

Role of Microbes in Resource Management in Arid Ecosystems

Jerry R. Barrow
Bobby D. McCaslin

Abstract—Three major classes of endophytic fungi were observed in the roots of native grasses and shrubs: (1) vesicular-arbuscular mycorrhizae (VAM), which are known to be important in water and nutrient uptake; (2) septate fungi that formed non-destructive interfaces within root cortex cells and exhibited mycorrhizal like characteristics; and (3) chytridiomycetes were the third major class of fungi observed and preliminary studies suggest they regulate colonization of VAM, septate fungi, and nutrient uptake. A hypothesis is proposed on how these fungi function in accessing and managing nutrients and water for survival in arid ecosystems.

In arid ecosystems many plant species live in harmony and are efficient and successful in accessing and managing limited nutrient and water resources. Generally, desert plants survive extended drought punctuated only briefly by cyclic and unpredictable precipitation events where moisture and nutrient mobility are sufficient for essential plant processes. We propose that this extraordinary task is accomplished by symbiotic associations. It is well documented that several classes of mycorrhizae increase production and survival of most vascular plants by enhancing water and nutrient uptake and in some cases they are essential for host plant survival (George and others 1991). Lichens are symbiotic associations between photosynthetic algae or bacteria with fungi. This association allows them to chemically weather rock surfaces through organic acid production to access highly immobile nutrients (Johnston and Vestal 1993). Similar microbe assisted mechanisms to access and manage nutrients and water in arid ecosystems may be essential for the survival and productivity of desert plants.

We surveyed dominant grasses and shrubs from the southwestern US and analyzed roots for endophytic fungal associations. Several experiments were conducted using native plants, alfalfa as a model plant, different fungi, nutrient levels and carbon sources. A brief summary of the results and the development of a hypothesis are presented here.

Incidence of Endophytic Fungi

Small (less than 1 mm in diameter) feeder roots of dominant grasses and shrubs were collected at different times from native populations from many sites varying dramatically in elevation, soil type, temperature, and precipitation.

In: Barrow, Jerry R.; McArthur, E. Durant; Sosebee, Ronald E.; Tausch, Robin J., comps. 1996. Proceedings: shrubland ecosystem dynamics in a changing environment; 1995 May 23-25; Las Cruces, NM. Gen. Tech. Rep. INT-GTR-338. Ogden, UT: U.S. Department of Agriculture, Forest Service, Intermountain Research Station.

Jerry R. Barrow is a Plant Geneticist, USDA, Agricultural Research Service, Jornada Experimental Range, Box 30003, NMSU, Dept. 3JER, Las Cruces, NM 88003. Bobby D. McCaslin is a Professor, Agronomy and Horticulture, New Mexico State University, Box 30003, Dept. 3Q, Las Cruces, NM 88003.

Roots were cleared by the method of Brundrett and others (1984) and stained with either acid fuchsin, trypan blue, or chlorazol black E, all specific for fungal tissue. Stained roots were analyzed microscopically for fungal colonization. In summary, three major classes of endophytic fungi were observed in both grasses and shrubs.

Hyphae and vesicles of vesicular-arbuscular mycorrhizal fungi are considered to be important in most plant species for nutrient and water uptake. Arbuscules, an important VAM structure, responsible for carbon and nutrient transfer between the host and fungus (Gianinazzi and Gianinazzi-Pearson 1979; Cox and Tinker 1976) were rarely observed. Since these structures are short lived when observed under mesic conditions, their existence under arid conditions would be brief and rare.

Secondly, root cortex cells were also consistently colonized both inter- and intracellularly by septate fungi. These formed non-destructive interfaces within the cells, occasionally a sheath surrounded the root, varying from a very loose to a dense fungal mat. Most frequently, however, the extramatrical hyphal growth extended directly from the root surface into the adjacent soil and aggregating soil and sand particles adjacent to the root surface. A septate isolate from fourwing saltbush formed "ectendomycorrhizal like" associations with alfalfa root organ cultures with internal interfaces similar to those observed in the native population. This isolate was identified as *Fusarium solani* by Dr. Robert Linderman at Oregon State University. Several additional isolates from native plants species also formed "mycorrhizal like" associations with root-organ cultures. Presently, positive identification of the other isolates has not been made, but they appear to be common soil saprobes.

A seedborne septate fungus, *Alternaria alternata*, was found consistently on the seed capsule or utricle of fourwing saltbush (*Atriplex canescens*) and formed similar non-destructive interfaces with the cortex cells of the emerging radicle. We compared vigor of seedlings from seed excised from the utricle with intact seed germinated on carbon free and cellulose supplemented medium. Seedlings from excised seed were significantly more vigorous when germinated on cellulose supplemented medium compared to carbon free medium. This suggested that internal microbes (we suspect chytrids) and septate fungi utilized cellulose for seedling vigor. Seedlings from intact seeds were more vigorous than excised seeds, suggesting that the septate fungi decomposed the utricle and transferred nutrients to the seedling. Maximum seedling vigor was obtained by germinating intact seeds on cellulose supplemented medium and suggests that the septate fungi accesses nutrients not only from the utricle but from external organic matter. Orchid mycorrhizae have been shown to access carbon from organic resources and transfer it to the developing host plant. These fungi have been identified as species pathogenic to other plants or are decomposing fungi (Harley and Smith 1983).

Similar septate fungi as well as VAM were observed in cultivated alfalfa populations. Alfalfa plants either inoculated with VAM + septate, septate fungi, or from surface sterilized seed were grown in phosphorus (P) deficient soil, or in P deficient soil supplemented with rates of elemental P ranging from 500 to 4,500 pounds per acre. These P rates are two to nine times greater than recommended fertilization rates in New Mexico and are considered excessive. Plants inoculated with VAM + septate fungi had the maximum growth response and plants inoculated with septate fungi were intermediate but significantly larger than the sterile controls in the non-P supplemented soil, suggesting a benefit from the septate fungi in P uptake. However, sterile plants were significantly larger than inoculated plants at all supplemental P treatments. This demonstrates that both VAM and septate fungi enhance P uptake at low concentrations and restrict or regulate uptake at high concentrations. In another experiment where toxic concentrations of mineral salts accumulated, plants inoculated with septate fungi were protected while non-inoculated plants died, further illustrating these organisms regulate mineral uptake.

No VAM or septate fungi were observed in plants from surface sterilized seed. However, from transmission electron microscope studies, seedlings from surface sterilized seed are colonized with internal microbes, which we suspect were either chytrids or bacteria that influence colonization of VAM and septate fungi, hydrolysis of cellulose, and nutrient uptake.

In summarizing several experiments we found that the morphological expression and colonization of VAM, septate fungi, and chytrids was greatly influenced by the level of P fertilization. Maximum expression of all three organisms were observed at low levels of P fertilization and fungal expression decreased as P concentrations increased.

Roots of broom snakeweed (*Gutierrezia sarothrae*) were also colonized with a septate fungus that formed dense sand and soil aggregates at the root surface. This isolate also formed "mycorrhizal like" associations with root organ cultures similar to those reported above. Scanning electron and petrographic microscope studies of the attachment of the fungal hyphae with clay "skins" coating the sand particles and extensive hyphal networks within porous caliche suggested a nutrient harvesting role of these fungi.

Discussion

Benefits of VAM fungi are well known and documented (Harley and Smith 1983). Peyronel (1924) reported that non-pathogenic septate fungal colonization was common in plants and suggested that they may have potential importance. Odell and Trappe (1992) found widespread septate fungi on native legumes in Oregon and Washington and found no evidence of harm to the host. Bethlenfalvai (1992) reviewed potential roles of mycorrhizal fungi in the soil other than nutrient uptake such as serving as agents of nutrient transport between the host plant and microbe populations in the soil. In essence, they are the means by which other soil microflora receive carbon to function. Miller and Jastrow (1994) described the contribution of mycorrhizal fungi to soil aggregation. First, the hyphae form a skeletal structure that physically holds soil particles by physical entanglement. Second, roots and hyphae produce physical and chemical conditions, such as organic and amorphous materials for the

binding particles. Third, the hyphae and roots, by forming these aggregates, create a means of water, carbon, and nutrient storage in the soil.

We propose that these three endophytic classes of fungi function in harmony or as a suite of organisms. They form non-destructive interfaces with the host cortex cells that allow bidirectional movement of water, carbon, and nutrients between the host, microbes, and soil. Nutrients and water would be accessed by the trophobiotic endophytic fungi and associated microbes from immobile organic and organic resources. These resources would be accumulated and immobilized in soil aggregates, or in fungal tissue, under its metabolic control. This would enhance survival of the host during extended stress periods such as drought. Ling-Lee and others (1975) suggested that phosphorus absorbed by uninfected roots is readily translocated throughout the plant. In ectomycorrhizal infected roots, phosphate is taken up into the sheath and is distributed between two phosphate pools: a small pool that supplies an immediate source of metabolic phosphate to the host and a larger immobile storage pool. When external phosphorus uptake ceases, phosphorus is slowly remobilized from the large pool and transferred to the metabolic pool, where it becomes available to the host. A similar mechanism operating between these endophytic fungi and the host would be a survival mechanism during periods of stress.

References

- Bethlenfalvai, G. J. 1992. Mycorrhizae and crop production. In: Mycorrhizae in Sustainable Agriculture. ASA Special Publication No. 54:1-27.
- Brundrett, M. C.; Piche, Y.; Peterson, R. L. 1984. A new method for observing the morphology of vesicular-arbuscular mycorrhizae. *Can. J. Bot.* 62:2128-2134.
- Cox, G.; Tinker, P. B. 1976. Translocation and transfer of nutrients in vesicular-arbuscular mycorrhizas. The arbuscule and phosphorus transfer: a quantitative ultrastructural study. *New Phytol.* 77:371-378.
- George, E.; Hausler, K.; Kothari, S. K.; Xiao-Lin, L. 1991. Role of mycorrhizal hyphae in nutrient and water uptake of plants. Abstracts of the Third European Symposium on Mycorrhizas. Mycorrhizas in Ecosystems—Structure and Function. August, 1991. Sheffield, England. p. 26.
- Gianinazzi, S.; Gianinazzi-Pearson, V. 1979. Enzymatic studies on the metabolism of vesicular-arbuscular mycorrhiza. III. Ultrastructural localization of acid and alkaline phosphatase in onion roots infected by *Glomus mosseae* (NICOL. & GERD.). *New Phytol.* 82:127-132.
- Harley, J. M.; Smith, S. E. 1983. Mycorrhizal symbiosis. Academic Press. New York.
- Johnston, C. G.; Vestal, J. R. 1993. Biogeochemistry of Oxalate in the Antarctic Cryptoendolithic Lichen-Dominated Community. *Microb. Ecol.* 25:305-319.
- Ling-Lee, M.; Ashford, A. E.; Chilvers, G. A. 1975. A histochemical study of polysaccharide distribution in eucalypt mycorrhizas. *New Phytol.* 75:329-335.
- Miller, R. M.; Jastrow, J. D. 1994. Vesicular-arbuscular mycorrhizae and biogeochemical cycling. In: Pflieger, F. L. and Linderman, R. G., (eds.) Mycorrhizae and Plant Health, APS Press. St. Paul, MN: 189-212.
- Odell, T. M.; Trappe, J. M. 1992. Root endophytes of lupin and some other legumes in Northwestern USA. *New Phytol.* 122:479-485.
- Odell, T. M.; Massicotte, H. B.; Trappe, J. M. 1993. Root colonization of *Lupinus latifolius* agardh. and *Pinus contorta* Dougl. by *Phialocephala Fortinii* Wang & Wilcox. *New Phytol.* 124:93-100.
- Peyronel, B. 1924. Prime ricerche sulla micorize endotrofica e sulla microflora radice normale della fanerogama. (Initial studies on endotrophic mycorrhizae and on the normal root microflora of phanerogams.) *Revista di Biologia.* 6:17-53.