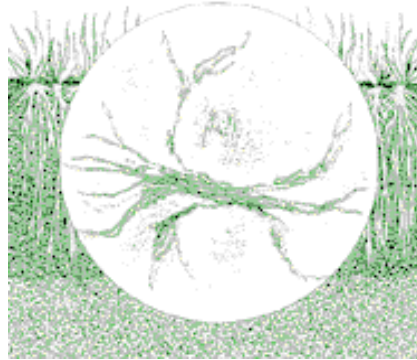


BENEFICIAL AND EFFECTIVE MICROORGANISMS



FOR A SUSTAINABLE AGRICULTURE AND ENVIRONMENT

Dr. Teruo Higa
Professor of Horticulture
University of the Ryukyus
Okinawa, Japan

and

Dr. James F. Parr
Soil Microbiologist
Agricultural Research Service
US. Department of Agriculture
Beltsville, Maryland, USA

International Nature Farming Research Center
Atami, Japan
1994

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FORWARD

In 1989, the National Research Council of the National Academy of Sciences issued a highly significant report on "Alternative Agriculture" which was defined as a system of food and fiber production that applies management skills and information to reduce costs, improve efficiency, and maintain production levels through such practices as crop rotations, proper integration of crops and livestock, nitrogen fixing legumes, integrated pest management, conservation tillage, and recycling of on-farm wastes as soil conditioner: and biofertilizers. The report encouraged the collective adoption of these practices by U.S. farmers as the best alternative to the continued and intensive use of chemical fertilizers and pesticides which have often impaired the quality of our soil, water, and food.

Again, in 1993 the National Academy of Sciences left no doubt as to these earlier concerns when the National Research Council released a report on "Pesticides in the Diets of Infants and Children" which concluded that people in this age group could be at considerable health risk from consumption of foods containing pesticide residues.

Both of these reports have raised considerable speculation about the future of our chemical-based agricultural production system. A growing consensus of consumers, environmentalists, legislators, and many farmers is that our current farming practices will have to change considerably to achieve a significant reduction in pesticide usage in U.S. agriculture. The ultimate goal of sustainable agriculture according to the National Research Council, and other sources as well, is to develop farming systems that are productive, profitable, energy conserving, environmentally sound, conserving of natural resources, and that ensure food

safety and quality. Consequently, the leading question that U.S. farmers are asking is, "How can I make these changes, reduce my chemical inputs, and achieve an acceptable level of economic and environmental sustainability?"

A successful transition from chemical-based farming systems to a more sustainable agriculture will depend largely on what farmers do to improve and maintain the quality of their agricultural soils. Indeed, soil quality is the "key" to a sustainable agriculture. Not surprisingly, the alternative agricultural practices advocated by the National Research Council are mainly those that can improve and maintain soil quality. Experience has shown that the transition from conventional agriculture to nature farming or organic farming can involve certain risks, such as initially lower yields and increased pest problems. Once through the transition period, which might take several years, most farmers find their new farming systems to be stable, productive, manageable and profitable without pesticides.

Dr. Teruo Higa, Professor of Horticulture, University of the Ryukyus, Okinawa, Japan has conducted pioneering work in advancing the concept of "Effective Microorganisms" (EM). He has developed microbial inoculants that have been shown to improve soil quality, crop growth and yield and have gained attention worldwide. As farmer: seek to change from chemical-based, conventional farming systems to more sustainable kinds of agriculture they will need to utilize the most effective means available if they are to be successful. Certainly, this includes the aforementioned alternative agricultural practices recommended by the National Research Council. We view EM technology as a potentially valuable tool that can help farmer: develop farming systems that are economically, environmentally, and socially sustainable.

Dr. James F. Parr
Agricultural Research Service
U.S. Department of Agriculture
Beltsville, Maryland, USA

INTRODUCTION

The uniqueness of microorganisms and their often unpredictable nature and biosynthetic capabilities, given a specific set of environmental and cultural conditions, has made them likely candidates for solving particularly difficult problems in the life sciences and other fields as well. The various ways in which microorganisms have been used over the past 50 years to advance medical technology, human and animal health, food processing, food safety and quality, genetic engineering, environmental protection, agricultural biotechnology, and more effective treatment of agricultural and municipal wastes provide a most impressive record of achievement. Many of these technological advances would not have been possible using straightforward chemical and physical engineering methods, or if they were, they would not have been practically or economically feasible.

Nevertheless, while microbial technologies have been applied to various agricultural and environmental problems with considerable success in recent years, they have not been widely accepted by the scientific community because it is often difficult to consistently reproduce their beneficial effects. Microorganisms are effective only when they are presented with suitable and optimum conditions for metabolizing their substrates including available water, oxygen (depending on whether the microorganisms are obligate aerobes or facultative anaerobes), pH and temperature of their environment. Meanwhile, the various types of microbial cultures and inoculants available in the market today have rapidly increased because of these new technologies. Significant achievements are being made in systems where technical guidance is coordinated with the marketing of microbial products. Since microorganisms are useful in eliminating problems associated with the use of chemical fertilizers and pesticides, they are now widely applied in nature farming and organic agriculture (Higa, 1991; Parr et al 1994).

Environmental pollution, caused by excessive soil erosion and the associated transport of sediment, chemical fertilizers and pesticides to surface and groundwater, and improper treatment of human and animal wastes has caused serious environmental and social problems throughout the world. Often engineers have attempted to solve these problems using established chemical and physical methods. However, they have usually found that such problems cannot be solved without using microbial methods and technologies in coordination with agricultural production (Reganold et al., 1990; Parr and Hornick, 1992a).

For many years, soil microbiologists and microbial ecologists have tended to differentiate soil microorganisms as beneficial or harmful according to their functions and how they affect soil quality, plant growth and yield, and plant health. As shown in Table 1, beneficial microorganisms are those that can fix atmospheric nitrogen, decompose organic wastes and residues, detoxify pesticides, suppress plant diseases and soil-borne pathogens, enhance nutrient cycling, and produce bioactive compounds such as vitamins, hormones and enzymes that stimulate

plant growth. Harmful microorganisms are those that can induce plant diseases, stimulate soil-borne pathogens, immobilize nutrients, and produce toxic and putrescent substances that adversely affect plant growth and health.

A more specific classification of beneficial microorganisms has been suggested by Higa (1991; 1994; 1995) which he refer to as "Effective Microorganisms" or EM. This report presents some new perspectives on the role and application of beneficial microorganism, including EM, as microbial inoculants for shifting the soil microbiological equilibrium in ways that can improve soil quality, enhance crop production and protection, conserve natural resources, and ultimately create a more sustainable agriculture and environment The report also discusses strategies on how beneficial microorganisms, including EM. can be more effective after inoculation into soils.

THE CONCEPT OF EFTECTIVE MICROORGANISMS: THEIR ROLE AND APPLICATION

The concept of effective microorganisms (EM) was developed by Professor Teruo Higa, University of the Ryukyus, Okinawa, Japan (Higa, 1991; Higa and Wididana, 1991a). EM consists of mixed cultures of beneficial an naturally-occurring microorganisms that can be applied as inoculants to increase the microbial diversity of soils and plant. Research has shown that the inoculation of EM cultures to the soil/plant ecosystem can improve soil quality, soil health, and the growth, yield, and quality of crops. EM contains selected species of microorganisms including predominant populations of lactic acid bacteria and yeasts and smaller numbers of photosynthetic bacteria, actinomycetes and other types of organisms. All of these are mutually compatible with one another and can coexist in liquid culture.

EM is not a substitute for other management practices. It is, however, an added dimension for optimizing our best soil and crop management practices such as crop rotations, use of organic amendments, conservation tillage, crop residue recycling, and biocontrol of pests. If used properly, EM can significantly enhance the beneficial effects of these practices (Higa and Wididana, 1991b).

Throughout the discussion which follows, we will use the term "beneficial microorganisms" In a general way to designate a large group of often unknown or ill-defined microorganisms that interact favorably in soils and with plants to render beneficial effects which are sometimes difficult to predict. We use the term "effective microorganisms" or EM to denote specific mixed cultures of known, beneficial microorganisms that are being used effectively as microbial inoculants.

UTILIZATION OF BENEFICIAL MICROORGANISMS IN AGRICULTURE

What Constitutes an Ideal Agricultural System?

Conceptual design is important in developing new technologies for utilizing beneficial and effective microorganisms for a more sustainable agriculture and environment. The basis of a conceptual design is imply to first conceive an ideal or model and then to devise a strategy and method for achieving the reality. However it is necessary to carefully coordinate the materials, the environment, and the technologies constituting the method. Moreover one should adopt a philosophical attitude in applying microbial technologies to agricultural production and conservation systems.

There are many opinions on what an ideal agricultural system is. Many would agree that such an idealized system should produce food on a long-term sustainable basis. Many would also insist that it should maintain and improve human health, be economically and spiritually beneficial to both producers and consumers, actively preserve and protect the environment, be self-contained and regenerative, and produce enough food for an increasing world population (Higa, 1991).

Efficient Utilization and Recycling of Energy

Agricultural production begins with the process of photosynthesis by green plants which requires solar energy, water, and carbon dioxide. It occurs through the plants ability to utilize solar energy in "fixing" atmospheric carbon into carbohydrates. The energy obtained is used for further biosynthesis in the plant, including essential amino acids and proteins. The materials used for agricultural production are abundantly available with little initial cost. However, when it is observed as an economic activity, the fixation of carbon by photosynthesis has an extremely low efficiency mainly because of the low utilization rate of solar energy by green plants. Therefore, an integrated approach is needed to increase the level of solar energy utilization by plants so that greater amounts of atmospheric carbon can be converted into useful substrates (Higa and Wididana, 1991a).

Although the potential utilization rate of solar energy by plants has been estimated theoretically at between 10 and 20%, the actual utilization rate is less than 1%. Even the utilization rate of C4 plants, such as sugar cane whose photosynthetic efficiency is very high, barely exceeds 6 or 7% during the maximum growth period. The utilization rate is normally less than 3% even for optimum crop yields.

Past studies have shown that photosynthetic efficiency of the chloroplasts of host crop plants cannot be increased much further; this means that their biomass production has reached a maximum level. Therefore, the best opportunity for

increasing biomass production is to somehow utilize the visible light, which chloroplasts cannot presently use, and the infrared radiation; together, these comprise about 80% of the total solar energy. Also, we must explore ways of recycling organic energy contained in plant and animal residues through direct utilization of organic molecules by plants (Higa and Wididana, 1991a).

Thus, it is difficult to exceed the existing limits of crop production unless the efficiency of utilizing solar energy is increased, and the energy contained in existing organic molecules (amino acids, peptides and carbohydrates) is utilized either directly or indirectly by the plant. This approach could help to solve the problems of environmental pollution and degradation caused by the misuse and excessive application of chemical fertilizers and pesticides to soils. Therefore, new technologies that can enhance the economic-viability of farming systems with little or no use of chemical fertilizers and pesticides are urgently needed and should be a high priority of agricultural research both now and in the immediate future (National Academy of Sciences, 1989; 1993).

Preservation of Natural Resources and the Environment

The excessive erosion of topsoil from farmland caused by intensive tillage and row-crop production has caused extensive soil degradation and also contributed to the pollution of both surface and groundwater. Organic wastes from animal production, agricultural and marine processing industries, and municipal wastes (e.i., sewage and garbage), have become major sources of environmental pollution in both developed and developing countries. Furthermore, the production of methane from paddy fields and ruminant animals and of carbon dioxide from the burning of fossil fuels, land clearing and organic matter decomposition have been linked to global warming as "greenhouse gases" (Parr and Hornick, 1992b).

Chemical-based, conventional systems of agricultural production have created many sources of pollution that, either directly or indirectly, can contribute to degradation of the environment and destruction of our natural resource base. This situation would change significantly if these pollutants could be utilized in agricultural production as sources of energy.

Therefore, it is necessary that future agricultural technologies be compatible with the global ecosystem and with solutions to such problems in areas different from those of conventional agricultural technologies. An area that appears to hold the greatest promise for technological advances in crop production, crop protection, and natural resource conservation is that of beneficial and effective microorganisms applied as soil, plant and environmental inoculants (Higa, 1995).

Beneficial and Effective Microorganisms for a Sustainable Agriculture Towards Agriculture Without Chemicals and With Optimum Yields of High Quality Crops.

Agriculture in a broad sense, is not an enterprise which leaves everything to nature without intervention. Rather it is a human activity in which the farmer attempts to integrate certain agroecological factors and production inputs for optimum crop and livestock production. Thus, it is reasonable to assume that farmers should be interested in ways and means of controlling beneficial soil microorganisms as an important component of the agricultural environment. Nevertheless, this idea has often been rejected by naturalists and proponents of nature farming and organic agriculture. They argue that beneficial soil microorganisms will increase naturally when organic amendments are applied to soils as carbon, energy and nutrient sources. This indeed may be true where an abundance of organic materials are readily available for recycling which often occurs in small-scale farming. However, in most cases, soil microorganisms, beneficial or harmful, have often been controlled advantageously when crops in various agroecological zones are grown and cultivated in proper sequence (i.e., crop rotations) and without the use of pesticides. This would explain why scientists have long been interested in the use of beneficial microorganisms as soil and plant inoculants to shift the microbiological equilibrium in a way that enhances soil quality and the yield and quality of crops (Higa and Wididana, 1991b; Higa, 1994:1995).

Most would agree that a basic rule of agriculture is to ensure that specific crops are grown according to their agroclimatic and agroecological requirements. However, in many cases the agricultural economy is based on market forces that demand a stable supply of food, and thus, it becomes necessary to use farmland to its full productive potential throughout the year.

The purpose of crop breeding is to improve crop production, crop protection, and crop quality. Improved crop cultivars along with improved cultural and management practices have made it possible to grow a wide variety of agricultural and horticultural crops in areas where it once would not have been culturally or economically feasible. The cultivation of these crops in such diverse environments has contributed significantly to a stable food supply in many countries. However, it is somewhat ironic that new crop cultures are almost never selected with consideration of their nutritional quality or bioavailability after ingestion (Hornick, 1992).

As will be discussed later, crop growth and development are closely related to the nature of the soil microflora, especially those in close proximity to plant roots, i.e., the rhizosphere. Thus, it will be difficult to overcome the limitations of conventional agricultural technologies without controlling soil microorganisms. This particular tenet is further reinforced because the evolution of most forms of life on earth and their environments are sustained by microorganisms. Most biological activities are influenced by the state of these invisible, minuscule units of life. Therefore, to significantly increase food production, it is essential to develop crop cultivars with

improved genetic capabilities (i.e., greater yield potential, disease resistance, and nutritional quality) and with a higher level of environmental competitiveness, particularly under stress conditions (i.e., low rainfall, high temperatures, nutrient deficiencies, and aggressive weed growth).

To enhance the concept of controlling and utilizing beneficial microorganisms for crop production and protection, one must harmoniously integrate the essential components for plant growth and yield including light (intensity, photoperiodicity and quality), carbon dioxide, water, nutrients (organic-inorganic) soil type, and the soil microflora. Because of these vital interrelationships, it is possible to envision a new technology and a more energy-efficient system of biological production.

Low agricultural production efficiency is closely related to a poor coordination of energy conversion which, in turn, is influenced by crop physiological factors, the environment, and other biological factors including soil microorganisms. The soil and rhizosphere microflora can accelerate the growth of plants and enhance their resistance to disease and harmful insects by producing bioactive substances. These microorganisms maintain the growth environment of plants, and may have secondary effects on crop quality. A wide range of results are possible depending on their predominance and activities at any one time. Nevertheless, there is a growing consensus that it is possible to attain maximum economic crop yields of high quality, at higher net returns, without the application of chemical fertilizers and pesticides. Until recently, this was not thought to be a very likely possibility using conventional agricultural methods. However, it is important to recognize that the best soil and crop management practices to achieve a more sustainable agriculture will also enhance the growth, numbers and activities of beneficial soil microorganisms that, in turn, can improve the growth, yield and quality of crops (National Academy of Sciences, 1989; Hornick, 1992; Parr et al., 1992).

CONTROLLING THE SOIL MICROFLORA: PRINCIPLES AND STRATEGIES

Principles of Natural Ecosystems and the Application of Beneficial and Effective Microorganisms

The misuse and excessive use of chemical fertilizers and pesticides have often adversely affected the environment and created many a) food safety and quality and b) human and animal health problems. Consequently, there has been a growing interest in nature farming and organic agriculture by consumers and environmentalists as possible alternatives to chemical-based, conventional agriculture.

Agricultural systems which conform to the principles of natural ecosystems are now receiving a great deal of attention in both developed and developing countries. A number of books and journals have recently been published which deal with many aspects of natural farming systems. New concepts such as alternative agriculture,

sustainable agriculture, soil quality, integrated pest management, integrated nutrient management and even beneficial microorganisms are being explored by the agricultural research establishment (National Academy of Sciences, 1989; Reganold et al., 1990; Parr et al., 1992). Although these concepts and associated methodologies hold considerable promise, they also have limitations. For example, the main limitation in using microbial inoculants is the problem of reproducibility and lack of consistent results.

Unfortunately certain microbial cultures have been promoted by their suppliers as being effective for controlling a wide range of soil-borne plant diseases when in fact they were effective only on specific pathogens under very specific conditions. Some suppliers have suggested that their particular microbial inoculant is akin to a pesticide that would suppress the general soil microbial population while increasing the population of a specific beneficial microorganism. Nevertheless, most of the claims for these single-culture microbial inoculants are greatly exaggerated and have not proven to be effective under field conditions. One might speculate that if all of the microbial cultures and inoculants that are available as marketed products were used some degree of success might be achieved because of the increased diversity of the soil microflora and stability that is associated with mixed cultures. While this, of course, is a hypothetical example, the fact remains that there is a greater likelihood of controlling the soil microflora by introducing mixed, compatible cultures rather than single pure cultures (Higa, 1991).

Even so, the use of mixed cultures in this approach has been criticized because it is difficult to demonstrate conclusively which microorganisms are responsible for the observed effects, how the introduced microorganisms interact with the indigenous species, and how these new associations affect the soil/plant environment. Thus, the use of mixed cultures of beneficial microorganisms as soil inoculants to enhance the growth, health, yield, and quality of crops has not gained widespread acceptance by the agricultural research establishment because conclusive scientific proof is often lacking.

The use of mixed cultures of beneficial microorganisms as soil inoculants is based on the principles of natural ecosystems which are sustained by their constituents; that is, by the quality and quantity of their inhabitants and specific ecological parameters, i.e., the greater the diversity and number of the inhabitants, the higher the order of their interaction and the more stable the ecosystem. The mixed culture approach is simply an effort to apply these principles to natural systems such as agricultural soils, and to shift the microbiological equilibrium in favor of increased plant growth, production and protection (Higa, 1991; 1994; Parr et al., 1994).

It is important to recognize that soils can vary tremendously as to their types and numbers of microorganisms. These can be both beneficial and harmful to plants and often the predominance of either one depends on the cultural and management practices that are applied. It should also be emphasized that most fertile and productive soils have a high content of organic matter and, generally, have large, populations of highly diverse microorganisms (i.e., both species and

genetic diversity). Such soils will also usually have a wide ratio of beneficial to harmful microorganisms (Higa and Wididana, 1991b).

Controlling the Soil Microflora for Optimum Crop Production and Protection

The idea of controlling and manipulating the soil microflora through the use of inoculants organic amendments and cultural and management practices to create a more favorable soil microbiological environment for optimum crop production and protection is not new. For almost a century, microbiologists have known that organic wastes and residues, including animal manures, crop residues, green manures, municipal wastes (both raw and composted), contain their own indigenous populations of microorganisms often with broad physiological capabilities.

It is also known that when such organic wastes and residues are applied to soils many of these introduced microorganisms can function as biocontrol agents by controlling or suppressing soil-borne plant pathogens through their competitive and antagonistic activities. While this has been the theoretical basis for controlling the soil microflora, in actual practice the results have been unpredictable and inconsistent, and the role of specific microorganisms has not been well-defined.

For, many years microbiologists have tried to culture beneficial microorganisms for use as soil inoculants to overcome the harmful effects of phytopathogenic organisms, including bacteria, fungi and nematodes. Such attempts have usually involved single applications of pure cultures of microorganisms which have been largely unsuccessful for several reasons. First, it is necessary to thoroughly understand the individual growth and survival characteristics of each particular beneficial microorganism, including their nutritional and environmental requirements. Second, we must understand their ecological relationships and interactions with other microorganisms, including their ability to coexist in mixed cultures and after application to soils (Higa, 1991; 1994).

There are other problems and constraints that have been major obstacles to controlling the microflora of agricultural soils. First and foremost is the large number of types of microorganisms that are present at any one time, their wide range of physiological capabilities, and the dramatic fluctuations in their populations that can result from man's cultural and management practices applied to a particular farming system. The diversity of the total soil microflora depends on the nature of the soil environment and those factors which affect the growth and activity of each individual organism including temperature, light, aeration, nutrients, organic matter, pH and water. While there are many microorganisms that respond positively to these factors, or a combination thereof, there are many that do not. Microbiologists have actually studied relatively few of the microorganisms that exist in most agricultural soil, mainly because we don't know how to culture them; i.e., we know very little about their growth, nutritional, and ecological requirements.

The "diversity" and "population" factors associated with the soil microflora have discouraged scientists from conducting research to develop control strategies. Many believe that, even when beneficial microorganisms are cultured and inoculated into soils, their number is relatively small compared with the indigenous soil inhabitants, and they would likely be rapidly overwhelmed by the established soil microflora. Consequently, many would argue that even if the application of beneficial microorganisms is successful under limited conditions (e.g., in the laboratory) it would be virtually impossible to achieve the same success under actual field conditions. Such thinking still exists today, and serves as a principle constraint to the concept of controlling the soil microflora (Higa, 1994).

It is noteworthy that most of the microorganisms encountered in any particular soil are harmless to plants with only a relatively few that function as plant pathogens or potential pathogens. Harmful microorganisms become dominant if conditions develop that are favorable to their growth, activity and reproduction. Under such conditions, soil-borne pathogens (e.g., fungal pathogens) can rapidly increase their populations with devastating effects on the crop. If these conditions change, the pathogen population declines just as rapidly to its original state. Conventional farming systems that tend toward the consecutive planting of the same crop (i.e., monoculture) necessitate the heavy use of chemical fertilizers and pesticides. This, in turn, generally increases the probability that harmful, disease-producing, plant pathogenic microorganisms will become more dominant in agricultural soils (Higa, 1991; 1994; Parr and Hornick, 1994). Chemical-based conventional farming methods are not unlike symptomatic therapy. Examples of this are applying fertilizers when crops show symptoms of nutrient-deficiencies, and applying pesticides whenever crops are attacked by insects and diseases. In efforts to control the soil microflora some scientists feel that the introduction of beneficial microorganisms should follow a symptomatic approach. However, we do not agree. The actual soil conditions that prevail at any point in time may be most unfavorable to the growth and establishment of laboratory-cultured, beneficial microorganisms. To facilitate their establishment, it may require that the farmer make certain changes in his cultural and management practices to induce conditions that will (a) allow the growth and survival of the inoculated microorganisms and (b) suppress the growth and activity of the indigenous plant pathogenic microorganisms (Higa, 1994; Parr et al., 1994).

An example of the importance of controlling the soil microflora and how certain cultural and management practices can facilitate such control is useful here. Vegetable cultivars are often selected on their ability to grow and produce over a wide range of temperatures. Under cool, temperate conditions there are generally few pest and disease problems. However, with the onset of hot weather, there is a concomitant increase in the incidence of diseases and insects making it rather difficult to obtain acceptable yields without applying pesticides. With higher temperatures, the total soil microbial population increases as does certain plant pathogens such as *Fusarium*, which is one of the main putrefactive, fungal pathogens in soil. The incidence and destructive activity of this pathogen can be greatly minimized by adopting reduced tillage methods and by shading techniques

to keep the soil cool during hot weather. Another approach is to inoculate the soil with beneficial, antagonistic, antibiotic-producing microorganisms such as actinomycetes and certain fungi (Higa and Wididana, 1991a; 1991b).

Application of Beneficial and Effective Microorganisms: A New Dimension

Many microbiologists believe that the total number of soil microorganisms can be increased by applying organic amendments to the soil. This is generally true because most soil microorganisms are heterotrophic, i.e., they require complex organic molecules of carbon and nitrogen for metabolism and biosynthesis. Whether the regular addition of organic wastes and residues will greatly increase the number of beneficial soil microorganisms in a short period of time is questionable. However, we do know that heavy applications of organic materials, such as seaweed, fish meal, and chitin from crushed crab shells, not only helps to balance the micronutrient content of a soil but also increases the population of beneficial antibiotic-producing actinomycetes. This changes the soil to a disease-suppressive condition within a relatively short period.

The probability that a particular beneficial microorganism will become predominant, even with organic farming or nature farming methods, will depend on the ecosystem and environmental conditions. It can take several hundred years for various species of higher and lower plants to interact and develop into a definable and stable ecosystem. Even if the population of a specific microorganism is increased through cultural and management practices, whether it will be beneficial to plants is another question. Thus, the likelihood of a beneficial, plant-associated microorganism becoming predominant under conservation-based farming systems is virtually impossible to predict. Moreover, it is very unlikely that the population of useful anaerobic microorganisms, which usually comprise only a small part of the soil microflora, would increase significantly even under natural farming conditions.

This information then emphasizes the need to develop methods for isolating and selecting different microorganisms for their beneficial effects on soils and plants. The ultimate goal is to select microorganisms that are physiologically and ecologically compatible with each other and that can be introduced as mixed cultures into soil where their beneficial effects can be realized (Higa, 1991; 1994; 1995).

Application of Beneficial and Effective Microorganisms: Fundamental Considerations

Microorganisms are utilized in agriculture for various purposes; as important components of organic amendments and composts, as legume inoculants for biological nitrogen fixation as a means of suppressing insects and plant diseases to improve crop quality and yields, and for reduction of labor. All of these are closely related to each other. An important consideration in the application of beneficial microorganisms to soils is the enhancement of their synergistic effects. This is difficult to accomplish if these microorganisms are applied to achieve

symptomatic therapy, as in the case of chemical fertilizers and pesticides (Higa, 1991; 1994).

If cultures of beneficial microorganisms are to be effective after inoculation into soil, it is important that their initial populations be at a certain critical threshold level. This helps to ensure that the amount of bioactive substances produced by them will be sufficient to achieve the desired positive effects on crop production and/or crop protection. If these conditions are not met, the introduced microorganisms, no matter how useful they are, will have little if any effect. At present, there are no chemical tests that can predict the probability of a particular soil-inoculated microorganism to achieve a desired result. The most reliable approach is to inoculate the beneficial microorganism into soil as part of a mixed culture, and at a sufficiently high inoculum density to maximize the probability of its adaptation to environmental and ecological conditions (Higa and Wididana, 1991b; Parr et al., 1994).

The application of beneficial microorganisms to soil can help to define the structure and establishment of natural ecosystems. The greater the diversity of the cultivated plants that are grown and the more chemically complex the biomass, the greater the diversity of the soil microflora as to their types, numbers and activities. The application of a wide range of different organic amendments to soils can also help to ensure a greater microbial diversity. For example, combinations of various crop residues, animal manures, green manures, and municipal wastes applied periodically to soil will provide a higher level of microbial diversity than when only one of these materials is applied. The reason for this is that each of these organic materials has its own unique indigenous microflora which can greatly affect the resident soil microflora after they are applied, at least for a limited period.

CLASSIFICATION OF SOILS BASED ON THEIR MICROBIOLOGICAL PROPERTIES

Most soils are classified on the basis of their chemical and physical properties; little has been done to classify soils according to their physicochemical and microbiological properties. The reason for this is that a soil's chemical and physical properties are more readily defined and measured than their microbiological properties. Improved soil quality is usually characterized by increased infiltration; aeration, aggregation and organic matter content and by decreased bulk density, compaction, erosion and crusting. While these are important indicators of potential soil productivity, we must give more attention to soil biological properties because of their important relationship (though poorly understood) to crop production, plant and animal health, environmental quality, and food safety and quality. Research is needed to identify and quantify reliable and predictable biological/ecological indicators of soil quality. Possible indicators might include total species diversity or genetic diversity of beneficial soil microorganisms as well as insects and animals (Reganold et al., 1990; Parr et al., 1992).

The basic concept here is not to classify soils for the study of microorganisms but for farmers to be able to control the soil microflora so that biologically-mediated processes can improve the growth, yield, and quality of crops as well as the tilth, fertility, and productivity of soils. The ultimate objective is to reduce the need for chemical fertilizers and pesticides (National Academy of Sciences, 1989; 1993).

Functions of Microorganisms: Putrefaction, Fermentation, and Synthesis

Soil microorganisms can be classified into decomposer and synthetic microorganisms. The decomposer microorganisms are subdivided into groups that perform oxidative and fermentative decomposition. The fermentative group is further divided into useful fermentation (simply called fermentation) and harmful fermentation (called putrefaction). The synthetic microorganisms can be subdivided into groups having the physiological abilities to fix atmospheric nitrogen into amino acids and/or carbon dioxide into simple organic molecules through photosynthesis. Figure 1 (adapted from Higa) is a simplified flow chart of organic matter transformations by soil microorganisms that can lead to the development of disease-inducing, disease-suppressive, zymogenic, or synthetic soils.

Fermentation is an anaerobic process by which facultative microorganisms (e.g., yeasts) transform complex organic molecules (e.g., carbohydrates) into simple organic compounds that often can be absorbed directly by plants. Fermentation yields a relatively small amount of energy compared with aerobic decomposition of the same substrate by the same group of microorganisms. Aerobic decomposition results in complete oxidation of a substrate and the release of large amounts of energy, gas, and heat with carbon dioxide and water as the end products. Putrefaction is the process by which facultative heterotrophic microorganisms decompose proteins anaerobically, yielding malodorous incompletely oxidized, metabolites (e.g., ammonia, mercaptans and indole) that are often toxic to plants and animals.

The term "synthesis" as used here refers to the biosynthetic capacity of certain microorganisms to derive metabolic energy by "fixing" atmospheric nitrogen and/or carbon dioxide. In this context we refer to these as "synthetic" microorganisms, and if they should become a predominant part of the soil microflora, then the soil would be termed a "synthetic" soil.

Nitrogen-fixing microorganisms are highly diverse, ranging from "free-living" autotrophic bacteria of the genus *Azotobacter* to symbiotic, heterotrophic bacteria of the genus *Rhizobium*, and blue-green algae (now mainly classified as blue-green bacteria), all of which function aerobically. Photosynthetic microorganisms fix atmospheric carbon dioxide in a manner similar to that of green plants. They are also highly diverse, ranging from blue-green algae to green algae that perform complete photosynthesis aerobically to photosynthetic bacteria which perform incomplete photosynthesis anaerobically.

Relationships Between Putrefaction, Fermentation, and Synthesis

The processes of putrefaction, fermentation, and synthesis proceed simultaneously according to the appropriate types and numbers of microorganisms that are present in the soil. The impact on soil quality attributes and related soil properties is determined by the dominant process. The production of organic substances by microorganisms results from the intake of positive ions, while decomposition serves to release these positive ions. Hydrogen ions play a pivotal role in these processes. A problem occurs when hydrogen ions do not recombine with oxygen to form water but are utilized to produce methane, hydrogen sulfide, ammonia, mercaptans and other highly reduced putrefactive substances most of which are toxic to plants and produce malodors. If a soil is able to absorb the excess hydrogen ions during periods of soil anaerobiosis and if synthetic microorganisms such as photosynthetic bacteria are present, they will utilize these putrefactive substances and produce useful substrates from them which helps to maintain a healthy and productive soil.

The photosynthetic bacteria, which perform incomplete photosynthesis anaerobically, are highly desirable, beneficial soil microorganisms because they are able to detoxify soils by transforming reduced, putrefactive substances such as hydrogen sulfide into useful substrates. This helps to ensure efficient utilization of organic matter and to improve soil fertility. Photosynthesis involves the photo-catalyzed splitting of water which yields molecular oxygen as a by-product. Thus, these microorganisms help to provide a vital source of oxygen to plant roots.

Reduced compounds such as methane and hydrogen sulfide are often produced when organic materials are decomposed under anaerobic conditions. These compounds are toxic and can greatly suppress the activities of nitrogen-fixing microorganisms. However, if synthetic microorganisms, such as photosynthetic bacteria that utilize reduced substances, are present in the soil, oxygen deficiencies are not likely to occur. Thus, nitrogen-fixing microorganisms, coexisting in the soil with photosynthetic bacteria, can function effectively in fixing atmospheric nitrogen even under anaerobic conditions.

Photosynthetic bacteria not only perform photosynthesis but can also fix-nitrogen. Moreover, it has been shown that, when they coexist, in soil with species of *Azotobacter*, their ability to fix nitrogen is enhanced. This then is an example of a synthetic soil. It also suggests that by recognizing the role, function, and mutual compatibility of these two bacteria and utilizing them effectively to their full potential, soils can be induced to a greater synthetic capacity. Perhaps the most effective synthetic soil system results from the enhancement of zymogenic and synthetic microorganisms; this allows fermentation to become dominant over putrefaction and useful synthetic processes to proceed.

Classification of Soils Based on the Functions of Microorganisms

As discussed earlier (Figure 1), soils can be characterized according to their indigenous microflora which perform putrefactive, fermentative, synthetic and zymogenic reactions and processes. In most soils, these three functions are going on simultaneously with the rate and extent of each determined by the types and numbers of associated microorganisms that are actively involved at any one time.

A simple diagram showing a classification of soils based on the activities and functions of their predominant microorganisms is presented in Fig. 2.

Disease-Inducing Soils. In this type of soil, plant pathogenic microorganisms such as *Fusarium* fungi can comprise 5 to 20 percent of the total microflora if fresh organic matter with a high nitrogen content is applied to such a soil, incompletely oxidized products can arise that are malodorous and toxic to growing plants. Such soils tend to cause frequent infestations of disease organisms, and harmful insects. Thus, the application of fresh organic matter to these soils is often harmful to crops. Probably more than 90 percent of the agricultural land devoted to crop production worldwide can be classified as having disease-inducing soil. Such soils generally have poor physical properties, and large amounts of energy are lost as "greenhouse" gases, particularly in the case of rice fields. Plant nutrients are also subject to immobilization into unavailable forms.

Disease-Suppressive Soils. The microflora of disease-suppressive soils is usually dominated by antagonistic microorganisms that produce copious amounts of antibiotics. These include fungi of the genera *Penicillium*, *Trichoderma*, and *Aspergillus*, and actinomycetes of the genus *Streptomyces*. The antibiotics they produce can have biostatic and biocidal effects on soil-borne plant pathogens, including *Fusarium* which would have an incidence in these soils of less than 5 percent. Crops planted in these soils are rarely affected by diseases or insect pests. Even if fresh organic matter with a high nitrogen content is applied, the production of putrescent substances is very low and the soil has a pleasant earthy odor after the organic matter is decomposed. These soils generally have excellent physical properties; for example, they readily form water-stable aggregates and they are well-aerated, and have a high permeability to both air and water. Crop yields in the disease-suppressive soils are often slightly lower than those in synthetic soils. Highly acceptable crop yields are obtained whenever a soil has a predominance of both disease-suppressive and synthetic microorganisms.

Zymogenic Soils. These soils are dominated by a microflora that can perform useful kinds of fermentations, i.e., the breakdown of complex organic molecules into simple organic substances and inorganic materials. The organisms can be either obligate or facultative anaerobes. Such fermentation-producing microorganisms often comprise the microflora of various organic materials, i.e., crop residues, animal manures, green manures and municipal wastes including composts. After these amendments are applied to the soil, their number: and

fermentative activities can increase dramatically and overwhelm the indigenous soil microflora for an indefinite period. While these microorganisms remain predominant, the soil can be classified as a zymogenic soil which is generally characterized by a) pleasant, fermentative odors especially after tillage, b) favorable soil physical properties (e.g., Increased aggregate stability, permeability, aeration and decreased resistance to tillage c) large amounts of inorganic nutrients, amino acids, carbohydrates, vitamins and other bioactive substances which can directly or indirectly enhance the growth, yield and quality of crops, d) low occupancy of *Fusarium* fungi which is usually less than 5 percent, and e) low production of greenhouse gases (e.g., methane, ammonia, and carbon dioxide) from croplands, even where flooded rice is grown.

Synthetic Soils. These soils contain significant populations of microorganisms which are able to fix atmospheric nitrogen and carbon dioxide into complex molecules such as amino acids, proteins and carbohydrates. Such microorganisms include photosynthetic bacteria which perform incomplete photosynthesis anaerobically, certain Phycomycetes (fungi that resemble algae), and both green algae and blue-green algae which function aerobically. All of these are photosynthetic organisms that fix atmospheric nitrogen. If the water content of these soils is stable, their fertility can be largely maintained by regular additions of only small amounts of organic materials. These soils have a low *Fusarium* occupancy and they are often of the disease-suppressive type. The production of gases from fields where synthetic soils are present is minimal, even for flooded rice.

This is a somewhat simplistic classification of soils based on the functions of their predominant types of microorganisms, and whether they are potentially beneficial or harmful to the growth and yield of crops. While these different types of soils are described here in a rather idealized manner, the fact is that in nature they are not always clearly defined because they often tend to have some of the same characteristics. Nevertheless, research has shown that a disease-inducing soil can be transformed into disease-suppressing, zymogenic and synthetic soils by inoculating the problem soil with mixed cultures of effective microorganisms (Higa, 1991; 1994; Parr et al., 1994). Thus it is somewhat obvious that the most desirable agricultural soil for optimum growth, production, protection, and quality of crops would be the composite soil indicated in Fig. 2, i.e., a soil that is highly zymogenic and synthetic, and has an established disease-suppressive capacity. This then is the principle reason for seeking ways and means of controlling the microflora of agricultural soils.

SUMMARY AND CONCLUSIONS

Controlling the soil microflora to enhance the predominance of beneficial and effective microorganisms can help to improve and maintain the soil chemical and physical properties. The proper and regular addition of organic amendments are often an important part of any strategy to exercise such control.

Previous efforts to significantly change the indigenous microflora of a soil by introducing single cultures of extrinsic microorganisms have largely been unsuccessful. Even when a beneficial microorganism is isolated from a soil, cultured in the laboratory, and reinoculated into the same soil at a very high population, it is immediately subject to competitive and antagonistic effects from the indigenous soil microflora and its numbers soon decline. Thus, the probability of shifting the "microbiological equilibrium" of a soil and controlling it to favor the growth, yield and health of crops is much greater if mixed cultures of beneficial and effective microorganisms are introduced that are physiologically and ecologically compatible with one another. When these mixed cultures become established their individual beneficial effects are often magnified in a synergistic manner.

Actually, a disease-suppressive microflora can be developed rather easily by selecting and culturing certain types of gram-positive bacteria that produce antibiotics and have a wide range of specific functions and capabilities; these organisms include facultative anaerobes, obligate aerobes, acidophilic and alkalophilic microbes. These microorganisms can be grown to high populations in a medium consisting of rice bran, oil cake and fish meal and then applied to soil along with well-cured compost that also has a large stable population of beneficial microorganisms, especially facultative anaerobic bacteria. A soil can be readily transformed into a zymogenic/synthetic soil with disease-suppressive potential if mixed cultures of effective microorganisms with the ability to transmit these properties are applied to that soil.

The desired effects from applying cultured beneficial and effective microorganisms to soils can be somewhat variable, at least initially. In some soils, a single application (i.e., inoculation) may be enough to produce the expected results, while for other soils even repeated applications may appear to be ineffective. The reason for this is that in some soils it takes longer for the introduced microorganisms to adapt to a new set of ecological and environmental conditions and to become well-established as a stable, effective and predominant part of the indigenous soil microflora. The important consideration here is the careful selection of a mixed culture of compatible, effective microorganisms properly cultured and provided with acceptable organic substrates. Assuming that repeated applications are made at regular intervals during the first cropping season, there is a very high probability that the desired results will be achieved.

There are no meaningful or reliable tests for monitoring the establishment of mixed cultures of beneficial and effective microorganisms after application to a soil. The

desired effects appear only after they are established and become dominant, and remain stable and active in the soil. The inoculum densities of the mixed cultures and the frequency of application serve only as guidelines to enhance the probability of early establishment. Repeated applications, especially during the first cropping season, can markedly facilitate early establishment of the introduced effective microorganisms.

Once the "new" microflora is established and stabilized, the desired effects will continue indefinitely and no further applications are necessary unless organic amendments cease to be applied, or the soil is subjected to severe drought or flooding.

Finally, it is far more likely that the microflora of a soil can be controlled through the application of mixed cultures of selected beneficial and effective microorganisms than by the use of single or pure cultures. If the microorganisms comprising the mixed culture can coexist and are physiologically compatible and mutually complementary, and if the initial inoculum density is sufficiently high, there is a high probability that these microorganisms will become established in the soil and will be effective as an associative group, whereby such positive interactions would continue. If so, then it is also highly probable that they will exercise considerable control over the indigenous soil microflora which, in due course, would likely be transformed into or replaced by a "new" soil microflora.

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Table 1.

Some Common Functions of Beneficial and Harmful Soil Microorganisms as they Affect Soil Quality, Crop Production, and Plant Health.

Functions of Beneficial Microorganisms

- Fixation of atmospheric nitrogen
- Decomposition of organic wastes and residues
- Suppression of soil-borne pathogens
- Recycling and increased availability of plant nutrients
- Degradation of toxicants including pesticides
- Production of antibiotics and other bioactive compounds
- Production of simple organic molecules for plant uptake
- Complexation of heavy metals to limit plant uptake
- Solubilization of insoluble nutrient sources
- Production of polysaccharides to improve soil aggregation

Functions of Harmful Microorganisms

- Induction of plant diseases
- Stimulation of soil-borne pathogens
- Immobilization of plant nutrients
- Inhibition of seed germination
- Inhibition of plant growth and development
- Production of phytotoxic substances

Fig 1

Flow chart of organic matter transformations by soil microorganisms and the development of soils that may be disease-inducing, zymogenic or synthetic (adapted from Prof. Dr. Teruo Higa).

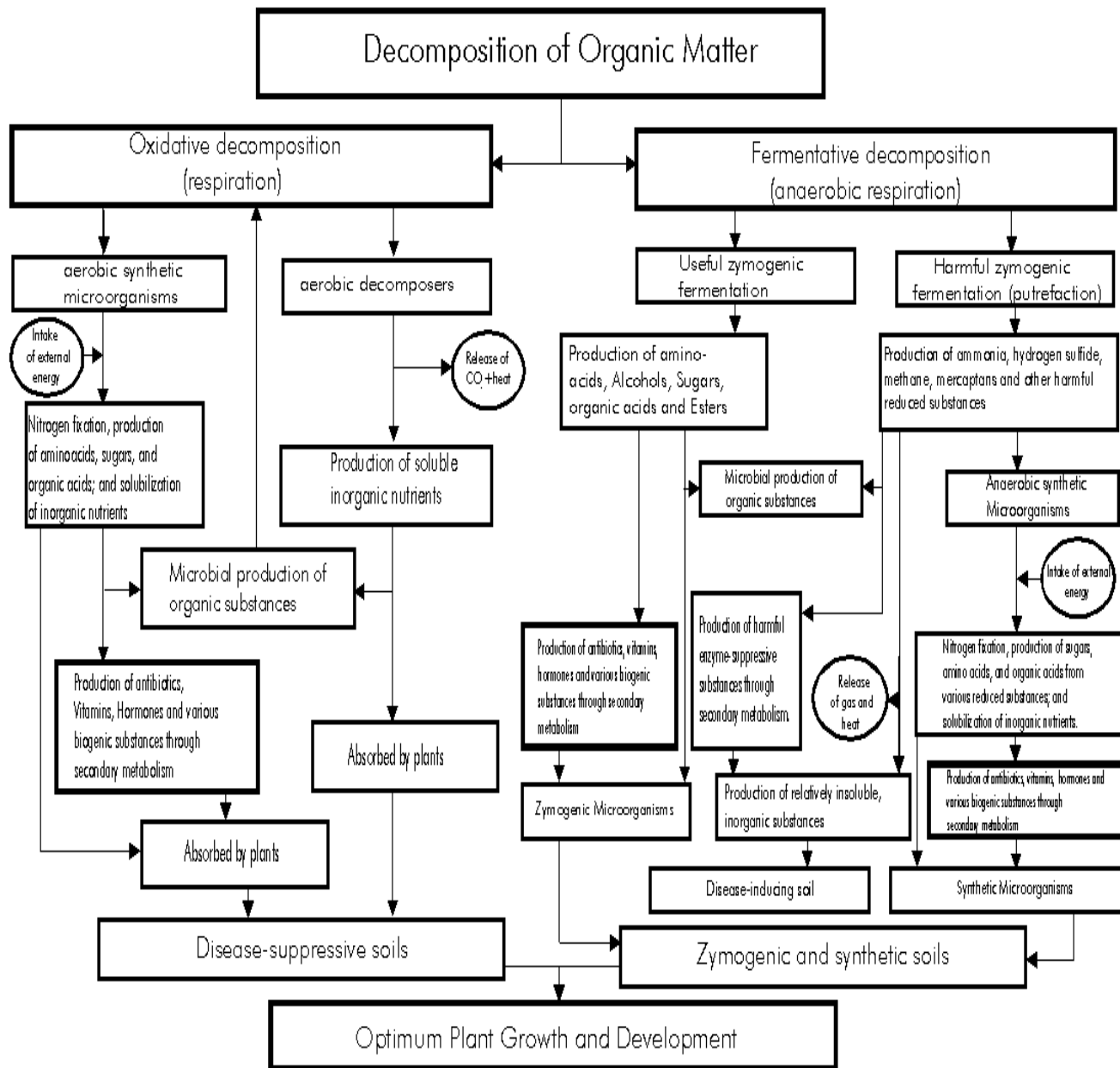


Fig 2

Classification of soils based on the activities and functions of their predominant microorganisms.

Disease-inducing soils can be transformed into disease suppressing, zymogenic and synthetic soils that are more conducive to growth and health of plants by introducing beneficial microorganisms as microbial inoculants and following best management practices. The ideal soil for a more sustainable agriculture is a composite of the other three soil types and contains associative groups of beneficial microorganisms to enhance the optimum growth, yield and quality of crops.

