

A review: Biofertilizer - A key player in enhancing soil fertility and crop productivity

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Plants nutrients are essential for the production of crops and healthy food for the world's ever increasing population. Soil management strategies today are mainly dependent on inorganic chemical-based fertilizers, which cause a serious threat to human health and the environment. Bio-fertilizer has been identified as an alternative for increasing soil fertility and crop production in sustainable farming. The exploitation of beneficial microbes as bio-fertilizers has become of paramount importance in agricultural sector due to their potential role in food safety and sustainable crop production.

Bio-fertilizer can be an important component of integrated nutrients management. Microorganisms that are commonly used as bio-fertilizer components include; nitrogen fixers (N-fixer), potassium and phosphorus solubilizers, growth promoting rhizobacteria (PGPRs), endo and ecto mycorrhizal fungi, cyanobacteria and other useful microscopic organisms. The use of bio-fertilizers leads to improved nutrients and water uptake, plant growth and plant tolerance to abiotic and biotic factors. These potential biological fertilizers would play a key role in productivity and sustainability of soil and also in protecting the environment as eco-friendly and cost effective inputs for the farmers.

Key Words: Biofertilizer; Soil fertility; Crop productivity

Agriculture has undergone series of advancement since the 12th century and is being practiced extensively throughout the world today. In 2007, it was recorded that one-third of the world's workers were employed in the area of agriculture. According to Food and Agricultural Development, 55% of people in Africa can earn a living through agriculture. In Nigeria, agriculture is the basic means of livelihood for people in most areas especially the rural areas and we solemnly depend on them for food (1). With the current fall in price of crude oil which, today is Nigeria's major source of income; the President of Nigeria, Muhammadu Buhari GCFR emphasized that the nation must go back to agriculture, particularly crop farming which used to be the country's source of income before the discovery of crude oil (2). To get food to feed Nigeria's teeming population which according to Nigerian population census (2006) is expected to rise to 221 million by the year 2020, there is need for very fertile soils, for sustainable crop production.

According to Khosro and Yousef (3) the most important constraint limiting crop yield in developing nations worldwide and especially among resource-poor farmers, is soil infertility. Therefore, maintaining soil quality can reduce the problems of land degradation, decreasing soil fertility, and rapidly declining production levels in large parts of the world that needed the basic principles of good farming-practice. Mfilinge et al. (4) said that low crop productivity is a general problem facing most farming systems in Sub-Saharan Africa (SSA). These low yields are pronounced in legumes and are often associated with declining soil fertility and reduced nitrogen fixation due to biological and environmental factors. Biological nitrogen fixation (BNF), a key source of nitrogen for farmers using little fertilizer, constitutes one of the potential solutions and plays a key role in sustainable production of legumes and even non legumes.

The continuous cultivation of the same piece of land year in year out due to increased population has resulted to a decline in soil fertility such that even with the application of chemical inorganic fertilizer, little is obtained in return.

Conventional agriculture plays an important role in meeting the food needs of a growing human population, which has led to an increasing dependence on the use of chemical fertilizers and pesticides for increased productivity (5). Chemical fertilizers are industrially made substances which are composed of known quantities of nitrogen, phosphorus and potassium.

The use of chemical fertilizers causes air and ground water pollution as a result of eutrophication of water bodies (6). According to Chun-Li et al. (7), though the practice of using chemical fertilizers and pesticides accelerates soil acidification, it also poses the risk of contaminating ground water and the atmosphere. It also weakens the roots of plants thereby making them to be susceptible to unwanted diseases. In this regard, attempts have recently been made towards the production of nutrient rich high quality fertilizer (Biofertilizer) to ensure bio-safety. Biofertilizer has been identified as an alternative to chemical fertilizer to increase soil fertility and crop production in sustainable farming. These potential biological fertilizers would play the key role in productivity and sustainability of soil and also protect the environment as eco-friendly and cost effective inputs for the farmers (3). Organic farming is one of such strategies that not only ensures food safety but also adds to biodiversity of soil (8). The application of biofertilizer to the soil increases the biodiversity which constitutes all kinds of useful bacteria and fungi including the arbuscular mycorrhiza fungi (AMF) called plant growth promoting rhizobacteria (PGPR) and nitrogen fixers. There are so many microorganisms thriving in the soil, especially in the rhizosphere of plant. A considerable number of these microorganisms possess a functional relationship and constitute a holistic system with plants. They have beneficial effects on plant growth (9). Application of beneficial microorganisms in agricultural practices started about 60 years ago and it is now evident that these beneficial microbes can also enhance plant resistance to adverse environmental stresses e. g., water and nutrient deficiency and heavy metal contamination.

Biofertilizers keep the soil environment rich in all kinds of macro and micro nutrients via nitrogen fixation, phosphate and potassium solubilisation or mineralization, release of plant growth regulating substances, production of antibiotics and biodegradation of organic matter in the soil (10). Biofertilizers, when applied as seed or soil inoculants, multiply and participate in nutrient cycling and leads to crop productivity. Generally, 60% to 90% of the total applied fertilizer is lost and the remaining 10% - 40% is taken up by plants. Hence biofertilizers can be important component of intergrated nutrient management systems for to sustaining agricultural productivity and a healthy environment (11).

Biofertilizers are products containing living cells of different of micro-

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organisms which have ability to convert nutritionally important elements from unavailable to available form through biological processes (9). This review is intended to x-ray the role of biofertilizers in sustainable agriculture thereby meeting the needs of agriculturists and plant biologists whose work focuses on creating clean and efficient means of to improving soil quality by nourishing and maintaining the useful and natural flora of microorganisms. Furthermore, it presents recent developments in the field of agricultural management that reveals the potentials of the application biofertilizers in terms of increased nutrient profiles, plant growth and productivity and an improved tolerance to environmental stress.

SOIL FERTILITY

Soil refers to the part of the earth on which plant grow (12). It consists of three layers: top soil, sub- soil, and parent material. However we are more concerned with the top soil since it is the part that favours plant growth. It contains minerals, air, water, living organism and inorganic and organic matter all of which have to be in a particular ratio with at least a medium pH to constitute a fertile soil. According to Purves et al. (12), a good quality soil is one that is 45% minerals (sand, silt, and clay), 25% water, 25% air, and 5% organic and living matter. The mineral portion of a soil which makes up half of the volume contains about 93% silica, aluminium and iron oxides; 4% calcium, potassium and magnesium oxides and 3% titanium, sodium and very small amount of nitrogen, sulphur, phosphorous, boron, manganese, zinc, copper, chlorine, molybdenum and many other elements. However, all these minerals only fourteen are essential to plant and these are called essential elements (12).

In Table 1, according to Barak (13), the essential nutrients are divided into micronutrient and macronutrients. The macronutrients are required in larger quantities whereas the micronutrients are required in small quantities by plants. The first six elements shown in Table 1 are macronutrient while the rest are micronutrients. The macronutrients are further divided into primary macronutrient (those that are always limited in the soil) and secondary macronutrients (those that are rarely limited). The primary nutrients are: nitrogen, phosphorous and potassium.

Nitrogen

Nitrogen is one of the primary macronutrient that is required by plants in large quantity. Its functions include: formation of base pair for RNA and DNA, formation of phosphate group of protein (example hemp group of chlorophyll) and hormones such as cytokines, metal uptake (phytosiderorphores), transport in xylem and phloem (example copper with amines), as osmoregulation (example in lettuce and spinach), alkaloids, miscobiochemicals e.g., mescaline, quinine (13).

Nitrogen is absorbed by plants in form of nitrates, ammonium and sometimes urea (1) and is added to the soil through fertilizer, biological nitrogen fixation, rainfall and thunder, and decomposition of organic matter. According to Barak (13) a plant deficient in nitrogen shows the following symptoms: stunted growth and light green or yellow leaves. Severe deficiency symptoms include necrosis starting from the tip of the older leaves and developing a 'v' pattern.

Phosphorus

Phosphorus is needed in relatively large amounts but at a quantity lower than that of nitrogen and potassium. It promotes legume growth and yield, nodule number and nodule mass. Its functions include: Phospholipids in membrane, phosphor proteins for life functions, improvement of crop yield and quality. It is important in seed formation. Analysis has shown that it forms a substantial component of seeds and fruits (14). Phosphorus is absorbed by plants in form of phosphates and is added to the soil through fertilizers, bone meal, and super phosphates. Barak (13) stated that phosphorus is phloem mobile and its deficiency symptoms are evenly spread throughout the plant. However, stunted growth and late maturity are often associated with its deficiency. In severe cases, grassy species such as corn show reddening of the leaves. According to Scalenghe et al. (14), excess phosphorus is not good as plants cannot properly assimilate it because most phosphorus is in inorganic form. Excess phosphorus is characterized by early maturation of plants and low crop yield in plants.

Potassium

Plants requirement for this element is only second to that of nitrogen. Potassium influences the water economy and crop growth through its effects on water uptake, root growth, maintenance of turgor, transpiration and stomatal regulation (4). The functions of potassium include the following:

It enhances the cold endurance of some plants as well as their resistance to fungal and bacterial diseases; It intensifies the synthesis of high molecular carbohydrates resulting in cell walls of the straw of cereals thicker. It catalyzes the activities of some enzymes and promotes the synthesis and accumulation of certain vitamins in plants (Thiamin and Riboflavin), It is essential for the activity of guard cells, It aids in photosynthesis, building up of protein and improving fruit quality (1,13).

Potassium is absorbed by plants in form of potassium ions which is insoluble in water. It is added to the soil by fertilizer, decaying organic matter and wood ash. According to Ifokwe (1), potassium deficiency upsets plant's metabolism, inhibits the activity of some enzymes and disrupts carbohydrate and protein metabolism. The author also stated that inadequate potassium supply reduces the viability of seeds, makes plant more prone to diseases and when harvested, they become difficult to preserve in marketable conditions. Other elements required by plants in large quantity include: oxygen, carbon, and hydrogen. These are obtained through air and water and therefore are bountiful in nature (13).

It has been shown that the overall yield of crops is determined by the combined effect of light, air, water, micro and macro nutrients, and pH. However, as the soil is continuously cultivated, nutrients in the soil progressively diminish because the rate at which they are removed is greater than the rate at which they are being replaced (15). According to Wander (15), apart from being taken up by plants, there are many other ways in which nutrients are lost from the soil. These include leaching, improper irrigation, over tillage, and bush burning. Loss of soil fertility is the primary cause of poor crops and the solution to this problem is by the use of an environment friendly bio-fertilizer. In recent years, agrochemicals such as chemical pesticides and chemical inorganic fertilizers are extensively applied to obtain high yield. Intensive application of agrochemicals leads to several agricultural problems and poor cropping systems. Some farmers use more chemicals fertilizers than the recommended levels for many crops. This practice accelerates soil acidification but also has the risk of contaminating underground water and the atmosphere, and also weakens the roots of plants and making them to be easy prey to unwanted diseases (7).

WHAT IS BIO-FERTILIZER?

A bio-fertilizer is simply a substance which contains living microorganisms which when applied to the soil, a seed or plant surface colonizes the rhizosphere and promotes growth by increasing the supply or availability of nutrients to the host plant (9). A bio-fertilizer is a modernized form of organic fertilizer into which beneficial microorganisms have been incorporated (16). According to Hari and Perumal (17) bio-fertilizer is most commonly referred to as selected strains of beneficial soil microorganisms cultured in the laboratory and packed in suitable carriers. In a large sense, the term bio-fertilizer may be used to include all organic resources for plant growth which are rendered in available form for plant absorption through microorganisms or plant associations or interactions (3).

History of bio-fertilizer

Bio-fertilizers such as *Rhizobium*, *Azotobacter*, *Azospirillum* and Blue green algae (BGA) have been in use a long time ago. The knowledge of applied microbial inoculum is a long history which passes from generation to generation of farmers. It started with culture of small scale compost production that has evidently proved the ability of bio-fertilizer (3). This is recognized when the culture accelerates the decomposition of organic residues and agricultural by-products through various processes and gives healthy harvest of crops (18). The commercial history of bio-fertilizer began with the launch of "Nitragin" by Nobbe and Hilther in 1895. This was followed by the discovery of *Azotobacter* and then Blue-green algae and a host of other microorganisms which are being used till date as bio-fertilizer (19).

In Malaysia, industrial scale production of microbial inoculants started in late 1940s and picking up in 1970s taking guide by *Bradyrhizobium* inoculation on legumes. Government Research Institute, the Malaysian Rubber Board (MRB) had been conducting research on *Rhizobium* inoculants for leguminous crops in the inter rows of young rubber trees in large plantation. Besides, University Putra Malaysia (UPM) also conducted many researches since 1980s on *Mycorrhiza* and initiated the research to evaluate the contribution of nitrogen from *Azospirillum* to oil palm seedling (18). Bio-fertilizers are usually prepared as carrier based inoculants containing effective microorganisms (9). Microorganisms used as bio-fertilizer include: Nitrogen fixers (N. fixers) e.g., *Rhizobium* Spp., *Cyanobacteria*, and *Azotobacter chroococcum*, potassium solubilizers (K - solubilizers) e.g., *Bacillus mucilaginosus*, phosphorus solubilizers (P - solubilizers) e.g., *Bacillus megaterium*, *Aspergillus*

fumigatus, Plant Growth Promoting Rhizobacteria (PGPR), Vesicular Arbuscular Mycorrhiza (VAM) e.g., *Glomus mosseae* and sulfur oxidizers (S - oxidizers).

Carrier materials used in making bio-fertilizer

Bio-fertilizers are usually amended with carrier material to increase effectiveness of the bio-fertilizers. It also increases water retention capacity (20). According to Somasegaran and Springer (21), a good carrier material must have the following characteristics. It must be cheap and readily available in adequate amounts, It must be easy to sterilize by autoclaving or gamma irradiation, It must be easy to process and must be free of lump-forming materials, It must be non-toxic to both the microorganisms and the plants, to which it is applied, It must have a good moisture absorption capacity, It must have good water retention capacity of more than 50%, It must have good adhesion to seeds, It must have good pH buffering capacity, It must have high organic matter content.

Khosro and Yousef (3) stated that the incorporation of microorganisms into carrier materials enables easy handling, long term storage, and effectiveness of the bio-fertilizer. They also stated that sterilization of carrier materials is essential to keep high numbers of inoculants on it during long storage period. Examples of carrier material include saw dust, talcum dust, manure, earthworm cast.

Gamma-irradiation is the most suitable way of carrier sterilization, because the sterilization process makes no change in the physical and chemical properties of the material. Another method of carrier sterilization is autoclaving. However, autoclaving may change the properties of some carrier materials resulting in the production of toxic substances that can kill some bacterial species (17).

Bio-fertilizer making

There are several things that need to be considered in making bio-fertilizers, such as microbes growth profile, types and optimum conditions of organism and formulation of inoculum. The formulation of inocula, method of application and storage of the products are all critical to the success of the biological product. In general, six steps are involved in making of bio-fertilizer. These are: choosing of active microorganisms, isolation and selection of target microbes, selection of method of propagation and carrier material, and phenotype testing, and large scale tests (3).

First of all, decision must be made on the active microorganisms to be used. For example, it must be decided whether to use organic acid bacteria or nitrogen fixer or a combination of some organisms, after which target microbes are isolated. Usually organisms are isolated from plant roots by luring it using a decoy such as placing cool rice underground beneath bamboo plants (3). Next, the isolated organism will be grown on Petri dishes before it is mass produced on flask. It is also important to choose the right carrier material. If the desire is to produce bio-fertilizer in powder form, then apioaca flour or peat are the right carrier materials to use. Selection of propagation method is mainly to find out the optimum growth condition of organism. This can be achieved by determining growth profile under different parameter and conditions after which the phenotype is tested and selection is made. Lastly, the bio-fertilizer is tested on large scale at different environment to analyze its effectiveness and limitations (3).

Mechanism of actions of bio-fertilizers

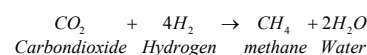
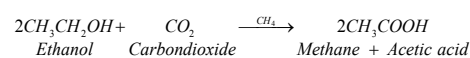
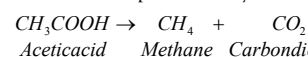
Among the PGPRs species, *Azospirillum* was suggested to secrete gibberellins, ethylene and auxins. Some plant associated bacteria can also induce phytohormone synthesis, for example lodge pine when inoculated with *Paenibacillus polymyxa* had elevated levels of IAA in the roots. *Rhizobium* and *Bacillus* were found to synthesize IAA at different cultural pH, temperature and in the presence of agro-waste as carrier material. Unlike other phytohormones, ethylene is responsible for inhibition of growth of dicot plants (22).

Biochemistry of bio-fertilizer production

The production of bio-fertilizer involves three biochemical steps which include the breaking down of complex materials into simpler substances in a process known as anaerobic digestion. (Anaerobic bio-digestion is a process whereby microorganisms breakdown biodegradable materials in the absence of oxygen (23). In the first stage, complex organic matter is broken down by cellulolytic microorganisms to produce simple molecules such as long chain fatty acids and other substances. In the second stage, the products from stage one are fermented leading to the production of simpler intermediates such as acetic acids, pyruvic acids, carbon dioxide and so on. In the third stage,

methanogens act on the products, giving off a mixture of gases known as biogas.

This can be represented by the following reactions:



Factors affecting bio-digestion

According Ezigbo (23), the rate of bio-degradation is affected by the following factors: 1. Moisture content-Bio-digestion occurs faster in the presence of moisture while lack of water makes the survival of degrading organisms difficult, 2. Surface area-This plays an important role in bio-degradation. The finer the particles, the faster the digestion rate, 3. pH- Bio-digestion occurs best at a medium to slightly acidic pH, 4. Temperature: High temperature favors bio-digestion while low temperatures slow it down.

Sophisticated bio-digesters have been built in countries such as Sweden and Japan (16) which combine all these parameters. The bio-digesters consist of a chamber where the substrates are screened thoroughly to avoid bringing plastic and metallic substances into the digester. It makes use of magnetic separation to separate the metals. Also, there is a chamber where pre-treatment and homogenization of the substance occur. The digesters make use of two temperatures; 37 °C for the growth mesophiles growth and 55°C for thermophiles. The gas given off during bio-digestion is known as "biogas". The bio-digesters have a chamber for collection of the biogas which has many uses. These bio-digesters make complete digestion in 24 hours (16).

Advantages of bio-digestion

Bio-digestion or bio-degradation of wastes has the following advantages: It leads to an increase in available nutrients, it leads to a decrease in weight thereby reducing handling and transportation problems arising due to bulkiness of the wastes, it reduces the volume of the wastes, it improves handling and transportation, High degradation temperatures kill pathogens and weed seeds, to enable graded wastes come into equilibrium with the soil, it reduces odor (24).

Advantages of bio-fertilizer over chemical fertilizers

Inorganic fertilizers have become very popular in Nigeria and throughout the world because they are easily affordable and have the advantage of fast action owing to their prompt release of nutrients. However, there has been much research on the demerits of inorganic fertilizers and this has revealed that they have disadvantages which cannot be overlooked. Most of the problems associated with harvested crops and some of the pollution of our natural environment occurred as a result of inorganic fertilizer use (25). These findings have led to the need for the provision of an environment friendly fertilizer known as bio-fertilizer. The world's demand for fertilizer has risen greatly in the past few decades. According to Fertilizer Suppliers Association of Nigeria (FESPAN) (2007), Nigeria's fertilizer demand is estimated to be 12 million metric tons per annum. All the fertilizers used in Nigeria are imported (1) and due to the high cost of importation, the price of the fertilizers become very high, preventing poor farmers from accessing it. Apart from the high cost, inorganic fertilizers when applied incorrectly, excessively or inadequately have negative effects. Many of the fertilizers imported into the country were wasted as farmers refused to purchase them. When interviewed, yam farmers complained that the fertilizers were responsible for the early decay of harvested yam tubers. It is also no secret that crops cultivated with inorganic fertilizers have less flavor, taste, and aroma than those cultivated without inorganic fertilizers (1). Excessive fertilizer application leads to salt burn and in most cases leads to the death of young plants (26). Because they are non-biodegradable, long term use of inorganic fertilizers result in accumulation of harmful substances and acidification of the soil thereby causing a decrease in the fertility of the soil (27). Due to their high solubility in water, inorganic fertilizers applied to the soil could be leached deep into the soil (where plant roots cannot reach) and into underground water causing pollution (1). According to Ifokwe (1), different types of fertilizers are suitable for different soil types. To get fertilizers which will suit a particular soil, the soil needs to be analyzed. According to him, most of the fertilizers imported into the country are not suitable for our soil thereby giving negative rather than positive results; besides one requires a good knowledge before applying it but today, every illiterate farmer applies fertilizers without understanding how it works and its side effects. All these

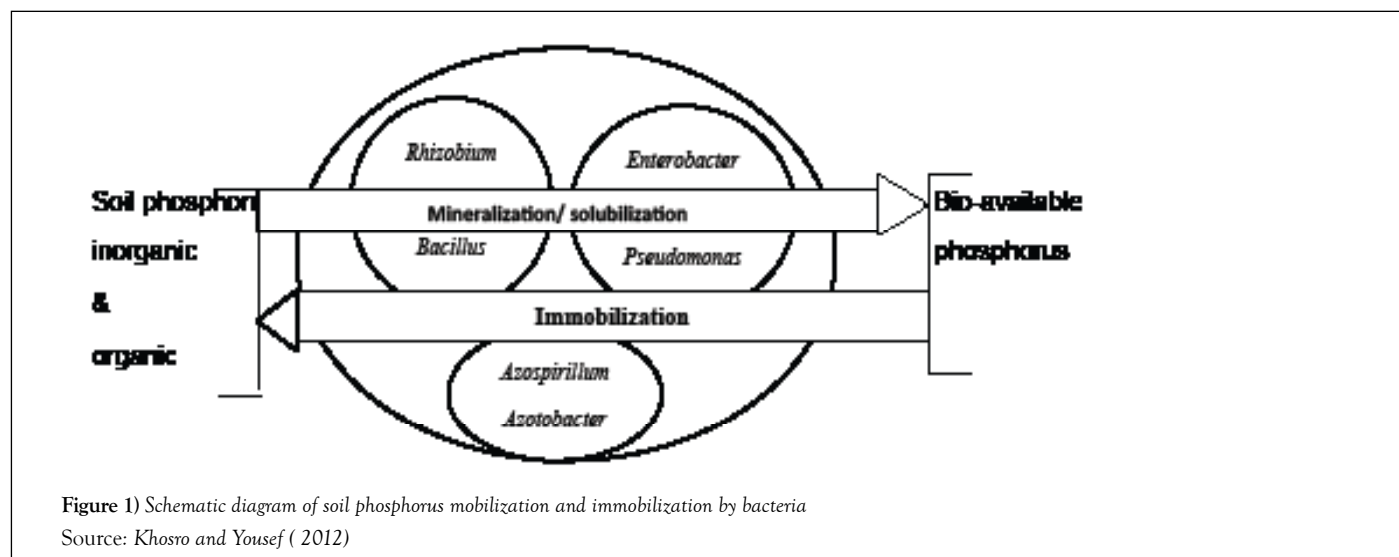


TABLE 1
Mean Nutrient Requirement of Plants

Element	Symbol	Mg/kg	Percent	Relative Number of Atom
Nitrogen	N	15,000	1.5	1, 000, 000
Potassium	K	10, 000	1	250, 000
Calcium	Ca	5, 000	0.5	125, 000
Magnesium	Mg	2, 000	0.2	80, 000
Phosphorus	P	2, 000	0.2	60, 000
Sulfur	S	1, 000	0.1	30, 000
Chlorine	Cl	100	-	3, 000
Iron	Fe	100	-	2, 000
Boron	B	20	-	2, 000
Manganese	Mn	50	-	2, 000
Zinc	Zn	60	-	1, 000
Copper	Cu	6	-	300
Molybdenum	Mo	1	-	1
Nickel	Ni	1	-	1

Source: Barak, 1999

TABLE 2
Different Microorganisms used in Bio-fertilizer Production

Groups	Examples
	Nitrogen fixing bio-fertilizers
Free-living	<i>Azotobacter, Bejerinkia, Clostridium, Klebsiella, Anabaena, Nostoc</i>
Symbiotic	<i>Rhizobium, Frankia, Anabaena, Azollae</i>
Associative symbiotic	<i>Azospirillum</i>
	Phosphate solubilizing bio-fertilizer
Bacteria	<i>Bacillus megaterium var, Phosphaticum, Bacillus subtilis, Bacillus circulans</i>
Fungi	<i>Penicillum Spp. Aspergillus awamori</i>
	Phosphate mobilizing bio-fertilizers
Arbuscular Mycorrhiza	<i>Glomus Spp., Gigaspora Spp., Acaulospora Spp.</i> <i>Scutellospora Spp. and Sclerocystis Spp.</i>
Ectomycorrhiza	<i>Laccaria Spp. Pisolithus Spp, Boletus Spp. and Amanita Spp.</i>
Ericoid Mycorrhiza	<i>Peizizella ericae</i>
Orchid Mycorrhiza	<i>Rhizoctonia solani</i>
	Bio-fertilizers for micronutrients
<i>Bacillus Spp</i>	Silicate and zinc solubilizers
	Plant growth promoting Rhizobacteria
<i>Pseudomonas</i>	<i>Pseudomonas fluorescens</i>

Source: Ritika and Uptal (2014)

problems can however be avoided by the use of indigenous fertilizers which is environment friendly. These fertilizers known as bio-fertilizers can achieve all that is achievable with inorganic fertilizers and even more without any side effects (1).

Bio-fertilizers are environment friendly and do not cause pollution unlike inorganic fertilizers which often 'run off' into water bodies causing eutrophication and 'blue baby syndrome' (acquired methemoglobinemia) when the nitrate level is above 10 mg/L (28). The issue of excessive application does not arise in the use of bio-fertilizer and special skills are not required for its application (1). Bio-fertilizers have long lasting effects due to their slow nutrient release. The nutrients from bio-fertilizers are released to plants slowly and steadily for more than one season. As a result, long term use of bio-fertilizer leads to the buildup of nutrients in the soil thereby increasing the overall soil fertility. In addition, bio-fertilizers have been found to help control of plant diseases such as pythium root rot, rhizoctonia root rot, chill wilt and parasitic nematode (24). Research has shown that some bio-fertilizers particularly those made with degraded tree barks and roots release chemicals that inhibit some plant pathogens. Disease control with bio-fertilizer has been attributed to four possible mechanisms: Successful competition for nutrients by beneficial microorganisms present in the fertilizer, production of antibiotics by the beneficial microorganisms, successful predation against pathogens by beneficial microorganisms, activation of disease resistant genes in plants by the microorganisms.

Bio-fertilizer acts as a soil conditioner adding organic matter to the soil which helps to bind the soil particles together preventing soil eructing, desertification, and erosion while increasing the water retention capacity of the soil (16). It enriches the soil with beneficial microorganisms while boosting the already existing ones unlike chemical inorganic fertilizers which acidify the soil making it hard for microorganisms to survive (16). Bio-fertilizers contain a wide range of nutrients which are often absent in inorganic fertilizers (these include trace elements). Studies have shown that application of nitrogen fertilizer in some weather conditions cause emission of nitrous oxide which has a global warming effect potential 296 higher times than that of an equal mass of carbon dioxide (29,30).

Methane emissions from crop fields (notably rice paddy fields) are increased by the application of ammonium based fertilizers whereas the composting of animal waste in a confined place or in an anaerobic condition (an important process in the production of bio-fertilizer), reduces the addition of methane to the atmosphere as these add methane to the atmosphere when left to decay on their own. Bio-fertilizer when compared to raw (undegraded) organic manure has the advantage of easier assimilation by plants and also the odor reduces after degradation (16). Again, the risk with raw organic manure is that it may contain pathogens such as *Salmonella* Spp. which may contaminate crops such as leafy vegetables and lead to the ingestion of the pathogen when the product is consumed. Bio-fertilizer also contains useful microorganisms which may not be present in organic (degraded) fertilizer (3). These bio-fertilizers can be produced from cheap waste materials which are abundant in Nigeria and the cost of production is low compared to inorganic fertilizers which required high energy.

DIFFERENCES BETWEEN BIO-FERTILIZER AND ORGANIC FERTILIZER

Before now, the term bio-fertilizer was used to include organic fertilizer. However, technically, there is a big difference between them. Vishal and Abhishek (31) in an attempt to distinguish between bio-fertilizer and organic fertilizer said "bio-fertilizers are microbial inoculants consisting of living cells of microorganisms like bacteria, algae, fungi, alone or a combination which may help in increasing crop productivity. Biological activities are markedly enhanced by microbial interactions in the rhizosphere of plants. Organic fertilizers on the other hand are obtained from animal sources such as animal manure or plant sources like green manure.

TYPES OF BIO-FERTILIZERS AND HOW THEY WORK

Bio-fertilizers are classified into different types depending on the type or group of microorganisms they contain. Table 2 shows the classification of bio-fertilizers on the bases of the different types of microorganisms used. The different types of bio-fertilizers include:

- **Nitrogen fixing bio-fertilizers (NFB):** Examples include *Rhizobium* Spp., *Azospirillum* Spp. and blue-green algae; these work by fixing atmospheric nitrogen and converting them to organic (plant usable) forms in the soil and root nodules of legumes, thereby making them available to plants. Nitrogen fixing bio-fertilizers are crop specific bio-fertilizers (32).

- **Phosphate solubilizing bio-fertilizer (PSB):** Examples include *Bacillus* Spp., *Pseudomonas* Spp. and *Aspergillus* Spp. These work by solubilizing the insoluble forms of phosphate in the soil, so that plants can use them. Phosphorus in the soil occurs mostly as insoluble phosphate which cannot be absorbed by plants (33). However, several soil bacteria and fungi possess the ability to convert these insoluble phosphates to their soluble forms. These organisms accomplish this by secreting organic acids which lower the pH of the soil and cause the dissolution of bound forms of phosphate making them available to plants (33).
- **Phosphate mobilizing bio-fertilizers (PMB):** Examples are Mycorrhiza. They work by scavenging phosphates from soil layers and mobilizing the insoluble phosphorus in the soil to which they are applied. Chang and Yang (34) stated that phosphorus solubilizing biofertilizer (PSB) sometimes act as phosphate mobilizers. Phosphate mobilizing bio-fertilizers are broad spectrum bio-fertilizers. Soil phosphorus mobilization and immobilization by bacteria are shown in Figure 1.
- **Plant growth promoting bio-fertilizer (PGPB):** Examples of plant growth rhizobacteria are *Pseudomonas* Spp. etc: these work by producing hormones and anti-metabolites which promotes root growth, decomposition of organic matter which help in mineralization of the soil thereby increasing availability of nutrients and improving crop yield (3,35). PGPB are crop specific bio-fertilizers.
- **Potassium solubilizing bio-fertilizer (KSB):** Examples include *Bacillus* Spp. and *Aspergillus niger*. Potassium in the soil occurs mostly as silicate minerals which are inaccessible to plants. These minerals are made available only when they are slowly weathered or solubilized. Potassium solubilizing microorganisms solubilize silicates by producing organic acids which cause the decomposition of silicates and helps in the removal of metal ions thereby making them available to plants. Potassium solubilizing bio-fertilizers are broad spectrum bio-fertilizers.
- **Potassium mobilizing bio-fertilizer (KMB):** Example of potassium mobilizing bio-fertilizer is *Bacillus* Spp. These work by mobilizing the inaccessible forms of potassium (silicates) in the soil. Some phosphate solubilizing bio-fertilizers such as *Bacillus* Spp. and *Aspergillus* Spp. has been found to mobilize potassium and also solubilize phosphorus.
- **Sulfur oxidizing bio-fertilizer (SOB):** Example of sulfur oxidizing microorganism is *Thiobacillus* Spp. These work by oxidizing sulfur to sulfates which are usable by plants.

MICROORGANISMS USED IN BIO-FERTILIZER

Organisms that are commonly used as bio-fertilizers components include nitrogen fixers (N - fixers), potassium solubilizers (K - solubilizer) phosphorus solubilizer (P - solubilizer), phosphorus mobilizers (P - mobilizers), used solely or in combination with of fungi. Most of the bacteria used in bio-fertilizers have close relationship with plant roots. *Rhizobacterium* has symbiotic interaction with legume roots, and *Rhizobacteria* inhabit root surfaces or rhizosphere soil (3). The phospho-microorganisms mainly bacteria and fungi make insoluble phosphorus available to the plants (33). Several soil bacteria and few species of fungi possess the ability to covert insoluble phosphate in soil into soluble forms by secreting organic acids. These acids lower the soil pH and bring about the dissolution of bound forms of phosphate (33). While *Rhizobium*, blue-green algae, and *Azolla* are crop specific, bio-inoculants such as *Azotobacter*, *Azospirillum*, phosphorus solubilizing bacteria (PSB), and Vesicular Arbuscular Mycorrhiza (VAM) could be regarded as broad spectrum bio-fertilizers (33). VAM are fungi that are found associated with majority of agricultural crops and enhanced accumulation of plant nutrients (3,36).

It has been suggested that VAM stimulate plant by physiological effects or by reducing the severity of diseases caused by soil pathogens. Examples of free living nitrogen fixing bacteria are obligate anaerobes *Clostridium pasteurinum* obligate aerobes, facultative anaerobes, photosynthetic bacteria *Rhodobacter*, *cyanobacteria* (*Azotobacter*), and some *Methanogens*. The most commonly used K - solubilizer is *Bacillus mucilaginosus* while P-solubilizers are *Bacillus megaterium*, *Bacillus circulans*, *Bacillus subtilis* and *Pseudomonas straita* (33).

THE ROLE OF BIO-FERTILIZERS IN ENHANCING PLANT TOLERANCE TO ENVIRONMENTAL STRESS

Abiotic and biotic factors are the major constraints that affect the productivity of crops. Many tools of modern science have been widely applied for crop improvement under stress, of which PGPRs' role as bio-protectant has

become of paramount importance in this regard (37). According to Hussain et al. (38) *Rhizobium trifoli* inoculated with *Trifolium alexandrinum* showed higher biomass and increased number of nodulation under salinity stress condition. *Pseudomonas aeruginosa* has been shown to withstand biotic and abiotic stress. *Pseudomonas putida* RS-198 enhanced germination rate and several growth parameters including plant height, fresh weight and dry weight of cotton under conditions of alkaline and high salt via increasing the rate of uptake of K^+ , Mg^{2+} and Ca^{2+} and by decreasing the absorption of Na^+ . Few strains of *Pseudomonas* conferred plant tolerance via 2, 4-diacetylphloroglucinol. *Mycobacterium phlei* provides tolerance to high temperatures and salinity stress. It has been demonstrated by Ansari et al. (22) that inoculation of plants with Arbuscular mycorrhiza fungi also improves plant growth under salt stress. Ansari et al. (22) also observe that a root endophytic fungus *Piriformospora indica* was found to defend host plant against salt stress. Combination of AM fungi and Nitrogen fixing bacteria help the legume plants in overcoming drought stress. Application of *Pseudomonas* Spp. to basal plants under water stress improves their anti-oxidant and photosynthetic pigment content. *Pseudomonas* Spp. was found to have positive effect on the seedling growth and seed germination under water stress (39). Ruiz-Sanchez et al., (40) reported that the photosynthetic efficiency and the anti-oxidative response of rice plant subjected to drought stress were found to increase after inoculation of arbuscular mycorrhiza. The beneficial effect of mycorrhizae has also been reported under both the drought and saline conditions.

PGPRs as biological agents proved to be one of the alternatives to chemical agents for providing resistance against various pathogens' attacks. Apart from acting as growth promoting agents, they can provide resistance against pathogens by producing metabolites (41). *Bacillus subtilis* GBO can induce defence related path ways. *Bacillus subtilis* N11 along with mature compost was found to control *Fusarium* infestation on banana root. By and large, the exploitation of PGPRs was found to be very significant in managing spotted wilt viruses in tomato, cucumber mosaic, virus of tomato and pepper, and banana bunchy top virus. In some cases, it was observed that along with bacteria, mycorrhizae can also confer resistance against fungi pathogens and inhibit the growth of many root pathogens such as *R. solani* and *Pythium* Spp.

IMPORTANCE OF BIO-FERTILIZERS

Bio-fertilizers play an important role in improving fertility of the soil. In addition, their application to soil improves the structure of the soil and minimizes the sole use of chemical fertilizers. Under low land conditions, the application of blue green algae (BGA) plus *Azospirillum* proved significantly beneficial in improving yield of grain. Bio-fertilizers inoculation with *Azotobacter* and *Rhizobium* and Vesicular Arbuscular Mycorrhiza gave the highest increase in straw and grain yield of wheat plants with rock phosphate as phosphate fertilizer (20). *Azolla* is inexpensive, economical, eco-friendly, which provides benefit in terms of carbon and nitrogen enrichment of soil. It was recorded microorganisms such as *Bacillus subtilis*, *Thiobacillus thiooxidans* and *Saccharomyces* species can fix atmospheric nitrogen symbiotically and about 80-90% nitrogen demand could be supplied by soya bean through symbiosis (20).

Bio-control, a modern approach of disease management can be a significant role of bio-fertilizer in agriculture. *Trichoderma* based bio-fungicides has been found promising to control root rot of mung bean (20). Growth, yield and quality parameters of certain plants significantly increased with bio-fertilizers containing bacterial nitrogen fixers, phosphate and potassium solubilizing bacteria and microbial strains of some bacteria (3). The importances of biofertilizers are highlighted below: Secretion of plant growth hormones which help in plant growth, Protection of the plant against attack by pathogens, Improvement soil fertility, No special care is necessary while using bio-fertilizer, Reduction in the use of chemical fertilizers, Bio-fertilizers are cost effective compared to synthetic fertilizer, Promotes growth of plants, Bio-fertilizers restore the soil's natural, nutrient cycle and build soil organic matter and Bio-fertilizer provides protection against drought.

LIMITATION OF BIO-FERTILIZER

The most important limitation of bio-fertilizer is their nutrient content when compared to inorganic fertilizers. This might result to deficiency symptoms in plants grown with the bio-fertilizer. However, this problem can be curbed by the addition of substances such as bone meal (rich in phosphorus), wood ash (rich in potassium) or other substances of natural origin such as phosphate rock to enrich the fertilizer. Also the use of nutrient rich wastes such as palm wastes (rich in potassium), wood ash (rich in potassium also) in making bio-fertilizer can help to remedy the problem. Mahimairaja et al. (24) stated that the addition of phosphorus to wastes makes the bio-fertilizer more balanced and reduces nitrogen losses. Again storage of bio-fertilizer goes a

long way in affecting its efficacy. Even though bio-fertilizer has many positive aspects, its use can sometimes not lead to the expected positive results and this could be because of exposure to high temperature or hostile conditions before usage. Bio-fertilizer should be stored at room temperature or in cold storage conditions away from heat or direct sunlight and polythene bags used in packaging bio-fertilizer should be of low density grade with a thickness of about 50 -75 microns (42). Other constraints limiting the use of bio-fertilizer technology may be environmental, human resource, unawareness, unavailability of suitable strains, and unavailability of suitable carrier and so on (20). Short shelf life, lack of suitable carrier material, susceptibility to high temperature, problem in transportation, and storage are biofertilizers bottlenecks that still need to be solved in order to obtain effective inoculation (43).

- **Unavailability of suitable strain due to lack of availability of specific strain:** This is one of the major constrains in the production of bio-fertilizer. Based on the fact that selected strains have ability to survive both in the broth and the inoculants carrier.
- **Unavailability of suitable carrier:** If suitable carrier material is not available, it is difficult to maintain the shelf-life of the bio-fertilizer. As per suitability, the order is peat, lignite, charcoal, farmyard manure, soil, rice bran.
- **Lack of awareness among farmers:** Farmers are not aware of bio-fertilizers and their usefulness in increasing crop yields. They are unaware of the damages caused on the ecosystem by continuous application of inorganic fertilizer.
- **Inadequate human resources and inexperienced staff:** This is another problem. This is because the unskilled and the inadequate staff farmers are not given proper instruction about the application.
- **Environmental constraints:** Soil characteristics like salinity, acidity, drought; water logging affects the use of bio-fertilizers (20).

CAUTION IN THE USE OF BIO-FERTILIZERS

- Never mix bio-fertilizers with nitrogen fertilizers.
- Never apply bio-fertilizers with fungicides.
- Never expose bio-fertilizers to sunlight directly.
- Bio-fertilizers are stored at room temperature, not below 0 °C and above 35°C
- Do not keep used solution overnight (17).

CONCLUSION

Our dependence on chemical fertilizers and pesticides has encouraged the thriving of industries that are producing life-threatening chemicals which are not only hazardous for human consumption but can also disturb the ecological balance. In fact, attention is now shifting from consuming food grown with chemical fertilizers to food grown with organic fertilizers because of the harmful effects that these foods have in the body when consumed. Bio-fertilizers can help solve the problem of food need of the ever increasing global population. It is important to realize the useful aspects of bio-fertilizers so as to apply it in modern agricultural practice. The application of bio-fertilizers containing beneficial microbes promote to a large extent, crop productivity. These potential biological fertilizers would play a key role in productivity and sustainability of soil and protect the environment as eco-friendly and cost effective inputs for the farmers as rightly stated by Khosro and Yousef (3). Using the biological and organic fertilizers, a low input system can help to achieve sustainability of farming.

The new technology when developed using the tool of molecular biotechnology can enhance the biological pathways of production of phytohormones if identified and transferred to the useful plant growth promoting rhizobacteria. This technology will help provide relief from environmental stresses. However, the ignorance regarding improved protocols of bio-fertilizers application to the field is one of the few limiting factors to bio-fertilizers usage.

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