Soil Microbiology FAQ's

David A. Zuberer

Over the years in my position as a soil microbiologist at Texas A&M University working with beneficial soil microbes and teaching soil microbiology to a very diverse audience of students I have been approached by many people with countless questions regarding the nature of soil microbes and their normal functions in soils, both cropped and uncropped, including turfgrass soils. The questions have been as diverse as the audiences with whom I have had the pleasure of addressing. Nevertheless, certain questions seem to come up over and over again.

In this brief article I will pose some of these questions and try to answer them against the framework of what is currently known about the functions of microbes in soils and what factors govern their activities.

FAQ #1 FAQ #2 FAQ #3 FAQ #4 FAQ #5 FAQ #6

FAQ 1 . . .

What kinds of microbes are found in a typical field or turfgrass soil and how abundant are they?

Normal, fertile soils teem with soil microbes. In fact, there may be hundreds of millions to billions of microbes in a single gram [about 4 hundredths of a pound and about the size of a navy bean in volume)(See Table 1)]. The most numerous microbes in soil are the **bacteria** (unicellular cells lacking a true nucleus) followed in decreasing numerical order by the **actinomycetes** (a specialized group of bacteria which contains many members that produce valuable antibiotics), the **fungi** (singular: fungus) which produce long, slender filaments nicely adapted for exploiting the three-dimensional pore network of the soil, soil **algae** and **cyanobacteria** ("blue-green "algae") (photosynthetic microbes which can add small amounts of carbon to soil and which also can be a nuisance in turfgrass golf greens) and soil **protozoa** (unicellular soil organisms that decompose organic materials as well as consume large numbers of bacteria).

Table 1. Numbers of Microbes in Soil

Microbial Group	No./Gram of soil
Bacteria	100,000,000 - 1,000,000,000
Fungi	100,000 - 1,000,000
Algae and Cyanobacteria	1000 - 1,000,000
Protozoa	1000 - 100,000

*Sylvia, Fuhrmann, Hartel and Zuberer, 1998.

Not only are the numbers of soil microbes generally very large, their combined mass (i.e. the soil microbial biomass) is also usually quite substantial. It can range from several hundred to thousands of pounds per acre of soil (Table 2).

Table 2. Microbial Biomass in typical fertile soils

Microbial Group	Wet wt. (lbs/ac)	Lbs/1000'sq ft.**
Bacteria	300-3,000	12
Actinomycetes	300-3,000	17
Fungi	500-5,000	35
Protozoa	50-200	8
Algae	10-1,500	3

** Data from Nelson, 1997b.

In addition to the microbes, there are numerous species of soil animals that inhabit soils. These include nematodes (microscopic roundworms which are generally beneficial but some of which are plant parasites of agricultural crops and turfgrasses), microarthropods (mites, springtails, etc.) and larger animals such as the earthworms, burrowing insects, etc. These larger organisms can exert beneficial effects through improved soil structure and improved aeration and drainage due to their channeling activities in the soil. Soil microbes are important for soil structure also but their effect is more subtle. Soil microbes produce lots of gummy substances (polysaccharides, mucilages, etc.) that help to cement soil aggregates. This cement makes aggregates less likely to crumble when exposed to water. Fungal filaments, called hyphae, also stabilize soil structure because these threadlike structures ramify throughout the soil literally surrounding particles and aggregates like a hairnet The fungi can be thought of as the "threads" of the soil fabric. It must be stressed that microbes generally exert little influence on changing the actual physical structure of the soil. That's the job of the larger "earthmovers".

Thus we see that a normal soil contains enormous numbers of microbes and substantial quantities of microbial biomass. This translates to an enormous **potential** for microbial activity when soil conditions (available carbon sources, moisture, aeration, temperature, pH, available inorganic nutrients such as nitrogen) are favorable. I stress potential for activity because under normal situations, the microbial population as a whole does not receive a constant supply of readily available substrates to sustain prolonged high rates of growth.

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FAQ 2 . . .
What beneficial processes do soil microbes carry out?

In addition to their role in cementing soil aggregates mentioned above, soil microbes are of paramount importance in cycling nutrients such as carbon (C), nitrogen (N), phosphorus (P), and sulfur (S). Not only do they control the forms of these elements [e.g. specialized soil bacteria convert ammonium N (NH_4^+) to nitrate N (NO_3^-)], they can regulate the quantities of N available to plants. This is especially critical in systems relying on organic fertilizers. **It is only through the actions of soil microbes that the nutrients in organic fertilizers are liberated for plants and use by other microbes.** Soil microbiologists call this process **mineralization** [the conversion of organic complexes of the elements to their inorganic forms, e.g., conversion of proteins to carbon dioxide (CO_2) ammonium (NH_4^+) and sulfate ($SO_4^=$)]. It is perhaps the single-most important function of soil microbes as it recycles nutrients tied up in organic materials back into forms useable by plants and other microbes. In fact, the so-called Principle of Microbial Infallibility (popularized by Dr. Martin Alexander of Cornell University) states that for every naturally occurring organic compound there is a microbe or enzyme system that can degrade it. Note that this applies to naturally occurring compounds. It is obvious that some our persistent pesticides did not conform to this principle and even some naturally occurring compounds are fairly resistant to microbial attack. It is through the process of mineralization that crop residues, grass clippings, leaves, organic wastes, etc., are decomposed and converted to forms useable for plant growth as well as converted to stable soil organic matter called **humus**. Herein lies another important role for the larger soil animals like earthworms. The large organisms function as **grinders** in that they reduce the particle size of organic residues making them more accessible and decomposable by the soil microbes. The soil microbial population also further decomposes the waste products of the larger animals. Thus, the activities of different groups of soil organisms are linked in complex "food webs".

One beneficial process carried out exclusively by soil microbes is called **nitrogen fixation**, the capture of inert N_2 gas (dinitrogen) from the air for incorporation into the bodies of microbial cells. In one well-known form of this process, symbiotic nitrogen fixation, soil bacteria such as Rhizobium and Bradyrhizobium actually inhabit specialized structures on the roots of leguminous plants (soybeans, cowpeas, beans clovers, etc.) where they fix substantial quantities of nitrogen that becomes available to the host plant. Unfortunately, the root nodule system is not found in the grasses so we cannot rely on it for "free" nitrogen. Nevertheless, free-living (nonsymbiotic) nitrogen-fixing bacteria do associate with roots of grasses where they fix small quantities of nitrogen using carbon compounds (root exudates, sloughed root cells, etc.) produced from the roots as energy sources to drive the energy-expensive nitrogen-fixing enzyme system. Another factor limiting the utility of free-living N_2 fixers is the fact that they will not fix N_2 when exposed to even very low levels of fertilizer nitrogen. Thus in fertile turfgrass soils this process is of limited importance whereas in unfertilized prairie soils the 10 to 25 pounds of N fixed per acre per year is ecologically relevant.

Another benefit of soil microbes is their ability to degrade pest control chemicals and other hazardous materials reaching the soil. Thus through the actions of the soil microflora, pesticides may be degraded or rendered nontoxic lowering their potential to cause environmental problems such as ground and surface water contamination. Of course, there is a "downside" to this microbial capability. In some instances, soil microbes have been shown to degrade soil-applied pesticides so rapidly as to reduce the ability of the chemicals to control the target pests. This phenomenon is known as **enhanced degradation** and usually results from repeated applications of a chemical to the soil. One way around this problem is to vary the use of pest control chemicals.

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FAQ 3 . . .

What factors control the rates of growth and activities of soil microbes?

This is an excellent question because an understanding of what it takes to support the growth and activity of soil microbes enables one to make decisions about soil management. In general, microbes need what all living things need to prosper: air (oxygen), water, food and a suitable habitat to live in (Table 3.).

Table 3. Principle environmental factors affecting soil microbes

- **Organic carbon** - grass clippings, crop residues, organic wastes, etc.
- **Moisture** - 50-60% of water holding capacity
- **Aeration** - balance of air and water filled pores
- **pH** - near neutral (pH 6.0-8.0)
- **Temperature** - 10 - 40^{o}C
- **Inorganic nutrients** - adequate N,P,K,S, etc., and trace metals

Interestingly, some soil bacteria (the anaerobes) do not even need air to grow and some are "poisoned" by exposure to oxygen. Generally, soil microbes grow best in soils of near neutral pH (7.0) having adequate supplies of inorganic nutrients (N and P, etc.), a balance of air- and water-filled **pore**

space (about 50-60% of water holding capacity) and abundant organic substrates (carbon and energy sources). When any one of these parameter gets too far beyond the normal range some segment of the population will likely be stressed. For example, aerobic (oxygen requiring) bacteria will be at a disadvantage when a soil becomes waterlogged and available O_2 is depleted through respiration of roots, microbes and soil animals. Conversely, anaerobic organisms may predominate leading to unique problems such as the formation of "black layer" caused, at least in part, by the anaerobic sulfate-reducing bacteria. Similarly, if soils become too acidic (down to pH 4 or 5) bacteria and actinomycetes usually decline and fungi assume a more dominant position. Except at cool and warm temperature extremes, the soil microbial population is usually not severely stressed. Most soil microbes grow best at temperatures between 15-30^o Celsius (about 60 to 85^{o}F) and their growth rates increase with increasing temperature up to a point. This is why it is harder to maintain soil organic matter in warm climates. Interestingly, some cold-loving microbes (called cryophiles) can actually grow and cause disease under blankets of snow cover. Such is the case with the so-called snow molds which can damage turfgrasses extensively during winter months. The opposite extreme is found in thermophilic microbes ("heat lovers") that thrive in composts reaching temperatures as high as 65^o C (150^o F). It is the biological heating of composts that actually reduces levels of pathogenic microbes, weed seeds and insects during the composting process.

Without a doubt, the **most important limiting factor for microbial growth in soil (assuming moisture is adequate) is the abundance of available organic carbon sources**. The vast majority of soil microbes require organic carbon compounds (these are called organotrophs) to oxidize for energy and to build the organic constituents of their cell bodies. Only a few types of soil bacteria get their carbon from CO_2 (autotrophs) and they contribute little to the overall organic matter content of a soil with the possible exception of the cyanobacteria on the surface of closely mown turfs where they may accumulate as dark slippery films. Organic inputs in turfgrass soils come mainly from the grasses themselves in the form of root exudates, lysed root cells, decomposing roots and any clippings returned to the soil. Of course, organic amendments may contribute some useable carbon as well but bear in mind that amendments such as compost, which is essentially microbially decomposed organic materials, do not contain high levels of readily available carbon. Rather, they provide slowly useable substrates and contribute directly to the soil organic matter pool. Also, as a general "rule of thumb" about one third of the organic carbon added to temperate soils remains in the soil as humus and microbial biomass whereas about two thirds of this carbon is returned to the atmosphere as CO_2 through microbial respiration. The microbial decomposition of grass clippings is the basis of the "Don't Bag It" programs of lawn maintenance which rely heavily on mulching mowers and the subsequent decomposition of clippings in the soil.

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FAQ 4 . . .
What can we do to increase microbial activity in soil?

Frequently turf managers ask what can be done to increase microbial activity in soil. No doubt this stems from a desire to capitalize on the known benefits attributed to the soil microflora. This question can also be turned around on the person asking it, i.e., Why do you want to increase microbial activity? Another way of phrasing this issue is “Can there be too much of a good thing"? Remember, increasing microbial activity increases organic matter decomposition, which can be good or bad. It might also be clear at this point that FAQ's #2 and #3 bear strongly on this question. The short answer to this question is relatively straightforward. To increase microbial activity in a soil one must make the environment optimal, or at least more favorable, in terms of aeration, moisture, and pH, and above all provide the organic substrates needed to fuel the population. It has been known for more than a century that the abundance of microbes in soil is directly proportional to the organic matter content. Thus soils receiving large amounts of organic residues support a larger microbial population. Generally there is an explosion in microbial numbers following the addition of available substrates. However, as the substrates are consumed microbial tissues are formed and CO_2 is given off so there is a loss of carbon from the soil with some storage in microbial biomass. Microbial cells themselves become food for other microbes and they too are decomposed through microbial activities. Eventually, microbial activity returns to a low level when substrates have been consumed or converted to compounds that are difficult to degrade that end up in the humus fraction. Thus we see that the increase in activity is transient. The normal state of affairs in soils not receiving large amounts of carbon on a regular basis is a microbial population subsisting on limited resources and metabolizing only very slowly. To effectively increase organic matter content in soil we must add more carbon than the microbes can decompose over a season. Regrettably, adding small amounts of organic materials like molasses to soils cannot do this. Soil microbes quickly use up substrates like these and little if any lasting effects are observed.

Another factor of great importance for decomposition of carbon in soil is the level of available nitrogen. When large amounts of available carbon are added to soils low in N, nitrogen becomes tied up, or **immobilized**, in the cells of the degraders. The net effect here is to induce nitrogen deficiency for plant growth due to swamping the system with available carbon. Careful attention should be paid to the **carbon to nitrogen** (C/N) ratio of organic materials added to soils for this reason.

Probably the most significant thing a turfgrass manger can do to sustain soil microbial populations is to maintain a vigorous, healthy turf. We know that grasslands are excellent microbial habitats and they can accumulate substantial microbial biomass. The same is true of well-managed turfgrass environments.

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FAQ 5 . . .

Do inorganic fertilizers and other chemical inputs harm the soil microbial population?

Frequently we see statements in the lay literature about chemical fertilizers killing soil microbes or, worse yet, statements indicating these management inputs "sterilize" the soil. Statements such as these should be viewed with much skepticism! Remember that as we learned in FAQ #1, the soil can contain tons of microbes. Short of incineration its hard to imagine a stress in a soil that would lead to complete extermination of the microbial populations. It is true that some inputs, e.g., anhydrous ammonia, cause reductions in microbial numbers in the immediate vicinity of the application. After all, ammonia is a toxic gas. However, it quickly equilibrates with the soil solution in the form of ammonium ions and the toxicity subsides. Certain pesticides have been shown to cause similar transient reductions in selected microbial population. But remember, in some cases the microbes simply view these chemicals as food and degrade them fairly quickly.

Organic fertilizers circumvent the criticisms leveled at "synthetic" fertilizers but it should not be forgotten that plants take up nitrogen in the form of ammonium (NH_4^+) or nitrate (NO_3^-) ions regardless of whether it was mineralized from an organic source or applied as in inorganic fertilizer like ammonium nitrate. An advantage of using organics, where practical, is that nutrients are liberated slowly as the microbes mineralize the organic materials. Thus there is low risk for fertilizer burn on plants and less risk for environmental problems due to runoff and leaching. Another potentially negative effect of long-term use of ammonia-based fertilizers is soil acidification due to ammonia oxidation by the nitrifying bacteria. Soil pH can drop below 5.0 after prolonged use of ammonia-based fertilizers and this can cause marked reductions in populations of bacteria and actinomycetes and simultaneous increases in the relative abundance of fungi. Such changes might favor the development of certain fungal plant pathogens. On the other hand, the potato scab disease is reduced by the low pH because the actinomycete which causes it is eliminated. These changes are easily reversed with applications of lime to the soil. Thus we see qualitative changes in the soil populations due to some management inputs but this is a long way from "sterilizing" or "killing" the soil.

With the advent of high-sand golf greens questions have arisen about the need for applying microbes during green construction and thereafter. Sand because of its lack of organic matter supports little microbial growth. However, when mixed with peats, composted rice hulls or other organic amendments it gains the microbial populations associated with those materials. Turfgrasses established from vegetative sprigs also bring their root-associated microbes with them! Once the turfgrass begins growing in the rooting medium of the green, microbes already present will colonize roots and the mechanics of soil organic matter formation will commence. A reasonable practice would be to add a small amount of normal pathogen-free soil to the greens mix as an inoculum. Thus far, there is little scientific evidence indicating the need to inoculate golf greens with selected microorganisms. The newly constructed green does afford us the possibility of customizing the soil population to some extent. Once we know what we want in these mixes it may be easier to add them "up front" than to add them into an established population already adapted to the prevailing conditions of a particular soil. As our knowledge of soil microbial biodiversity and the factors that control it increases we may find ways of tailoring microbial populations in given environments. At this point, we are limited in what we can do to this effect.

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FAQ 6 . . .

Why are biological products more variable in producing desired results?

Considerable research has been done on applying various microbes as inoculants for various purposes including their use as agents to control plant diseases, (including turfgrass pathogens; Nelson, 1997a), to stimulate plant growth (the so-called plant-growth-promoting rhizobacteria; PGPR) and more recently their use in various forms of bioremediation processes. Perhaps the most outstanding example of beneficial use of a soil bacterium is the practice of inoculating legumes with bacteria such as Rhizobium and Bradyrhizobium. Some crops are nearly self-sufficient in meeting their nitrogen requirements through this process. The process is so successful because the plant essentially selects the bacterium and builds a habitat, the root nodule, where conditions for nitrogen fixation are optimized. However, even with this remarkable symbiosis there are failures for one reason or another. Thus one of the nagging problems of using organisms as inoculants is the tendency for erratic control of pests or failure to observe any benefit from inoculation. Reasons for inconsistencies in response to inoculation can be manifold. What are some biological reasons for the failure of these types of products? There are many reasons why introduced bacteria do not become established when added to the soil in very low numbers. Some biological factors are listed in Table 4. Here we see a number of problems that an introduced microbe must overcome in order to establish itself among the normal population. These include inhibition by toxins, predation by other soil microbes such as the protozoa and a bacterium called *Bdellovibrio*, lysis by viruses called bacteriophages, and a simple inability to compete with the native organisms.

Table 4. Some biotic factors responsible for the elimination of introduced microbes:

- Microbially produced toxins
- Predatory protozoa
- Lysis by bacteriophage (bacterial viruses)
- Lysis by *Bdellovibrio bacteriovorus*
- Lysis by microbial enzymes
- Inability of introduced microbe to compete

Compounding our problems with introducing microbes to the soil is the fact that soil environmental factors (Table 5.) often contribute to the demise of added cells. For example high or low soil pH, toxic concentrations of metals, extreme temperatures, etc., can cause failures in establishment of introduced microbes.

Table 5. Some abiotic factors responsible for the elimination of introduced microbes:

- High or low pH
- High concentrations of Mn, Al, etc.
- Extreme heat or cold
- Many others

It is well to recall that each soil has an indigenous microbial population that is selected by the prevailing biotic and abiotic factors unique to that soil. Typically it is difficult to add or displace microorganisms to or from a system in such an equilibrium. An axiom of microbial ecology often referred to as **Beijerinck's Rule** (Beijerinck was a Dutch microbiologist who is often considered the "Father" of microbial ecology) states that "Everything (microbes) is everywhere and the milieu (i.e. the environment) selects". Thus each soil is endowed with a stable community of microbes uniquely selected by and adapted to the prevailing physical, chemical, and biological conditions of that soil. Minor perturbations have little effect on this balance.

From the above discussion, one can see that there are many factors, both biotic and abiotic, that can come together to foil our attempts to use beneficial microbes in practical applications. It is because of these inconsistencies that biological alternatives are often met with reluctance by users. There is a greater comfort factor in using a chemical formulation that delivers more consistent results when applied as directed. However, as research progresses and we gain a clearer understanding of the characteristics that make an organisms successful in the soil or rhizosphere environment it is likely that we will see the development of useful microbial products for a number of purposes including increasing plant growth, protecting crops from disease, organisms for use in bioremediation or for enhancing the cleanup of pesticides in rinsates etc. However, one thing will be reasonably certain, those that come to the forefront will be based on sound biological principles and will be backed up by substantial research demonstrating the efficacy of the product in meeting the claims of the manufacturer. In the meantime a few pointers for testing new products should be considered (see Table 6). Testing new products is an expensive proposition. However, without well-designed, replicated field trials useful information about the effectiveness of a product cannot be developed. After all, the proof is in the performance of the product under normal user conditions whether it be for turfgrass management, agricultural production or some other specific application. Microbes can and do indeed accomplish wonderful things. However, our abilities to harness and successfully manipulate beneficial microbes remains a "work in progress".

Table 6. Testing Microbial Fertilizers and Soil Activators (Biostimulants)

- Testimonials should be viewed with skepticism. Ask to see original data.
- Test products in replicated plots with valid statistical designs
- Test products across multiple soil types
- Test products across locations, climate, etc.
- Minimally: test product in strips in fields and measure yields, turf performance, etc.

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Suggested readings:

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