NebGuide
Nebraska Extension
Research-Based Information That You Can Use
G2327 · Index: Crops, Soil Management
Issued May 2020

Soil Microbiology in Nebraska

Charles Wortmann, University of Nebraska–Lincoln (UNL) Extension soil management specialist

Javed Iqbal, UNL Extension nutrient management and water quality specialist

Bijesh Maharjan, UNL Extension soil and nutrient management specialist

Laila Puntel, UNL Extension soil fertility and precision agriculture specialist

Soil microbiology is a universal and essential component of agricultural soils. This NebGuide provides basic information of soil microbiology, microbial functions, microbial enhancement, and microbial assessment.

Soil organisms are essential for many soil processes and functions. The surface 6" of soil for typical agricultural land in Nebraska may contain 3 ton/acre of soil organisms, which is equal to about 0.3% of soil weight on a dry weight basis. These include soil macrofauna (nematodes, earthworms, beetles, mites, springtails, mice, etc.) and microbes such as bacteria, fungi (including mycorrhiza), viruses, and microscopic eukaryotes (including protozoa and algae). Archaea are an important microbial group, but are treated as bacteria in this paper due to similarities. This NebGuide is focused on soil microbes other than viruses.

A teaspoon of soil may contain more than a billion bacteria and archea plus yards of fungi mycelium, and thousands of other microbes. Soil organisms represent much diversity, consisting of more than 10,000 species with the estimated number of species increasing as speciation improves. The microbes form complex and dynamic communities, with the relative dominance of species and microbial groups responding rapidly to changing conditions. The biomass of microbial communities may on average turnover 1.5 times per year in Nebraska and contributes organic material to the soil. Microbial turnover provides an easily decomposed source of energy, carbon (C), and nutrients for new microbial growth.

Bacteria are free-living, single-cell organisms that can reproduce in as little as 30 minutes under ideal growth conditions, or survive for extended times in a suspended or dormant state. Bacteria are by far the most numerous and diverse group of soil microbes with thousands of species. Bacteria include actinomycetes, or actinobacteria, which are gram-positive bacteria and responsible for the earthy smell of soil. Actinomycetes are larger, but fewer than other bacteria, and may have similar biomass (t/ac) as other bacteria. Rhizobia are specialized bacteria that form nodules on the roots of legumes for the symbiotic fixation of atmospheric nitrogen (N). Some free-living bacteria (e.g. cyanobacteria, *Pseudomonas, Azospirillum, and Azotobacter*) cause non-symbiotic N fixation.

Fungi produce long strings of cells called hyphae, which form webs of mycelium. Arbuscular mycorrhiza are specialized fungi that colonize roots and produce hyphae, which extend 2 to 4 inches from roots to greatly increase contact with the soil for nutrient and water uptake. The hyphae of mycorrhiza aid in soil aggregation by entangling soil particles and producing glue-like glomalin. Fungi require a consistent source of food, but adapt to changes in soil conditions more readily than do bacteria.

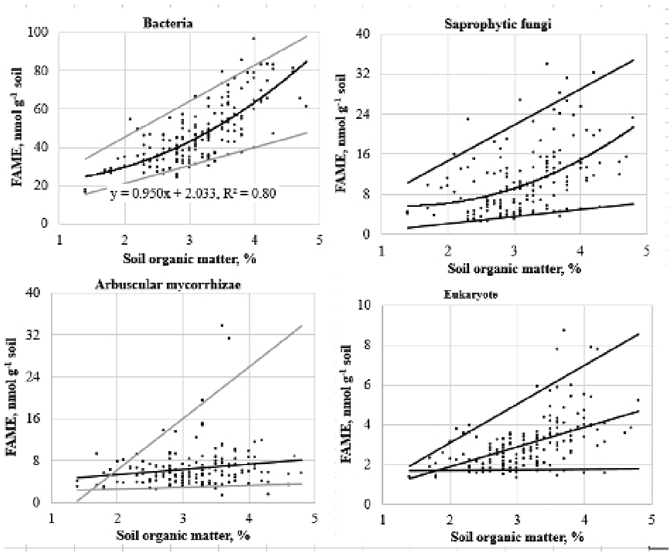

Figure 1. Distribution of microbial fatty acid methyl esters (FAME), indicators of microbial biomass, relative to SOM for organically managed cropland in eastern Nebraska with the upper and lower boundaries each determined from 5% of the samples.

Protozoa are much larger and more complex than bacteria. They are single-cell eukaryotes with nuclei that feed on other microbes and organic material and include groups such as flagellates and amoebas. Feeding by protozoa results in the release of ammonium-N for the supply of N to other microbes and plants. Nematodes are another type of multi-cellular soil microorganism that feeds on other microbes. Nematodes may require three days to three years to reproduce. Both protozoa and nematodes are aquatic and need adequate soil water for mobility.

Most soil microorganisms are saprophytic and therefore gain energy and nutrients from dead organic material. Soil microbial biomass and activity tend to increase with increased soil organic matter (SOM), which is related to the amount of organic material returned to the soil. Bacteria and saprophytic fungi biomass tends to be more with more soil organic matter (SOM), whereas eukaryotes and arbuscular mycorrhiza were found to be less affected by SOM levels in Nebraska soils (Figure 1). Soil microbes tend to be more active near roots, or in the rhizosphere, of plants due to root exudation of sugars and proteins. Management to increase SOM may be a feasible means to increase microbial activity, especially for sites where surface soil was lost, such as to erosion (NebGuide G2283).

Factors Affecting Microbial Biomass and Activity

Competition among soil microbes is great. The microbes are "starved" by insufficient resources and are dormant or in other states of quiescence much of the year. On average throughout the year, 90% of soil bacteria are inactive. Microbial communities dynamically adjust with microbial groups and species gaining or losing prominence with ongoing changes in factors affecting overall microbial activity. Some microbial species are adapted to soil extremes of temperature, water and air availability, pH, salinity, and the C:N ratio of organic materials.

Overall soil microbial activity approximately doubles with each 10°F increase in soil temperature to peak activity at about 85°F, above which activity eventually declines. Overall microbial activity is very low at < 30 and > 110° F, but some microbial species are adapted to the extremes such as with high microbial activities at 140–160° F during composting.

More soil water availability favors overall soil microbial activity until excess soil water limits soil air movement. Soil water at field capacity may be optimal for sandy soil, but microbial activity can be limited at field capacity with high soil water-filled pore space and little air movement for clayey soils and in poorly aggregated or compacted soil.

Microbes compete for energy, C, N, and other nutrients. Soil microbial activity increases with the addition of organic material to soil, especially with material of low C:N ratio, if other factors affecting microbial activity are favorable. The ratio of fungal to bacterial activity is expected to increase as the C:N ratio of applied organic material increases.

Overall soil microbial activity is expected to be very low at < 4.5 soil pH, partly due to reduced plant growth and effects of exchangeable aluminum. The ratio of fungal to bacterial activity decreases greatly as soil pH declines from 8 to 4.5. The optimal pH range for rhizobia varies with crop and the associated rhizobia species, but biological fixation of atmospheric N is commonly optimized at soil pH of 6.0 to 7.2.

Soil salinity affects microbial activity through reduced plant growth, drawing water from cells, and toxic levels of some ions. Salinity-tolerant microbes become more prominent under saline conditions.

Soil Microbial Processes and Functions

Organic material decomposition. Much organic material is added to the soil annually for highly productive agricultural land. For example, estimates of annual organic material dry weight contribution for a 240 bu/ac corn grain crop include about 2 t/ac of roots, 0.5 t/ac of root exudates, 6 t/ac of non-harvested aboveground crop and weed residue, 1.1 t/ac of soil microbial biomass turnover to a 4'

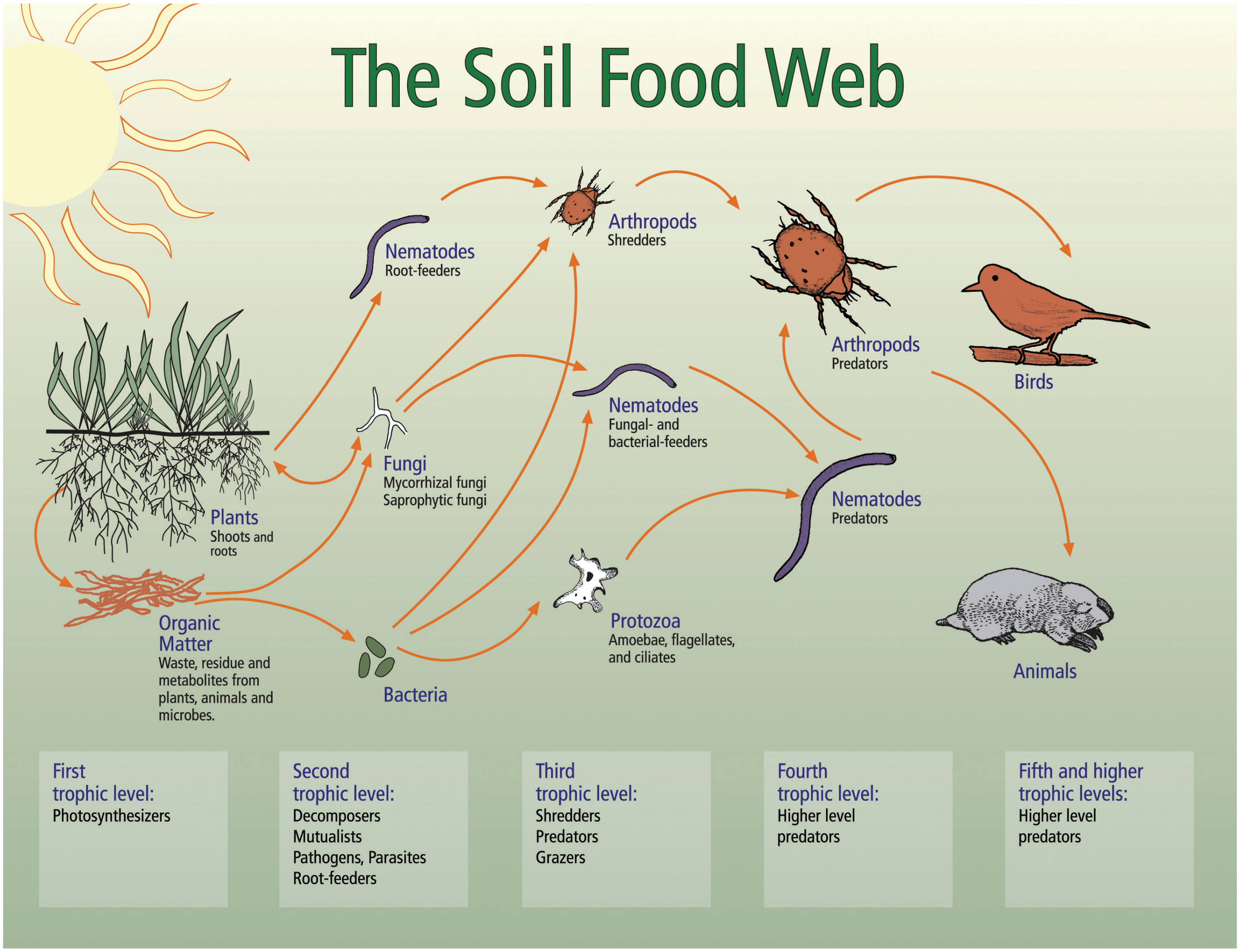

Figure 2. The bottom-up tropic cascade of the soil food web © USDA Natural Resources Conservation Service.

soil depth, maybe 1 t/ac of manure, maybe 1 t/ac of cover crop growth, and smaller quantities of urea and pesticides. The decomposition of about 11 t/ac/yr of organic material in the upper 4-6 inches of soil demonstrates the high level of soil microbial activity associated with highly productive land. Decomposition provides C, energy, and nutrients for new microbial cell growth. The approximate fate of applied organic C is 60–80% as emitted carbon dioxide (CO_2), 3–8% in living microbes, 3–8% in non-humic compounds, and 10–30% in humic compounds, the latter of which are very resistant to decomposition.

The addition of organic material (plant and animal residues, manure, other organic waste, and dead microbial biomass) to soil stimulates an upward trophic cascade referred to as the soil food web (Figure 2). Bacteria are the "first responders" to newly available organic material. Through enzymatic action, bacteria are the primary cause of decomposition of organic materials with low C:N ratio. Bacteria are 10–30% N by dry weight and use 20–30% of C mineralized from the decomposition of organic materials for cell growth. Bacteria quickly decompose proteins and sugars, leaving behind cellulosic materials and lignin. Fungi and actinomycetes account for much decomposition of relatively resistant organic materials of high C:N ratios, also through enzymatic activity. Fungi convert 40–55% of digested C to fungal cell C, and fungal biomass is < 10% N. Protozoa consume organic materials and bacteria. The feeding of protozoa and nematodes on microbes results in a release of ammonium-N. Turnover of microbial biomass is an important energy and nutrient source to bacteria due to the low C:N ratio and low lignin and cellulose contents of microbial biomass. Nematodes feed on organic materials, bacteria, fungi, protozoa, and other nematodes. Anthropods feed on nematodes, and birds and animals feed on anthropods. All die eventually providing an important energy and nutrient source throughout the food chain.

Nutrient cycling. Nutrient cycling involves the flux of nutrients between the various organic and inorganic nutrient pools in the soil, including plant uptake from various soil depths and the re-deposition of nutrients in organic material. The silt loam and silty clay loam soils of Nebraska may contain > 4,000 lb/ac phosphorus (P) in the top 4' of soil. Only a small fraction of this P becomes plant available during a growing season. Most is inorganic P that is made more available to plants by reaction with acidic products produced through microbial fermentation and by bacteria production of enzymes ('-ase' compounds such as phosphatase) for release of inorganic P and cellulase for organic P mineralization. Some mineral nutrients, e.g., iron and manganese, are made more or less available to plants, depending on the bacterial action. Turnover of nutrients in microbial biomass is important to maintaining nutrients in soil solution, while microbes may stimulate root growth for greater capacity for nutrient and water uptake.

Soil organic nutrient mineralization. Each 1% SOM for 6" soil depth contains about 1,000 lb N and 100 lb each of P, potassium (K), and sulfur (S) which would be worth about $588/ac if these were fertilizer nutrients (Table 1). Most SOM is very stable with very low rates of microbial decomposition. Soils of similar SOM can differ greatly in the nutrient mineralization potential depending on the amount of active relative to passive and stable SOM. For example, tropical soils with 2.5% SOM mineralized 1/3 as much N as a Nebraska soil with 2.5% SOM. For a similar amount of organic material returned to the soil, the rate of SOM accumulation will decrease with increasingly humid and warmer conditions.

Nitrogen dynamics. Nitrogen is assimilated by soil microbes, causing immobilization of N, but N is also released as ammonium by microbial decomposition of organic materials. The released ammonium-N is available for uptake by plants or microbes. Applied fertilizer-N may be taken up directly by plants, but much will go through a cycle, and maybe several cycles, of microbial conversion or assimilation and release before uptake by plants. Some ammonium goes through the microbial process of nitrification for conversion to nitrite by *Nitrosomonas* bacteria and nitrite conversion to nitrate by *Nitrobacter* bacteria. Denitrification and nitrous oxide production are also microbially dependent processes.

Symbiotic biological fixation of atmospheric N requires the enzyme nitrogenase, which is effective only under anaerobic conditions. Symbiotic N fixation with legumes occurs in nodules where nitrogenase is protected from oxygen. Non-symbiotic fixation of N from the air by free-living bacteria also involves nitrogenase that requires anaerobic conditions, which can be created by some specialized microbes.

Table 1. The nutrient content and fertilizer equivalent value of nutrients in soil organic matter assuming: 2 million pounds soil in top 6 inches; 1% organic matter = 10,000 lb organic C in top 6 inches; the soil C:N ratio = 10:1.

Nutrient	Amounts and value
Nitrogen	1,000 lb * $0.44/lb N = $440
Phosphorus	100 lb * $.46/lb P = $46
Potassium	100 lb * $0.32/lb K = $32
Sulfur	100 lb * $0.50/lb S = $50
Carbon	10,000 lb or 5 t * $4/t = $20
Value of nutrients/acre/1% SOM = $588	

Modified from ohioline.osu.edu/factsheet/SAG

Contaminant degradation. The breakdown of contaminants, including pesticides, is an important microbial function. Such degradation reduces loss of contaminants to water bodies. It reduces carry-over of some herbicides, such as atrazine, to the next crop. The half-life of pesticides is expected to decrease as microbial populations become more adapted to using these as energy and nutrient sources. For example, atrazine may have a half-life of < 50 years in soils with a decades-long history of frequent application of atrazine compared to > 100 days in other soils lacking such an application history.

Hormones and antibiotics, along with their metabolic by-products, occur from natural microbial sources and from applied livestock and human organic materials. Antibiotics for health care have often been produced from soil isolates as some soil microbe species naturally produce antibiotics for enhanced competitiveness. The applied contaminants are subject to microbial degradation, but have the potential to drive increased microbial resistance and the development of more resistant disease strains.

Soil aggregation. Soil microbes produce complex sugars, proteins, and other compounds in mucilage that bind soil particles together for improved soil aggregation. Fungal hyphae form mycelial webs around and through soil aggregates to further stabilize aggregates. Mycorrhiza produce glue-like glomalin. Earthworm casts contribute to soil aggregation.

Other functions. Water is cleansed as it percolates through the soil and the underlying vadose zone before it reaches groundwater or seepage areas due to physical filtration and chemical reactions that may depend on microbial activity, such as pesticide or other contaminant degradation. Soil microbes have roles in pest control such as degradation of weed seeds and suppression of plant disease. Soil microbes can aid in plant tolerance to stress.

Enhancement of Soil Microbial Activity

Highly productive agricultural lands with large amounts of crop residue returned to the soil typically have high and dynamic soil microbial communities. A very high level of activity is not necessarily optimal for agronomic purposes. The loss of ground cover by crop residues may be more rapid than desired for reduced soil erosion and soil water evaporation. Microbial conversion of organic N and ammonia-based fertilizer N to leachable nitrate in a short period of time is undesirable both from environmental and agronomic perspectives. However, targeted enhancement of microbial activity or effectiveness may be desired and may be agronomically feasible. Most important to the enhancement of microbial activity is the optimization of factors addressed above that affect microbial activity. In addition, increased supply of organic material, inoculants or bio-stimulants, crop rotation, and increased duration of crop presence may enhance microbial activity.

Increased supply of organic material. The importance of SOM level to soil bacteria and saprophytic fungi biomass was addressed above (Figure 1). Management to increase SOM may be a feasible means to increase microbial activity, especially for sites where surface soil was lost, such as to erosion. Application of manure or other organic materials can stimulate microbial activity for improved aggregation of soil, at least for a short time, and thereby increase water infiltration and resistance to erosion, soil crusting, and yield depending on the previous soil conditions.

Tillage. Soil microbial biomass and activity is higher in the 0–2 inch soil depth compared with deeper depths. This stratification can be reduced with inversion plow tillage. The change in microbial biomass in the 0–12 inch soil depth at 2–3 years after one-time moldboard or miniboard plow tillage, compared with continuous no-till, were no change for bacteria including actinomycetes and saprophytic fungi, but a decrease in arbuscular mycorrhiza. The decreases in arbuscular mycorrhiza persisted in one of three fields for five years, but nutrient uptake and grain yield were not decreased. Overall, bacteria are more responsive and adaptive than fungi to soil disturbance.

Inoculants. The most common use of inoculants is the inoculation of legume seed with bacteria for symbiotic N fixation Fertilizer Recommendations for Soybean. This can result in increased yield, depending on the legume species and on past land use and management. In an analysis of results from 187 studies of soybean response to inoculation with *Bradyrhizobia* with fields of past soybean production in the U.S., the mean yield increase due to inoculation was 0.9 bu/ac with greater increases with soil pH > 6.8, late planting, and relatively low-yield situations. Soil or seed may also be inoculated with free-living bacteria that fix N non-symbiotically, such as cyanobacteria, *Azobacter*, and *Azospirillum*.

Inoculation with P solubilizing bacterial, e.g. *Bacillus*, and fungal microbes can be effective when well-matched with the right soil conditions and may be most effective for calcareous soils. Inoculation with mycorrhiza is of interest for increased uptake of soil P, other nutrients, and water. Inoculation with other bacteria or fungi, or microbial stimulants, may be done to promote plant growth or suppress plant pathogens. Inoculation to suppress plant pathogens may be through activation of plant defense such as to strengthen cell walls or release antibiotics.

Inoculation often fails to give positive results, maybe due to: (1) already adequate activity of the microbial group or species such as is common for *Rhizobia* or *Bradyrhizobia* for fields with a history of producing the associated legume crop; (2) failure to compete with other microbes for resources; (3) predatory protozoa, viruses, and microbial enzymes; and (3) extremes of soil pH or temperature. The quality of the product is another concern as such products are generally not regulated as are fertilizers and pesticides, with regulatory exceptions in some states. Yield increases due to the application of microbial inoculants and growth stimulants have been rare in Nebraska and the North Central U.S. as might be expected for already highly productive systems. Between 2010 and 2018, 17 soil microbial inoculation or growth stimulation products were evaluated in 76 on-farm trials conducted by the Nebraska On-Farm Research Network with yield increases for only 8% of the trials.

Careful targeting of microbial inoculates and stimulates to specific field conditions may improve effectiveness. However, product use instructions commonly do not address targeting. Current and future research on rhizosphere dynamics, microbial species differentiation, and product targeting is expected to lead to more success with such products. For example, inoculation of non-legumes with a non-symbiotic N-fixing bacteria may be more successful for cases of low soil N availability as the introduced microbes may better compete for needed resources with other microbes that are stimulated by high N availability. The same principle may apply for inoculants for increased P availability where overall microbial competition may be less with low soil P availability. The inoculant may also be specific to causes of low P availability such as a very P-depleted soil or P tie-up under calcareous conditions or under acid soil conditions.

Crop rotation and duration of crop presence. Increased yield is common for a crop rotated with another crop compared with monoculture. This increase may occasionally be due to, among other factors, an agronomically favorable enhancement in microbial activity; however, rotation does not always increase microbial biomass or diversity. Assuming similar soil water availability, the duration of crop growth is expected to affect the immediate microbial activity with more soil microbial activity expected with some perennial grasses than with annual crops, long- compared with short-duration crops, and double cropping compared with a single crop per year.

Assessing Microbial Communities and Activity

The ability to assess microbial community composition and activity has changed greatly over the decades. For example, in a study conducted in Nebraska in the 1940s, microbe counts per gram of soil were estimated by repeated dilutions until the microbial concentration was low enough to make counts feasible, and then back-calculating to determine numbers per gram. It later became feasible to measure the amounts or activities of a specific enzyme. Later, analysis for a suite of enzymes became feasible. Further research identified organic compounds such as fatty acid methyl esters (FAMEs) and phospholipid fatty acids (PLFA) specific to microbial groups allowing the study of soil microbial community structures. Advances in genomics and DNA analysis enabled better differentiation of microbial species and more in-depth study of soil microbiomes. The rate of scholarly publication about the soil microbiome has increased from about 1,000 during the 2000–2005 period to approximately 20,000 from 2015 to 2019.

Soil respiration and soil microbial activity are indicated by the CO_2 generated in a given time, such as during one day, from a freshly collected soil sample. The CO_2 generated from a dried and then re-wetted soil sample, however, is more indicative of microbial biomass than of activity as drying followed by abrupt re-wetting kills many microbes causing a flush of CO_2 release. High CO_2 generation and microbial activity are expected with near-ideal soil water, temperature, and aeration conditions coupled with an adequate supply of organic material and N at the time of sampling. Very high activity may be preferred if a high rate of nutrient cycling is desired while the duration of ground cover by crop residues such as for erosion and soil water evaporation control is expected to benefit from the relatively low activity.

Soil microbial community structures are often studied with FAME (Figure1) and PLFA analyses. These analyses give estimates of the amounts or activities of microbial groups (e.g., actinomycetes, other bacteria, saprophytic fungi, arbuscular mycorrhiza, and eukaryotes) for determination of microbial community profiles at the time of sampling the soil. The group ratios can be calculated, but only the saprophytic fungi:bacteria ratio has had much validation for edaphic, agronomic and environmental interpretations of the results.

Interpretations of such assessments are generally of little agronomic value. The assessments are most useful in making direct comparisons for evaluation of the effects of different practices, times of the year, or soils for better understanding of soil microbial dynamics. It is important to remember that the results represent the time and depth of soil sampling, and further inference is limited without additional sampling and analyses.

Summary

Soil microbiology is a universal and essential component of agricultural soils. This NebGuide provides basic information of soil microbiology, microbial functions, microbial enhancement, and microbial assessment. Soil microbial communities are very diverse and highly responsive to changed conditions. The relative abundance and activity of microbial species and types adjusts to changes in soil temperature, water and air availability, nutrient availability, and the type and amount of organic material present. Generally, soil microbial communities are 'well-fed' with highly productive cropland, although less so with crop residue harvest. Major functions of soil microbiology have been addressed including organic material decomposition, nutrient mineralization and cycling, contaminant degradation, soil aggregation, soil water cleansing, and others. The potential and possible means to the enhancement and assessment of soil microbial biomass and activity were addressed.

This publication has been peer reviewed.
Nebraska Extension publications are available online at http://extensionpubs.unl.edu/.