



Building Soil Organic Matter Biologically

The Benefits of Mycorrhizal Seed Inoculation

by Mike Amaranthus, Jeff Anderson,
Zack Amaranthus & Dave Perry

Loss of organic matter in soils following conversion of natural to agricultural ecosystems has been a large historical source of the steep rise in global levels of CO₂ (see “Soil Life & Carbon,” *Acres U.S.A.*, March 2008). Conversion has resulted in a 60 percent loss of soil organic matter in temperate regions and a 75 percent loss in cultivated tropical soils. It is estimated that up to a third of all of the increase in global CO₂ since the industrial revolution can be attributed to carbon losses from soils as a result of agricultural practices.

Soils are also a potentially powerful sink for accumulating carbon in soil organic matter. Atmospheric CO₂ can be recaptured in the soil under a variety of conditions. Activities that have been shown to facilitate carbon movement into soils include those that slow soil decomposition rates, input greater amounts of plant biomass, reduce soil erosion, and produce glomalin as a result of mycorrhizal activity. Land management practices such as no-till, winter cover crops, perennial crops, manure and compost inputs are being studied for their ability to build soil carbon inputs.

Of recent interest has been the discovery of glomalin in 1996 by Agricultural Research Scientist Sara Wright. Produced by arbuscular mycorrhizal fungi, glomalin permeates organic matter, binding it to silt, sand, and clay particles. Not only does glomalin contain 30 to 40 percent carbon, but it also forms aggregates that add structure to soil and keep other stored soil carbon from escaping. Studies have shown glomalin

can represent up to 30 percent of the total carbon in soil and can last 40 years.

The only producers of glomalin are arbuscular mycorrhizal fungi, which are found living on plant roots around the world. Wright named glomalin after *Glomales*, the taxonomic order to which arbuscular mycorrhizal fungi belong. The arbuscular mycorrhizal relationship commonly referred to as *endomycorrhizae* occurs with approximately 80 percent of the world’s plants species in their native habitat. The fungi use carbon from the plant to grow and make glomalin and a variety of other organic compounds in the soil. In return, the fungi’s hairlike filaments, called *hyphae*, dramatically extend the reach of plant roots. These hyphae function as pipes to funnel more water and nutrients to the plant. Unfortunately many factors such as erosion, organic matter removal, compaction, cultivation, fallow, and the use of certain chemical fertilizers and pesticides have reduced and in many cases totally eliminated mycorrhizal fungi from large expanses of disturbed lands.

Crop lands and grasslands around the world are now being recognized as potentially valuable for offsetting carbon dioxide emissions from industry and vehicles. In fact, some private markets have already started offering carbon credits for sale by owners of such land. However, little is known regarding growing practices and how they might affect the relative rates of carbon buildup in the soil. How might adding mycorrhizal fungi back to agricultural soils affect carbon sinks? How might carbon inputs be influenced by glomalin production? To address such

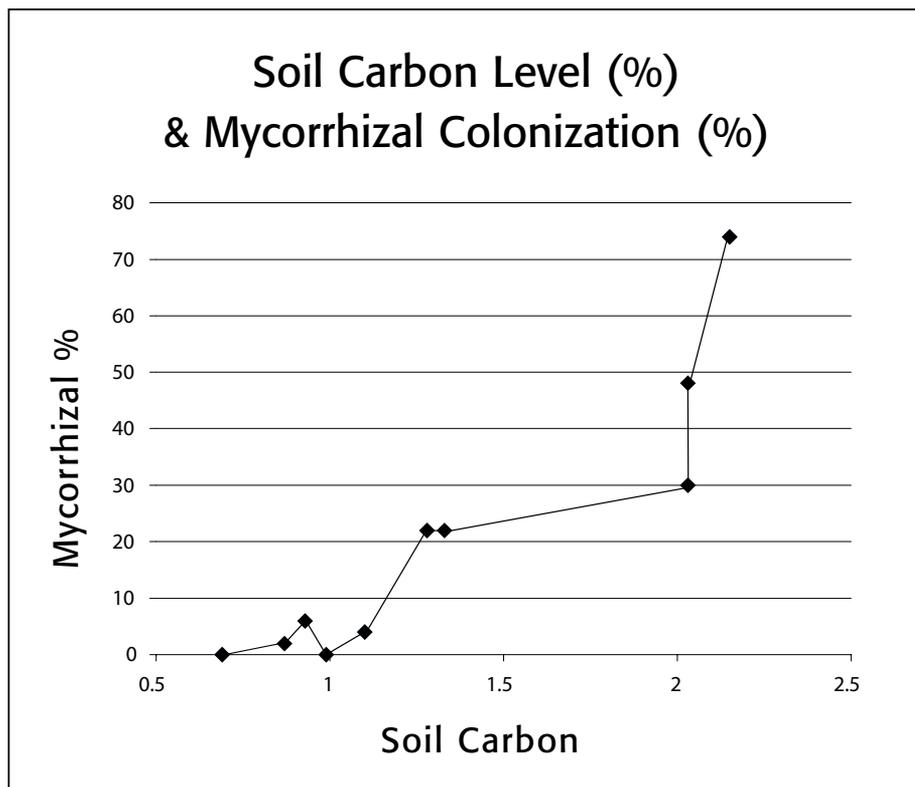
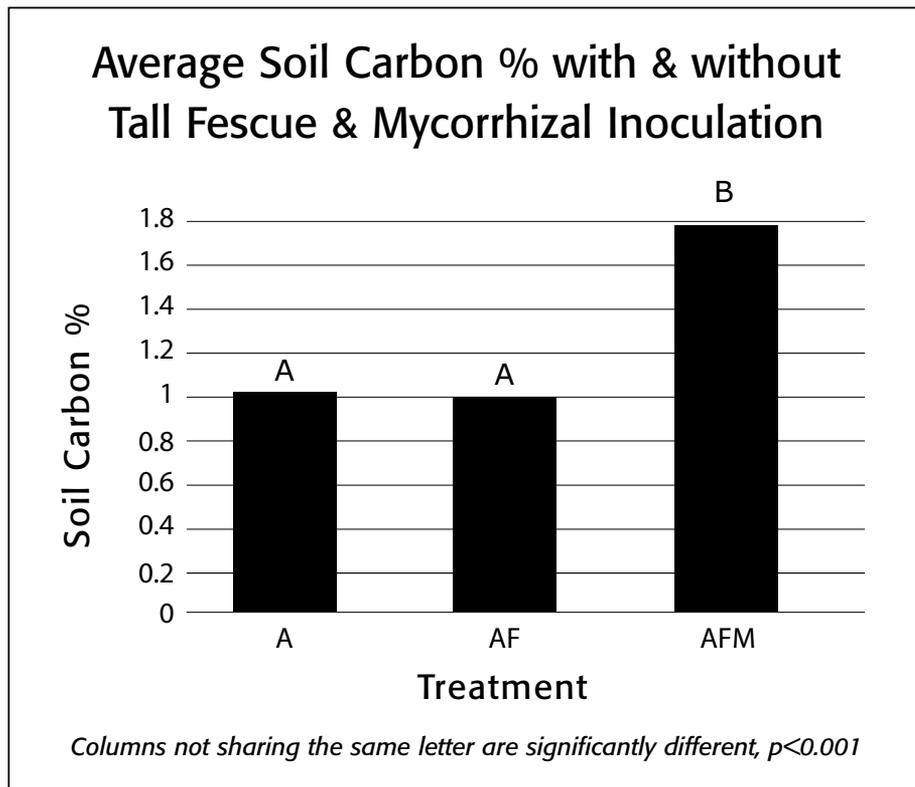
questions, we designed a study to examine tall fescue grown with and without mycorrhizal inoculation and to determine the level of carbon and glomalin in the soil at the end of one year.

METHODS

Eighteen five-gallon pots were filled with a non-swelling bentonite clay (Ag-sorb) and amended with 15 grams slow-release Apex fertilizer 23-2-11. There were three treatments: bentonite only (treatment A), bentonite seeded with tall fescue (treatment AF), and bentonite seeded with tall fescue and inoculated with a mycorrhizal inoculant (MycApply; treatment AFM). On July 8, 2007, treatment A pots were filled with bentonite, and 15 grams of fertilizer was mixed thoroughly through the soil media. Treatment AF pots were filled with bentonite with 15 grams of fertilizer mixed thoroughly through the soil media. AF pots also received 20 grams of tall fescue seed spread evenly across the top of the soil and capped with a 0.5 centimeter layer of bentonite. Treatment AFM pots were filled with bentonite with 15 grams of fertilizer mixed thoroughly with the soil media. AFM pots also received 20 grams of tall fescue seed spread evenly with 5 grams endo-granular mycorrhizal inoculant across the top of the soil and capped with a 0.5 centimeter layer of bentonite. Each gram of granular inoculant contained 32 propagules each of mycorrhizal fungi *Glomus intraradices*, *Glomus aggregatum*, *Glomus mosseae* and *Glomus etunicatum*. Each treatment was replicated six times in a randomized block design.

On July 11, 2008, pots were brought into the lab. In AF and AFM treatments, 0.2 grams of grass roots were removed from the upper and lower third of each pot and placed in perforated plastic capsules for analysis of percent mycorrhizal colonization. To clear the roots, capsules were submerged in KOH for 72 hours, rinsed and placed in HCl for 15 minutes.

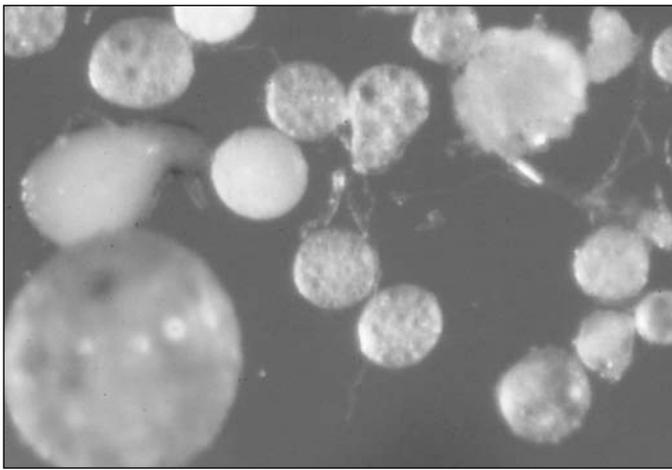
After rinsing, the capsules were submerged for one hour in blue indigo Indian ink for staining of the mycorrhizal structures. Root samples were then spread evenly on a gridded Petri dish and examined on a 40x-power dissecting microscope for the presence or ab-



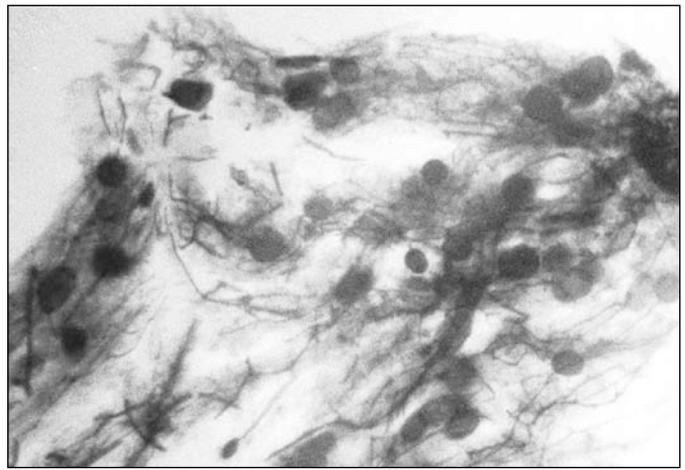
sence of mycorrhizal structure using the gridline method and examination of 100 grid intersections.

For each pot and treatment soil samples of 100 grams were sent to A&L West-

ern Agricultural Laboratory in Portland, Oregon, for determination of percentage of organic matter. For AF and AFM treatments, soil was separated from the roots of each plant using a fine mesh screen.



Inoculation with glomus spores (seen here at 80x magnification) helps form the mycorrhizal relationship with fescue.



Dark-stained areas within roots are colonized by mycorrhizal fungi (80x magnification).



This mycorrhizal inoculated tall fescue crop is an effective soil carbon sink.

Percentage of carbon for each sample was determined by multiplying the percent organic matter by 0.58. For pots that demonstrated mycorrhizal colonization, soil samples devoid of roots were sent to the University of California, Davis, for assessment of glomalin concentrations using the methods outlined by Rosier et al. ("Intraradical protein and glomalin as a tool for quantifying arbuscular mycorrhizal root colonization," *Pedobiologia*, vol. 52 [2008], pages 41-50).

RESULTS

The mycorrhizal inoculation (AFM) treatment significantly increased carbon in the tall fescue grown soils ($p < 0.001$; see page 17). Percentage of soil carbon

content was 1.66 for AFM compared to 0.90 for AF and 0.88 for A treatments, respectively. Soil carbon content was not significantly different when comparing AF and A treatments.

AFM pots had significantly higher levels of glomalin compared to AF pots ($p < 0.04$). The fescue root systems in AFM pots averaged 35.7 percent mycorrhizal colonization compared to 2.0 percent colonization in AF pots.

For individual pots there was a significant positive correlation between the level of mycorrhizal colonization and the level of soil carbon ($R^2 = 0.781$, $p < 0.001$). For individual pots there was also a significant positive correlation between the level of mycorrhizal colonization and

the level of soil glomalin ($R^2 = 0.575$, $p < 0.001$).

CONCLUSIONS

Under the simplified soil conditions of this study, inoculation of tall fescue with AM fungi nearly doubled soil carbon percentage in just one year, while no increase was seen with tall fescue that was not inoculated.

Glomalin production was likely a contributing factor to the increase in soil carbon, as glomalin concentrations increased with increased mycorrhizal colonization. The strongest effects were seen between 0 and 30 percent colonization; colonization rates higher than 30 percent produced only small additional increases in both soil carbon and glomalin.

This study indicates soils and perennial grasses are a potentially powerful sink for accumulating carbon in soil organic matter when mycorrhizal fungi are present. Activities such as introducing mycorrhizal fungi and corresponding glomalin production into soils where they have been eliminated can be an important mechanism to facilitate rapid carbon movement into soils.

Mike Amaranthus is an associate professor (adjunct) at Oregon State University and president of Mycorrhizal Applications, Inc; Dave Perry is a professor at Oregon State University; Zack Amaranthus is a biological technician at Mycorrhizal Applications; and Jeff Anderson is field projects coordinator at Mycorrhizal Applications, Inc. For more information, visit www.mycorrhizae.com.