Koni	*Coniothyrium minitans*	*Sclerotinia* spp.		BIOVED Ltd., Hungary www.bioved.hu
Novacide	*Chaetomium cupreum*	*Fusarium oxysporum f.sp. lycopersici*	Powder	Nova Science, Thailand
Polygandron Polyversum	*Pythium oligandrum*	*Pythium ultimum*	Granule or powder	Plant Protection Institute, Slovak Republic
Prestop Primastop	*Gliocladium catenulatum* Strain JI446	*Pythium* spp. *Rhizoctonia solani* *Botrytis* spp. *Didymella* spp.	Wettable powder	Kemira Agro Oy, Fingland
Promote	*Trichoderma harzianum* *Trichoderma viride*	*Pythium* *Rhizoctonia* *Fusarium*		JH Biotech Inc., Ventura, CA, USA www.jhbiotech.com
RootShield PlantShield	*Trichoderma harzianum* Strain T-22	*Pythium, Rhizoctonia, Fusarium. Sclerotina*	Granules and Wettable powder	Bioworks, Inc. NY. USA www .bioworksbiocontrol.com
Sentinel®	*Trichoderma* spp. Strain LC52	*Botrytis cinera*	Wettable powder	Agrimms Technologies Ltd. www.vinevax.com
SoilGard (GlioGaid)	*Gliocladium virens* GL21	Several plant diseases Damping-off and root pathogens	Granules Alginate prill	Thermo Triology, USA.
Trichodex	*Trichoderma harzianum* T-39	Fungal diseases e.g. *Botrytis cinerea* *Colletotrichum, Monilinia laxa, Plasmospara viticola Rhizopus stolonifer*	Wettable powder	Makhteshim-Agan, DeCeuster, Belgium
Vinevax™	*Trichoderma* spp.	Wood-infecting fungal pathogens of vineyard, orchard, ornamental trees, and vines	Wettable powder	Agrimms Technologies Ltd, www.vinevax.com

3. Mycoinsecticides

There are some fungal species that can act as parasites of insects. The fungi which are used to attach and kill insects are called as mycoinsecticides. Use of fungi and bacteria as insecticides for control of harmful insects has been a part of integrated pest management. It is reported that approximately 750 species of fungi are pathogenic to insects. However around 12 species have been utilized as the insecticides.

(*i*) Insect-Pathogenic Fungi

One of the earliest discovered insect pathogenic fungus is *Beauveria bassiana* (Bb) which is found world-wide in soils and plants. Table 10.8 gives the list of some insect pathogenic fungi and their insect hosts.

TABLE 10.8. Insect-pathogenic fungi with their hosts

Fungi	Insects affected
Beauveria bassiana	White flies, aphids grasshoppers, termites, colorado potato beetle, Mexican been beetle, Japanese beetle, boll weevil, cereal leaf beetles, bark beetles, lygus bugs, chenchbug, fireants, coding moth, etc. (It is a broad range mycoinsecticide)
Eutomophthera muscae	Flies, onion maggots, cabbage maggots, seed corn maggots
Pandora neoaphids	Naturally occurring pathogen of aphids
Zoophthora radicans	Broad host range. Several caterpillars, leaf hoppers, aphids, weevils
Metarhizium anisopliae	Corn root worm, white grubs, root weevils, spittle bugs of sugarcane and alfalfa
Verticellium lecani	Green house white flies, trips and aphids
Neozygites floridana	Spider mites
Nomuraea rileyi	Green clover worm, army worms, corn ear worm, tobacco budworm
Hirsutella thumpsonii	Mites

(*ii*) Mode of Action

These fungi generally invade the insects by penetrating their cuticle or the skin. After entering into the body they rapidly multiply. Death is caused by tissue destruction and sometimes by toxins produced by the fungus. The fungus frequently emerges from the insect body to produce spores which spread by rain or wind or contact to other insects thereby affecting the insect population.

(*iii*) Mode of Applications and Effectiveness

They are applied as spore suspensions on the insects at proper dose recommendations. In some cases a combination of *B. brassiana* and *M. anisopliae* is given for effective control.

Where the larval stage of the insect are found in the soil, granular solution of the mycoinsecticides are applied.

The granular solutions are produced in a variety of ways. In one approach the bran or grains are coated with dry spores. In another method drying and fragmentation of mycelia is done to held spores in starch medium. The production of the fungus on whole kernels of grain followed by drying and pulverization of the kernel produces a good granular formulation.

The effectiveness of mycoinsecticides depends on the correct fungal species and strain with the susceptible insect life stage at the appropriate humidity, soil texture, and temperature.

The application of fungicides, herbicides kill or inhibit the growth of these mycoinsecticides. So during their application the use of fungicides, herbicides etc. should be avoided.

(*iv*) Commercially available mycoinsecticides

Now the mycoinsecticides have been produced in commercial scale and sold with specifications and recommendations.

(*a*) *Beauveria bassiana* strain GHA is sold under the trade name Mycotrol GHOF and Mycotrol GH-ES to control grasshoppers, locusts, and mormon crickets on rangeland, improved pastures, alfalfa, corn, cotton, potatoes, rapeseed, sunflowers, etc.

(*b*) *M. anisophiae* is registered mycoinsecticide on US for control of household cockroaches.

(*c*) *Paecilomyces fumosoroseus* Apopka strain 97 has been approved for use on ornamentals, nonfood crops in greenhouses, aphids, trips, spider mites, etc.

(*d*) A new strain of *M. anisophiae* called ARSEF-49 has been developed by genetic engineering which is very effective. It contains the gene AalT which gets activated once the fungus enters the bloodstream of the insect.

(*v*) Advantages of Mycoinsecticides

(*a*) The fungus does not grow in warm blooded organisms or it survives long in water bodies.

(*b*) Spores can withstand long periods of dryness and other harsh environmental conditions.

(*c*) it does not hurt plants.

(*d*) they are non-poisonous.

(*e*) insects do not develop resistance to these quickly.

Inspite of this, the mycoinsecticides have not yet replaced the chemical insecticides. The rapid increase in pest population is sometimes difficult to control by this group as they take sometime to proliferate in the body. Besides, the infectivity is moderate and some times inconsistent.

It requires still more research in this field to increase the virulence of the fungal species, and make it economical.

4. Myconematicides

Plant parasitic rematodes cause serious economic loss to the agriculture. Control of nematodes by chemical nematicides although successful, they have great environmental hazards. Besides, the nematodes develop resistance to chemically and the chemicals persist in the environment. Therefore biocontrol of nematodes is an eco-friendly approach. Use of fungi as bicontrol agents against nematodes was first suggested by Lohde in 1874 but proper organized research started in 1960s. By now nearly 200 species of soil fungi are known to have potentials in controlling nematode population either parasitizing them or eating them directly or killing them through the substances they secret. Those fungi which are able to kill and control nematodes are called as **myconematicides.**

The nematophagus fungi can be broadly grouped into three groups :

(*a*) **Predacious or nematode trapping fungi**—which produce mechanical or adhesive traps,

(*b*) **Endoparasitic fungi or endozoic group**—which infect nematodes when their conidia (spores) are ingested or adhere to the cuticle,

(*c*) **Opportunistic fungi or egg parasites**—which attach the female nematodes or their eggs when they come in contact with them.

The different fungi on these three groups used in nematode control are given in the Table 10.9.

TABLE 10.9. List of fungi used as nematicides

Predacious fungal genera	Endozoic fungal genera	Opportunistic fungal genera
Arthrobotrys	*Meria*	*Cylindrocarpum*
Dactylella	*Cutenaria*	*Fusarium*
Monacrosporium	*Nemaptophthera*	*Exophiala*
Geniculifera	*Hirsutella*	*Gliocladium*
	Harposporium	*Paecilomyces*
	Haptoglossa	*Phoma*
	Myzocytium	*Verticilium*

(*a*) **Nematode trapping fungi.** They are mostly the genera belonging to Moniliaceae of Deuteromycotina. Such trapping fungi develop some trapping structures like adhesive networks, adhesive knob and non-constricting or constricting rings. They capture the nematode prey by these devices that they develop and cover the entire surface of the nematode.

The species like *Arthrobotrys arthrobotryoides, A. conoides, A. oligospora. Dectyllella lobuta* when added to the field mixed with organic substances reduce the root galling by nematode *Meloidogynae hapla* in red pepper.

D. oviparasitica is successful against *Meloidogyne* spp. also *D. megalospora* which both trapp and absorb the nematodes is found effective against 18 genera of plant parasitic nematodes.

Monacrosporium ellipsosporium helps in controlling the root knot of tomato in the green home and field conditions.

(*b*) **Endoparasitic Fungi.** Species of *Hirsutella, Acrostallagmus, Harposporium, Meric* belonging to Hypomycetes are included in this group. The zoosporic forms of endoparasitic fungi are species of *Catenaria, Myzocytium, Haptoglossa*, etc.

The zoosporic fungi *Catenaria auxilians* and *Nematophthora gynophila* are used for control of parasitic nematodes. *Hirsutella heterodevae* acts against the sugarcane beet cyst nematode.

Although 50 species of this group of fungi have been identified having the nematicidal activities, the difficulties in obtaining masses of propagules, the endozoic fungi are rarely added to the soil for the purpose.

(*c*) **Opportunistic fungi or egg parasites.** These fungi usually live in the soil on saprophytes. When they come in contact with the nematodes they parasitize them. Generally these fungi affect the female nematodes and the eggs. Out of several fungi of the group the most commonly used myconematicide fungus is *Paecilomyces lilacinus* and *Verticillium chlamydosporium*. These fungi are studied thoroughly and they are found against a wide range of nematodes which attack nearly 90% of the food and fiber crops.

The spores of *Paecilomyces lilacinus* grown on rice halls and rice bran mixture or on chopped water lily are collected and incorporated into the soil. It controls the nematodes in corn, tomato, okra crops. Later a strain of this fungus called *P. lilacinus* strain 251 is developed which is very effective against a wide range of nematodes. It is sold under the brand name **Paecilo** commercially.

Now commercial preparations of these myconematicides are available in many countries. Some examples are Royal 300 (contains *Arthrobotrys robusta*), Royal 350 (contains *Arthrobotrys superba*) which act against root knot nematode. Real *Trichoderma asperillum* isolate TR 900 is unique Real I PM and is most cost effective. It acts both as nematicide and fungicide.

Although several fungi have shown nematicidal activities, few are practically used in the field and for limited crops. Therefore chemicals obtained from these fungi are isolated and are also used as bicontrol agents for nematodes in the field in preference to chemical nematicides. It is hoped further research in this field will develop effective myconematicides commercially for wide use in future.

10.8. FUNGI AS BIOFERTILIZER

Biofertilizer is defined as a substance which contains living microorganism which when applied to seeds, plant surfaces or soil colonize the rhizosphere or the interior of the plant and promotes growth by increasing the supply or availability of primary nutrients to the host plant. There are also other definitions to a biofertilizer. In general biofertilizers comprise microbial inocula or assemblages of living microorganisms which exert direct or indirect benefits on plant growth and crop yield through different mechanisms.

These organisms are able to fix atmospheric nitrogen or solubilize phosphorus, decompose organic material or oxidize sulphur in the soil that are beneficial to agricultural production interms of nutrient supply.

Uses of cyanobacteria, and bacteria are common as biofertilizers used mainly for increasing the nitrogen fertility of soil. Fungi also contribute to the plant growth and yield as biofertilizers.

(*a*) Mycorhizal fungi used as biofertilizers

The mycorrhizal fungi (or called mycorrhizae) from mutualistic symbiotic relationships with plant roots of more than 80% of land plants excluding the crops and forest tree species. The two dominant types or mycorrhizae called **ectomycorrhizae** (ECM) and **arbuscular mycorrhizae** are very helpful to the plants growth and development for which they are used as biofertilizers.

Ecotomycorrhizal fungi (ECM) do not penetrate the living cells of the roots but only surround them. Most of the ECM fungi found associated with the forest trees are members of Basidiomycetes. The common members are species of *Amanita, Lactarius, Pisolithus, Rhizopogon*. Some ascomycetan members are the species of *Cenococcum, Elaphomyces, Tuber*, etc. They help the plants in the following ways :

(*i*) Absorb and accumulate nitrogen, phosphorus, potassium, calcium more rapidly than the normal root,

(*ii*) Break down complex minerals and organic substances in the soil,

(*iii*) Increase the tolerance of the plants to drought, high soil temperature, soil toxins, soil pH conditions,

(*iv*) Protect the roots from other pathogens.

The most common ECM fungi used as biofertilizers are—*Pisolithus tinctorius* and *Piriformospora indica*. The inocula of these fungi are applied as vegetative mycelia in a peat vermiculite carrier.

The **endomycorrhizal fungi (EM)** are intercellular and penetrate the root cortical cells. They form vesicle like structures called **arbuscular vesicles for which they are known as vesicular arbuscular mycorrhiza (VAM).** In some cases these vesicles are not found and these fungi are called *asarbuscular mycorrhizae* (AM). The agriculturally produced crop plants that form endomycorrhizae of the VAM type are now called as arbuscular mycorrhizae (AM). Such AM fungi are :

Acaulospora	*Archaespora*	*Enterophospora*
Gerdemannia	*Geosiphon*	*Gigaspora*
Glomus	*Paraglomus*	*Seutellospora*

These fungi play an important role in plant growth, health and productivity. They help the plant in the following ways :

(*i*) absorb nutrients especially the less available nutrients like copper, molybdenum, zinc, phosphorus,

(*ii*) increase seedling tolerance,

(*iii*) improve soil structure,

(*iv*) increase herbivory tolerance, increase pollination, increase soil stability, and heavy metal tolerance to the plants.

They also provide protection against soil borne pathogens and provide resistance against root rot diseases. They do it by competitive colonization or through competition for nutrients or by production of fungistatic compounds.

VAM fungi have been used as biofertilizers for more than 30 years in agriculture, horticulture, landscape restoration, soil remediation. Mass production of AM fungi has been done with several species—

Acaulospora laevis, Glomus clarum, G. etunicatum, G. intraradices, Gigaspora ramisporophora, G. rosea, etc. But *Glomus intraradius* is the most common inoculum of EM products.

(*b*) Production of Mycorrhizal Biofertilizers

VAM fungi are obligate (or strict) symbionts. Therefore they cannot be grown in synthetic media without the host for which separate procedures are followed for their mass production.

(*i*) Root based bulk inoculum production technology utilizes mass production of seedlings that are grown in sterilized soil infected with the selected fungi using spores from fruiting bodies from cultivated plants. The seedlings root system get infected with the fungi. These infected roots along with the adhering soils are collected, chopped up and used as the starter inoculum for scale up productions.

The inocula are produced in bulk by infecting fresh seedlings of the selected plants. The root inocula are kept in polythene bags and used for pelleting seeds or in the preparation of granules for seed bed inoculation.

(*ii*) Other methods like soil-free aeroponic, nutrient film, root organ culture systems are also developed for production of such biofertilizers. But they are not cost effective hence not alluring for commercial productions.

(*iii*) Tissue culture techniques may also be followed. Plants may be produced in mass in tissue culture in sterile agar media. The mycorrhizal; fungi may be induced using spores from fruiting bodies of the selected mycorrhizal fungi. The dried root tissues and fungal mycelia could then be developed into mycorrhizal seedling product.

The mycorrhizal fungi which are selected for production of biofertilizer in commercial scale must colonize roots rapidly after inoculation; absorb phosphate from soil, transfer phosphorus to plants, increase plant growth, persist in soil and reestablish again during following seasons and form propagules that remain viable during and after inoculum production.

The success of formulation depends on whether

- it is economically viable to produce,
- it does not alter the viability and function of the inoculum
- it is easy to carry and enhance dispersal during application.

The inoculum formulation may contain one or more mycorrhizal fungi with other organisms which together enhance the ability of the inoculum to form mycorrhizal association with the target plant.

The formulation are available in different form—as powder, tablets or pellets, granules, gel beads or balls.

The applications methods vary depending on the formulation. They may be used by scattering by hand, in furrow application, seed coating, root dipping or seed inoculation.

Commercially available mycorrhizal biofertilizers. Commercial mycorrhizal biofertilizers have evolved in recent years due to advances in technology. More than 30 companies all over the world are producing and marketing these biofertilizers (Table 10.10) which are used in horticulture, agriculture, forestry and restorations.

(c) Other fungi used as biofertilizers

Some species of *Penicillium, Aspergillus, Chaetonium and Trichoderma* have been used for production of fungal biofertilizers also in commercial scales.

(*i*) *Penicillium bilaiae* has been formulated as a commercial product named Jumpstart and it is a wettable powder released in 1999 to the market. It increases dry matter, phosphorus uptake, and seed yield in canola (*Brassica napus*).

There are two other species of *Penicillium* such as *P. radicum* and *P. italicum*. *P. radicum*, isolated from wheat rhizosphore, promotes plant growth. *P. italicum* is able to solubilise tri-calcium phosphate and promotes growth in soybean.

(*ii*) *Aspergillus flavus, A. niger, A. terreus* are reported to be involved in solubilization of phosphates through the production of various organic acids like—citric, oxalic, gluconic, glycolic acids. So they are now useful as biofertilizer.

(*iii*) *Chaetomium globusum* and *Chaetomium cupreum* products are also prepared for use as biofertilizer. The formulation is available under the name ketomium. This acts as a mycofungicide and also as a growth stimulant in tomato, corn, rice, pepper, citrus, plants.

(*iv*) *Trichoderma* spp. are known for their fungicidal activities and used as mycofungicides. But some have dual roles : as mycofungicide and biofertilizer. They increase mineral uptake, release minerals from soil and organic matters, enhance plant hormone production, induce systematic resistance mechanisms, induce root system in plants. Some of the *Trichoderma* products used as biofertizer has been given in the Table 10.10.

There is no doubt that mycorrhizal fungi help the plants in various ways. But there is a controversy as to whether they can be called as biofertilizer or not ? A group of scientists do not prefer to call mycorrhizal fungi as biofertilizer as the fungal symbiont in the relationship is a consumer instead of being a producer of nutritive elements. It does not bring anything from outside (like the N_2 fixing organisms). It only promotes the soil environment for the benefit of the plant. They prefer to call mycorrhizae as plant growth promoting organism. Whatever the controversy may be, all agree that mycorrhizae are beneficial to the plant growth and yield.

TABLE 10.10. Some fungal biofetilizers available globally

Products	Fungi	Companies
AgBio-Endos	Ectomycorrhizal fungi	AgBio Inc. Westminter, USA
AgBio-Ectos	Endomycorrhizal fungi	Agbio-inc.com
AM 120	Mycorrhizal fungi	Reforestation Technologies International, USA www.reforest.com
Bioorganic Plus	*Trichoderma harzianum* *Trichoderma hamatum*	NovaScience Co. Ltd, Thailand.
BioVam	Mycorrhizal fungi *Trichoderma* spp.	T&J Enterprises, USA www.tandjenterprises.com
BuRize	AM fungi	BioScientific Inc, Arizona, USA www.biosci.com
Diehard™ mycorrhizal inoculant	Mycorrhizal fungi *Trichoderma* spp.	Horticultural Alliance, Inc, Fl. USA www.horticulturalalliance.com
Endomycorrhizal inoculant (BEI), inoculant micronized (BEIM), Mycorrhizal root dip	Endomycorrhizal fungi	Bio-Organics, Oregon, USA www.bio-organics.com
MycoApply® Endo MycoApply® Endo/Ecto MycoApply® Maxx	Ectomycorrhizal fungi Endomycorrhizal fungi	Mycorrhizal application Inc, Oregon, USA www.mycorrhizae.com
Plant Success™ Mycogrow™	Ectomycorrhizal fungi Endomycorrhizal fungi	Fungi perfecti, LLC, WA., USA www.fungi.com
Mycomax	AM fungi (*Glomous intraradices*)	JHBiotech Inc. California, USA www.jhbiotech.com
Myke	Mycorrhizal fungi	Premier Tech Biotechnologies, Canada