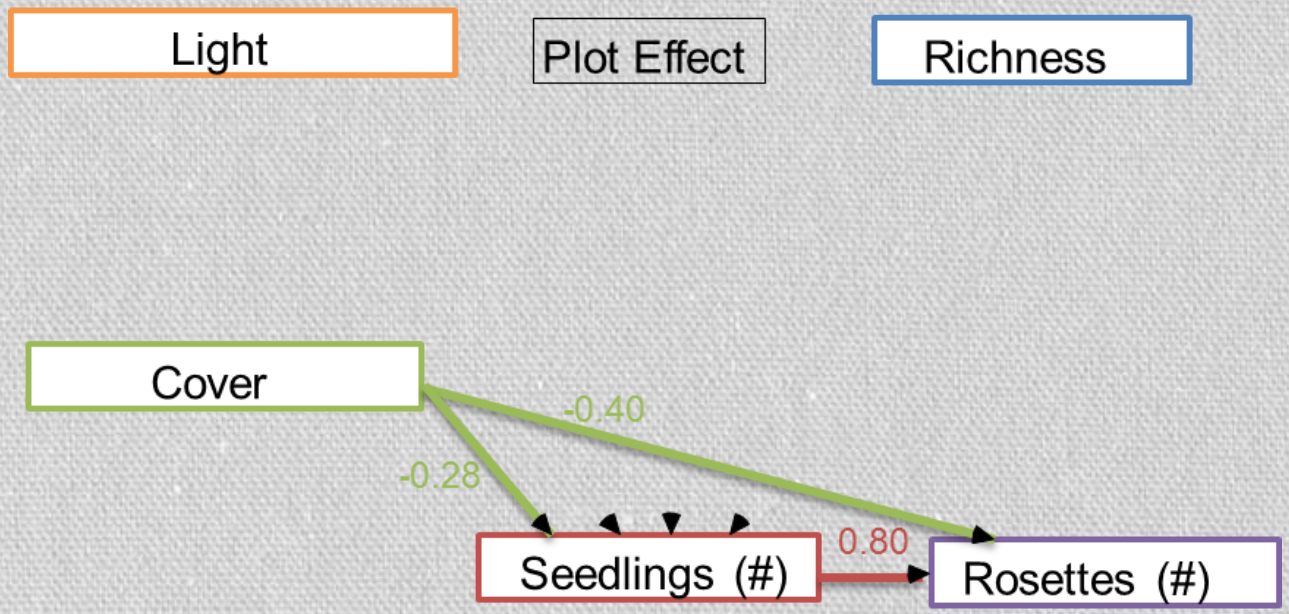

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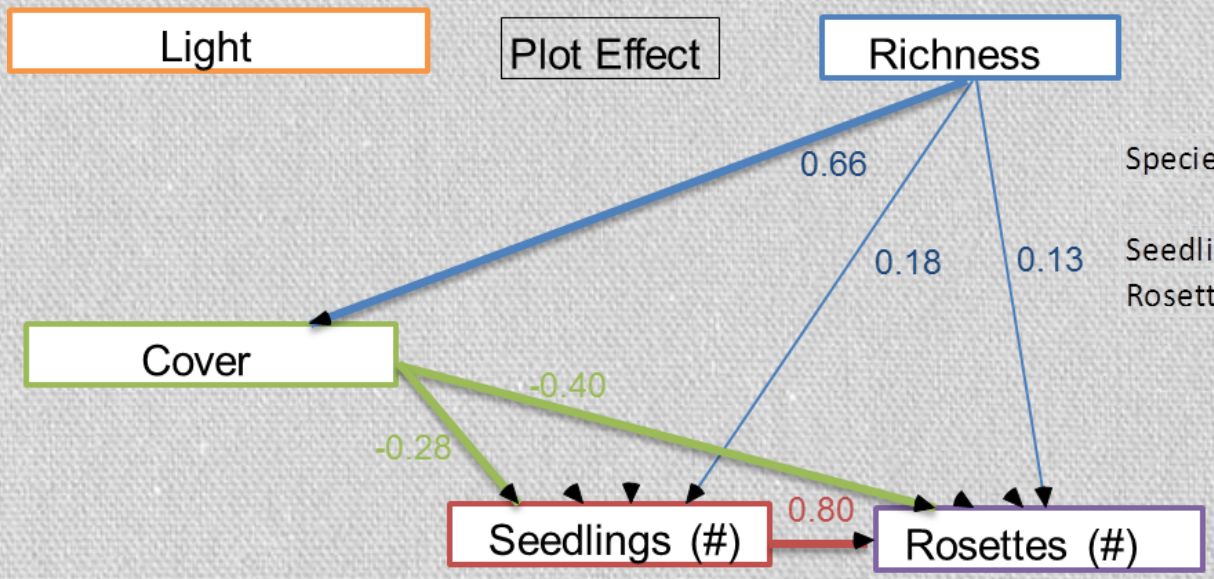
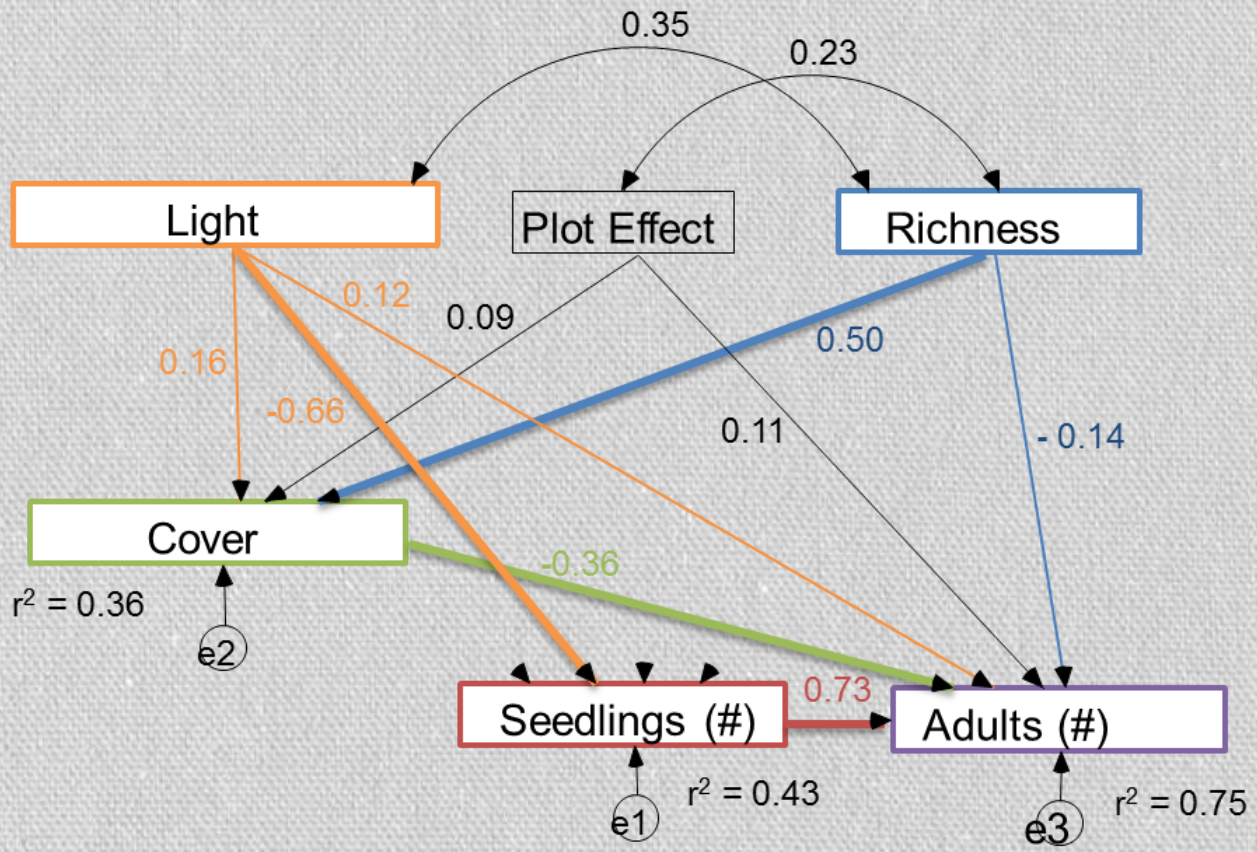



Species Richness

| | Indirect | Net Effect |
|---|---|---|
| Seedlings | -0.18 | 0.001 |
| Rosettes | -0.26 | -0.14 |

# Native herb impacts – garlic mustard (2nd year)



# Native herb impacts on garlic mustard - summary

| | Seedling # | Rosette # | *Adult #* | Avg. Biomass | Total Biomass | Avg. Siliques | Total Siliques |
|---|---|---|---|---|---|---|---|
| Light | - *p < 0.001* | +/- | +/- | + | + *p = 0.01* | + | + |
| Richness | +/0 | +/- *p = 0.07* | - *p = 0.09* | + | - | +/- | - |
| Cover | - *p = 0.05* | - *p < 0.001* | - *p < 0.001* | - *p < 0.001* | - *p < 0.001* | - *p < 0.001* | - *p < 0.001* |
| Seedling # | | + *p < 0.001* | + *p < 0.001* | - *p < 0.001* | - | - *p = 0.02* | - |

- +/- = Direct/total effects
- Light: positive effect on cover (year 1: *p = 0.002*; year 2: n.s.)
- Richness: positive effect on cover (*p < 0.001*; both years)
  - Negative NET effect of richness on rosettes and all adult measures

# Native herb impacts - conclusions

- Native plant cover has significant negative effects on garlic mustard establishment, growth, survival and reproductive output
- Species richness:
  - Indirect negative effect on garlic mustard via effects on cover
- "Empty" forest floors may be more vulnerable to invasion

- Light – strong negative effects on garlic mustard seedling establishment
  - Low-light forests may be more vulnerable to initial garlic mustard invasion

*Thalictrium dioicum* in high-cover plots at WNC

# Implications for restoration

- Native herbs may be somewhat resistant to (AMF) effects of garlic mustard
  - Long-term effects?
  - Multiple stressors?
    - Effects of lost AMF may only manifest in stressful conditions
    - Sensitive AMF may already be lost
    - Increased soil nutrients may counteract
- Potential for restoration following garlic mustard control

- Woodlands lacking ground layer may be most vulnerable to garlic mustard invasion
- Restoration/management to increase native herbs may help reduce reinvasion following garlic mustard removal

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