Fair trade underground:
Nutrient exchange in mycorrhizal symbioses

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The importance of mycorrhizal symbioses

For the Plant
- Phosphorus and N uptake from the soil
- Improved water relations
- Heavy metal nutrition and resistance

For The Fungus
- Pathogenic resistance to root pathogens

Inoculum Production
Global carbon economy
Horticultural uses
Low input agriculture
Arbuscular mycorrhizae versus Ectomycorrhizae (EcM)

• both occur in the fine root system

• difference in penetration of cortex cells
  • AM fungal hyphae penetrate the cortex cells forming vesicles and arbuscules (endomycorrhizae)
  • EcM do not penetrate cell walls of cortex cell
  • (ectomycorrhizae)
• Arbuscules - highly branched structures that are the site of nutrient transfer; they do not penetrate cell membrane; short-lived structures

• Vesicles - oval-shaped, darkly staining structures that are thought to function as nutrient reservoir
EcM do not penetrate cell walls of cortex cells

• EcM form a puzzled-shape covering of hyphae over the cortex cells - called a Hartig net
• site of nutrient transfer
• relatively long-lived structures
Alteration of root morphology

• VAM do not significantly alter root morphology
  • fine roots possess root hairs

• EcM alter root morphology; no root hairs; produce a mantle
Occurance
- estimated that 300,000 plant species to have VAM
  only ~150 spp. fungi participate
- est. 2000 plant species have EcM
  ~5000 fungi participate
- taxonomic distribution
  VAM restricted to Glomerales
  ectomycorrhiza in >70 genera in 9 orders
  (Basidiomycota, Ascomycota)
- host distribution
  ectomycorrhiza mostly trees
  Gymnosperms - e.g., Pinaceae
  Angiosperms - e.g., Fagaceae, Betulaceae, Salicaceae, Dipterocarpoideae, Myrtaceae
  VAM: woody & herbaceous plants
  90% of vascular plants normally assoc./w VAM fungi
AM and EcM mycorrhizal symbioses are mutualisms

Fungal benefits
• carbohydrates (photosynthates, monosaccharides) that are converted to trehalose, mannitol, glycogen
• necessary cofactors for spore germination

Plant benefits
• greater absorptive area
• uptake of P, N, Ca, K, Cu, Mb, Mg, Zn
• water
• protection against soil borne pathogens?
Ecosystem Distributions

VAM
- VAM are common in most habitats
- Dominant mycorrhizae in grasslands and tropical ecosystems

EcM
- dominant in coniferous forests, especially boreal or alpine regions
- common in many broad-leaved forests in temperate and mediterranean regions
- also occur in some tropical or subtropical savanna or rain forests habitats

Nonmycorrhizal (NM) Plants
- most common in disturbed habitats, or sites with extreme environmental (high latitude or elevation) or soil conditions
- appear to be more common in Australia than on other continents
Mycorrhizal fungi strongly influence plant communities.
Root colonization by Glomerales

hyphae enter the root through root hairs or by forming appressoria between epidermal cells;

hyphae grow intracellularly and also penetrate the cell walls of cortical cells, causing invagination of the plasma membrane.

form arbuscules and, if produced by a species, vesicles.
• highly branched haustorium-like structures
• extend through the host cell wall, but not cell plasma membrane
• increased surface area between the fungus and the host cell plasma membrane
• bidirectional transfer of metabolites and nutrients between the two mycorrhizal partners
• short-lived: remains alive only for a few days before disintegrating and being digested by the cells of the plant
• in a healthy VAM mycorrhizal relationship there is a continuous sequence of development and disintegration of arbuscules
The life cycle and morphology of an asexual coenocytic obligate symbiont:
A phylum of their own - Glomeromycota
Nutrient exchange is the basis for one of the world’s most important relationships.

Because of mycorrhizal symbioses, billions of tons of carbon move annually from shoots to roots, and hundreds of millions of tons of mineral nutrients move in the other direction.

Extraradical mycelium (ERM)
Studying metabolism in AM

- Soil culture
- Monoxenic culture
- Axenic culture

**Labeling with stable isotopes**

**In vivo**
- NMR & MS

**In vitro**
- Radiolabeling (regulation)
- Gene expression (regulation)
- Gene identification (validation)

NMR & MS spectra

(Interpretation)

- Trehalose
- Triose
- Acetyl CoA
- Glyoxylate
- Lipids
- Exogenous hexose
- CO₂
- OAA
- Glutamate
- Arginine

**TCA**

Interpretation of metabolites:

- Exogenous hexose
- Trehalose
- Triose
- Acetyl CoA
- Glyoxylate
- Lipids
- Exogenous hexose
- TCA
Carbon movement in the AM symbiosis

Hexose → TAG

Hexose → Trehalose

Hexose → PPP

PPP → Triose

Triose → TAG

TAG → Glyoxylate cycle

β-oxidation

Glycogen

Hexose → PPP

Trehalose → Triose
Carbon Transfer between plants via the mycorrhizal network?
Carbon supplied as 14C glucose to one root system is transferred to a second root system via a common mycorrhizal network.

14C in donor (▲), and recipient (▼), mycorrhizal roots

14C in extraradical hyphae of donor (▲) and recipient (▼) root compartments

14C in the medium of donor (▲) and recipient (▼) root compartments
But mass spectra of fungal and host metabolites after supplying the donor roots with 13C-1 glucose show that the carbon transferred remains in fungal compounds.

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**Trehalose**

- Unlabeled standard = natural abundance 13C level
- Extracted from donor roots
- Extracted from recipient roots
- Simulated assuming 12.5% labeling in alternate positions

**Sucrose**

**Fatty acid (16:1)**
Carbon transferred between root systems (usually) stays in the fungus.
A guess about the movement of Nitrogen

Hexose

Intraradical carbon pool

NH$_4^+$

Urea

Orn

Arg

PolyP

AA’s

Extraradical carbon pool

NO$_3^-$

NH$_4^+$

Gln

Arg

PolyP

P
Labeling in free amino acids after exposure to $^{15}$NH$_4^+$ in the fungal compartment as measured by GC mass spectrometry.
Labeling in free amino acids after exposure to $^{15}\text{NH}_4^+$ in the fungal compartment for 1 week

Free amino acids of mycorrhizal roots

Free amino acids of extraradical hyphae
Average 15N Enrichment in Root Protein Amino Acids following 15N and/or 13C labeling of the ERM for 8 Weeks

<table>
<thead>
<tr>
<th>Label Supplied</th>
<th>% label in Protein Amino Acids</th>
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<tr>
<td>13C-1,2 acetate + 15NO3</td>
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<td>13C –2 acetate</td>
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<tr>
<td>Tyr</td>
<td>-2.8</td>
</tr>
</tbody>
</table>

But carbon doesn’t accompany the nitrogen.
Arginine is translocated from ERM to IRM

Mass spectrometry data of 13C Arg labeling expt

Mass isomers of arginine (m/z)

Unlabeled standard
Arg in unlabeled roots
Arg in ERM after labeling
Arg from roots after labeling
Arg from root protein
Arg from root protein+*Arg
Comparing the expression of genes involved in primary nitrogen metabolism in the intraradical and extraradical fungal tissues

- Glutamine Synthetase
- Glutamate dehydrogenase
- Ornithine amino-transferase
- Urease associated protein
Current status of testing the model
Carbon status regulates Phosphorus handling

Sinks → Growth → Photosynthesis → Sucrose → Sucrose

Hexose → Hexose → Hexose-P → Hexose-P

PolyP → PolyP

Host

Interfacial Apoplast

Soil

ERM

IRM

P_i