Arbuscular Mycorrhizal Fungi (AMF): A Natural Companion of Plants against Biotic Stress; A Review

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Abstract:

Arbuscular mycorrhizal fungi (AMF) are a type of dominant obligate symbionts that establish their relationship with roots of the plants. Arbuscular Mycorrhizal Fungi have significant roles in the mycorrhizosphere. More or less about 71% of the vascular plant are in microsymbiont associations, where AMF solely relies on host photosynthetic phenomenon. As well as, they induced the tolerance ability of plants against the biotic stresses like viral, fungal, herbivores as well as bacteria. This mechanism involves competition for nutrients, space, rhizosphere alternation and host photosynthates to induce the defense in the host. Moreover, the effectiveness of AMF in tolerating the biotic stresses primarily depends on the interaction between host plant and culture systems through receptors after AMF colonization. This review designates the induced defense mechanism, factors, receptors, their particular functions and pathways that are mediated by AMF to induce tolerance against biotic stresses.

Keywords: Arbuscular mycorrhizal fungi; symbiosis; biotic stress; defense; receptors

1. Introduction

Plants being an essential and significant part of the environment depends mostly on the natural rhizosphere microbiome for proper development and growth [1]. Plants develop relations with the rhizosphere microbiome to aid the natural plant defense mechanism against biotic stresses. Biotic stresses involve pest, virus, fungal and bacterial attacks that lead to diseases in the plants [2]. One of the relations that the plant has developed is Arbuscular mycorrhizal fungi (AMF). Arbuscular mycorrhizal fungi (AMF) form mutualistic relationships with the roots of most plants, and are a group of soil-borne fungi [3]. These are the fungi that colonize the roots of plants and extend hyphae (thread-like structures) into the surrounding soil, where they are able to access all the nutrients that the plant roots are unable to reach themselves. As a mutualistic relationship is established, in return for the nutrients, the plant provides the AMF with carbohydrates, mostly sugars that are produced through photosynthesis [4]. Moreover, AMF are considered to be one of the former and the most widespread plant-microbe symbioses on the planet Earth, with fossil evidence dating back to the Devonian period. These are found in almost all terrestrial ecosystems, including forests, grasslands, deserts and agricultural systems [5].

AMF provides benefits to the plants, by extending the reach of the plant's roots, fungi are able to increase the plant's access to nutrients such as phosphorus, which is often limited in soil. The uptake of plant nutrients had also been improved by colonization of AMF such as potassium, zinc and copper. Additionally, AMF are able to protect the plant from pathogens by producing compounds that inhibit growth of harmful bacteria, pests and fungi [6]. AMF are also important for soil health. They help to refine soil structure by enhancing the amount of organic matter as well as increasing the soil's water-holding capacity. It also plays a key role in the cycling of nutrients, making them available for plants and other organisms. In agriculture, AMF has significance as it can be used as a sustainable replacement to the chemical fertilizers. They have been observed to improve crop yields and to reduce the need for chemical inputs.

Additionally, plants grown with AMF have been shown to be more resilient to environmental stressors such as drought. There are many different species of AMF, each with their own unique characteristics [7]. Moreover, research is ongoing to identify the most effective species for specific crops and ecosystems. Therefore, AMF are beneficial for plants, improving their access to essential nutrients and protecting them from pathogens. They also play a vital role in soil health and have potential for use in sustainable agriculture [8]. The symbiotic relationship of AMF and plants are shown in figure:

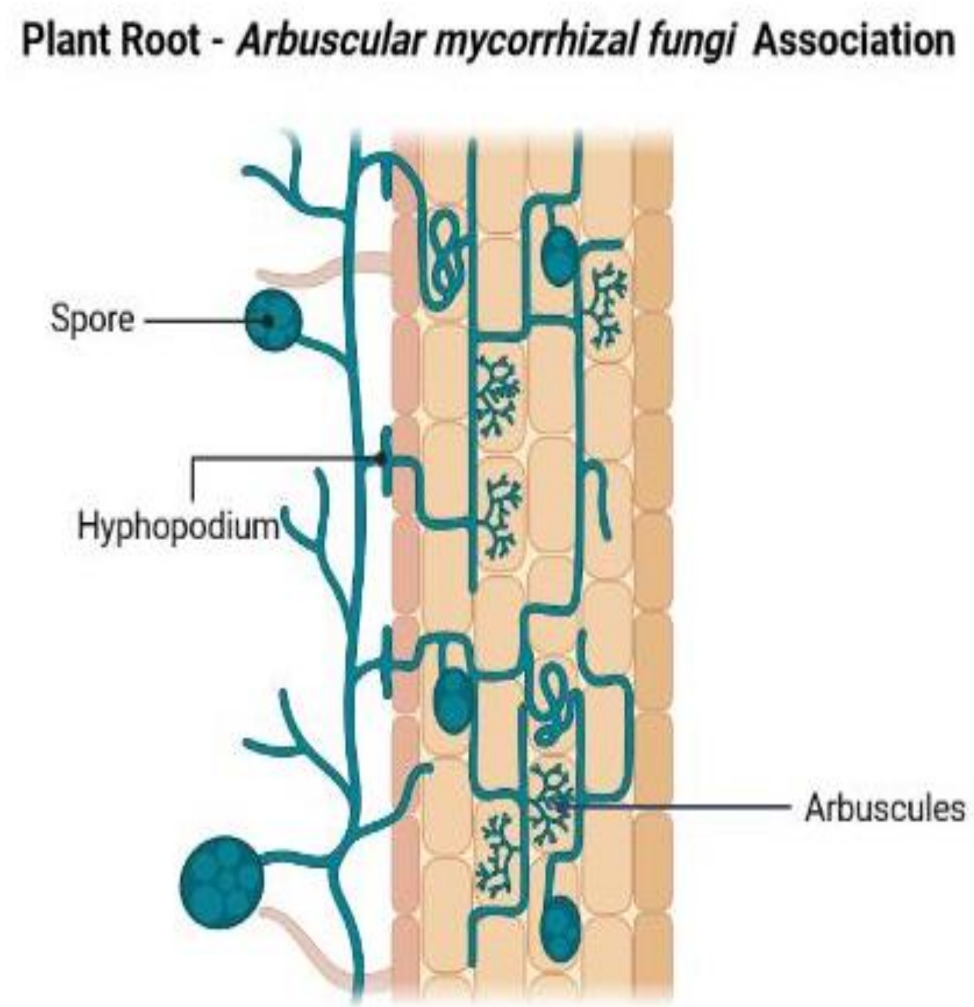

Figure 1: Root with Arbuscular mycorrhizal fungi association

2. Role of AM Fungi as Biocontrol agent

Some approaches that are used particularly for biotic control involve use of proteins such as Cry1Ac and Cry2A [9]. On the other hand, AMF has been playing a vital role in biotic stress management and has gained prime importance. Recent studies reported that damage to plants has been controlled by microsymbionts associations [10]. For example, the authors summarize their findings that AMF reduced the root-rot disease caused by severity of charcoal in soyabean. Mycorrhiza-induced resistance (MIR) has been induced by colonization of AMF. Moreover, the production of antioxidant enzymes provides oxidative stress and aids in the defense mechanism of the plant by downregulating lipoxygenase enzyme [11]. Also regulates the aquaporin gene, transcription factors and ABA responsive gene that are involved in the expression. It improves the nutrient uptake by translocating from the external environment into the internal environment, soil moisture, level of fertility and quality of soil which finally leads to the improved developmental growth of symbiotic plants [12].

These induced defense mechanisms proved to be helpful in the mycorrhizosphere. Therefore, it should also be stated that the beneficial effects of AMF and its effectiveness solely relies on interactions between host plant and culture systems [13]. The interactions that occur between them was established through certain receptor substrate bindings [14]. The particular gene expresses a particular receptor for further interactions. Few of the genes that take part in the defense mechanism of plants after colonization of AMF have been mentioned down below in a table.

Table 1: Genes that are involved in defense against biotic stresses / pathogens after AMF’S colonization

Sr. No.	Name of Genes	The product they form	Best Source	Related Function they perform
1.	*TC101060*	Protein with more Cysteine content	M. truncatula	Activity against fungus
2.	*TC104515*	More Cysteine content in protein	M. truncatula	Activity against fungus
3.	*TC98064*	Cysteine rich protein	*Medicago truncatula*	Activity against fungus
4.	*PR-1a*	PR-1a protein	Tomato	Pathogenesis-related (Antimicrobial)

			(Solanum lycopersicum)	
5.	*BGL*	β 1,3 Glucanase (Belong to PR family of protein)	Tomato (*Solanum lycopersicum)*	Antifungal property
6.	*VCH3*	Protein Chitinase	*Vitis amurensis.* (Grapevine)	Antifungal property against Meloidogyne incognita
7.	*Ltp*	PLTP (Protein transfers lipids)	Rice *(Oryza sativa)*	Having role against microbes
8.	*Pal*	(PAL) enzyme (phenylalanine ammonia lyase)	Rice *(Oryza sativa)*	Substances like phytoalexins and phenolic are produced
9.	*PR10*	PR 10	Pea *(Pisum sativum)*; Parsley	Pathogenesis-related protein (have RNases activity)
10.	*pI 49*	pI 49	Pea *(Pisum sativum)*	Member of multigene family PR 10 (have Ribonuclease activity)
11.	*pI 176*	pI 176	Pea *(Pisum sativum)*	Involved in the activity of ribonuclease, belongs to the PR 10 a multigene family

12.	*pI 206*	pI 206	Pea *(Pisum sativum)*	Role in defense against plant pathogen interaction
13.	Unknown	Chalcone isomerase	Pea *(Pisum sativum)*	Involved in the phytoalexins production
14.	Unknown	Transcinnamic acid 4-hydroxylase	Pea *(Pisum sativum)*	Involved in the phytoalexins production
15.	Unknown	Basic A1-chitinase	Pea *(Pisum sativum)*	Having vacuolar compartmentation and involved in the activity against fungus

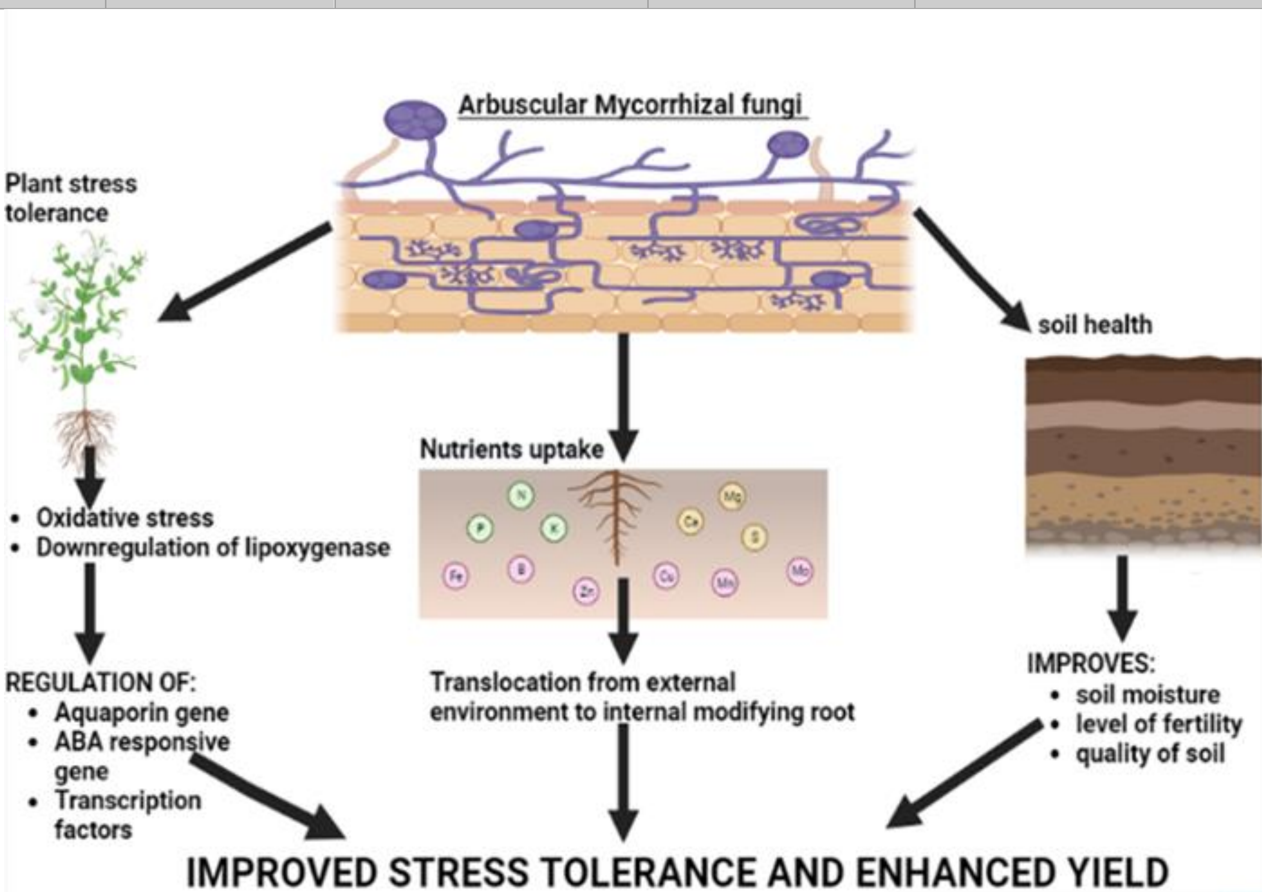

Figure 2: AMF association with plant for improved stress tolerance and enhanced yield

2.1. Arbuscular mycorrhizal fungi prevent plant from fungal infections

Plant pathogens affect plants and cause economic as well as yield losses. Among the plant pathogen fungi is one of the foes of plants [15]. They are one of the most prominent threats to cause disease in plants. They adopt different strategies to infect their host and cause diseases in them. Different strategies could be adopted to control the fungal virulence against plants [16]. Many chemical and biological methods have been applied till date for their control. Recently arbuscular mycorrhiza has shown to impart beneficial effects on plants which help it in its defense against fungal attacks and infections [17]. As evidenced by work of Campo et al., which performed research on rice varieties and inoculated it with *Funneliformis mosseae* and *Rhizophagus irregularis*. These AMF showed beneficial symbiotic relationship and not only enhanced productivity but also confer resistance against rice blast fungus *Magnaporthe oryzae.* Hence protecting it from infection.

Similar results were shown by Tian and his team which conducted experiments on rice using different species of AMF and reported resistance against *Magnaporthe oryzae*. Beneficial effects of AMF are also evident by the work of Berdeni et al. which conducted experiments on apple *(Malus pumila)* plants in symbiosis with beneficial fungi. The colonized AMF not only improved growth of apple plants but conferred the resistance against the canker disease too mediated by fungal pathogens [18]. While AMF proved to be beneficial in plant's defense against fungal disease but it could be genome specific as shown by researchers in China conducting experiments on different varieties of tomato against Fusarium wilt [19]. They reported that the protective role of mycorrhiza was evident in wild type, it was not prominent in mutated cultivar of tomato, hence suggesting that AMF mediated disease protection could be genome-specific property depending on regulation of certain metabolic pathway i.e., Jasmonic acid signaling pathway [20]. Better defense and resistance to pathogen fungi was shown by red sage plant *(Salvia miltiorrhiza)* of China when inoculated with *Glomus versiforme*. Plant showed enhanced biomass production and better photosynthetic capacity when grown with fungi and also it displayed better protein production and enhanced expression of defense related genes in response to infection with *Fusarium oxysporum.* A study conducted on banana seedlings inoculated with AMF also showed increased defense against wilt disease. Moreover, the productivity and growth of plants was also considerably increased [21]. Association of plants with arbuscular mycorrhizae fungi can cause greater expression of antioxidant pathways which in turn helps alleviate stress induced by pathogens such as *Verticillium dahliae* which may cause verticillium wilt in artichoke [22]. The role of AMF to confer resistance against black leaf stack disease (BLSD) has been reported by Gbongue et al. by inducing silicon uptake AMF *Rhizophagus irregularis* decreases the chances of banana acquiring BLSD. Protection of potato, pepper and tobacco by species of AMF has also been reported. Beneficial microbial colonies can help plants against *Phytophthora* spp. such as, *Phytophthora infestans*, *Phytophthora nicotianae* and *Phytophthora capsica* [23].

Table 2: AMF against pathogenic fungi

AMF species	Host plant	Protection against fungi	References
Funneliformis mosseae, Rhizophagus irregularis	Rice *(Oryza sativa)*	Blast fungus (*Magnaporthe oryzae)*	[21]
Rhizoglomus intraradices	Rice *(Oryza sativa)*	Blast fungus (*Magnaporthe oryzae)*	[8]
Funneliformis mosseae, Rhizophagus irregularis	Apple (*Malus pumila)*	Canker (*Neonectria ditissima)*	[17]
Rhizophagus irregularis	Tomato *(Solanum lycopersicum)*	Fusarium wilt (*Fusarium oxysporum*)	[24]
Glomus versiforme	Red Sage/Danshen *(Salvia miltiorrhiza)*	Fusarium wilt (*Fusarium oxysporum*)	[25]
Rhizophagus irregularis	Banana *(Musa sp.)*	Fusarium wilt *(Fusarium oxysporum f. sp. Cubense)*	[9]

Glomus viscosum	artichoke *(Cynara scolymus L.)*	Verticillium wilt *(Verticillium dahliae)*	[26]
Rhizophagus irregularis	Banana *(Musa sp.)*	Black Leaf Stack Disease (BLSD) *Pseudocercospora fijiensis*	[27]
***Chaetomium* spp.**	Potato, tomato and tobacco	*Phytophthora* spp	[19]

2.2. Arbuscular mycorrhizal fungi (AMF) prevent plants from bacterial infection

The natural environment of plants-AMF association most of the taxa of bacteria are linked with the AMF mycelia and plays their roles to colonize the surfaces of spores and extra radial hyphae, which leads to the formation structures that are similar to biofilms [28]. Bachir Iffis et.al reported that the bacterial taxa have been observed on the surface of mycelia that belongs to the α-, β-, and γ-Proteobacteria and Firmicutes, using cultured dependent and culture independent. Moreover, in some of the AMF species, bacteria were living in the cytoplasm in the form of endobacteria [29]. Therefore, the propagules of AMF that are associated with the genera of different bacteria have been isolated from *Solidago rugosa* include *Pseudomonas sp. Streptococcus sp. Bacillus sp. Lactobacillus sp*. by using 16S rRNA gene sequencing [30] has been mentioned below in table.

Table 3: The propagules of AMF isolated from Solidago rugosa

Source *(Solidago rugosa)*	Bacterial taxa
	Pseudomonas sp.
	Streptococcus sp.
	Bacillus sp.
	Lactobacillus sp.

About 250, 000 cells of bacteria are present that reside in the cytosol of the bacteria (endobacteria). Endobacteria are unable to grow without AMF as they are the obligate biotrophs. Number of authors reported in their work that the AMF with plants and bacteria should consider tripartite associations which result in the consortium involved in the plant development and growth [31]. Moreover, they play major roles in nutrient cycling, decomposition and disease repression. Some types of bacteria can also form symbiotic relationships with plants, known as rhizobia, which can fix nitrogen in the soil, making it available for plant growth. In order for this symbiotic relationship to form, plants have receptors that recognize and respond to specific bacterial signals [32]. These receptors are called rhizobial receptors and they are responsible for initiating the formation of the symbiotic relationship. Arbuscular mycorrhizal fungi interact with plants to protect against the bacterial toxins, through direct and indirect mechanisms. AMF protects plants from bacteria by priming the plant's defense mechanisms. AMF can activate the plant's immune system by releasing signaling molecules that trigger the production of the defense-related compounds, like phytohormones, enzymes etc. These compounds can then help the plant to fend off attacks from pathogens, including bacteria. Some AMF can colonize the root system of the plant, making it difficult for bacteria to establish themselves [33].

AMF can also compete with pathogens for nutrients, which can reduce their ability to infect the plant. Another way that AMF interacts with bacteria is by forming mutualistic relationships with certain types of bacteria, known as rhizobia via rhizobial receptors which can fix nitrogen in soil, making it accessible for plant growth. Bhaskar Dowarah et. al reported in his findings that plant AMF association shows positive effects in protection against the bacterial species [34] mentioned in the table listed below.

Table 4: Species of AMF that affect the bacterial plant pathogens

AMF species	Pathogenic bacteria	Host	References
Glomus versiforme	*Ralstonia solanacearum*	Tomato *(Solanum lycopersicum)*	[3]
Entrophospora infrequens	*Pseudomonas syringae pv. glycinia*	Soybean *(Glycine max)*	[35]
Gigaspora intraradices	*Xanthomonas campest*	Barrelclover *(Medicago truncatula)*	[36]
Funneliformis mosseae	*Xanthomonas translucens*	Wheat *(Triticum aestivum)*	[37]
Rhizophagus irregularis	*Candidatus*	Tomato *(Solanum lycopersicum)*	[38]

2.3. Arbuscular Mycorrhizal fungi protects plant plants from Viruses

Viruses are the obligatory type microorganisms that spread disease to a wide range of plant species and resultantly reduce the crop yields around the globe [39]. Instead of their commercial significance, there is a little and conflicting detail on how the Arbuscular Mycorrhizal fungi affects viral infections. The plant viruses are a significant barrier to the economic farming of a variety of crops [40]. Moreover, with time, mycorrhizal plants may become more susceptible to the viral presence as virus concentration steadily rises in comparison to non-mycorrhizal controls. In early stages, the mycorrhizal plants exhibit less severe symptoms or no difference in virus infection compared to the non-mycorrhizal ones [41].

One of the significant issues preventing the economic production of many of the crops is the presence of the plant viruses. Certain strategies have been proposed to overcome different kinds of viruses in different plants [42] The impact of AM fungus on the viral infections varies and the shoot virus infection has received a lot of research in this regard. Moreover, tobacco mosaic virus (TMV) and the Rhizophagus intraradices (Arbuscular Mycorrhizal fungus) interactions in the tobacco leaves were examined with the goal of determining the severity of disease. It was found that the disease symptoms are more pronounced in mycorrhizal plants than non-mycorrhizal ones [43].

Arbuscular Mycorrhizae supports the plant by inspecting the soil outside the rhizosphere and also functions as an addition of the root system. Because the extraradical mycelium has a high absorptive capability i.e. the host plant can more easily acquire soil water and gradually release mineral compounds particularly the phosphate and nitrogen ions that improves plant's nutritional condition as well as strength [44]. In return the plants provide an environment that enables the Arbuscular Mycorrhizal fungi to consume energy in order to complete their life cycle. In another review, it was observed that the arbuscular mycorrhizal fungi improved pepper plant's resistance to the tomato brown rugose fruit virus (ToBRFV) which is a destructive virus harming tomatoes and peppers, resulting in the losses that might be 100% but the use of AM resulted in the appearance of fewer symptoms [45].

Mycorrhiza-induced resistance is a mechanism by which the tomato plants have their relationship with AMF, which can enhance the plant nutrition and boost systemic plant defenses against viral attacks [46]. However, restricted and inconclusive research on the impact of AMF on viral infection suggests that AMF colonization may negatively affect plant's ability to defend themselves against viruses hence for these circumstances, the term mycorrhiza-induced susceptibility (MIS) has been suggested [47].

Table 5: Species of AMF that affect the Viral Species

Mycorrhizal species	Crop species	Virus	References
G. etunicatum	*Citrus Aurentium*	CLRV-2 Tristeza virus T3	[48]
G. etunicatum	*Citrus Macrophylla*	Tristeza virus T1	[7]
Endogone macrocarpa var. geospora	*Fragaria Chiloensis Ananassa*	Arabis Mosaic virus	[13]
E. macrocarpa var. geospora	*Lycopersicon Esculentum*	Tobacco Mosaic virus	[42]
G. mosseae *G. intraradices*	*Nicotiana Tabacum*	Tobacco Mosaic virus	[47]
E. macrocarpa var. geospora	*Petunia sp.*	Arabis Mosaic virus	[33]

2.4. Arbuscular mycorrhizal fungi (AMF) protect plants from pest:

Pests are organisms that feed on plants, causing damage and reducing growth and health. Pests can be insects, pathogens or herbivores and they can cause direct damage to the plant by feeding on the leaves, stems and roots or by transmitting plant diseases [49]. This can lead to reduced plant growth, reduced crop yields and increased costs for farmers, thus leading to plant death [50]. Arbuscular mycorrhizal fungi and pests are both essential factors which can affect plant growth and health. AMF are soil fungi that colonize root cells of plants and extend their hyphae into surrounding soil [51]. The relationship between AMF and plants is mutualistic i.e. both increase the plant's ability to absorb phosphorus, zinc and other essential nutrients by up to 100-fold. AMF can help to protect plants from pests by increasing the plant's resistance to stress like they play a critical role in plant water uptake, helping plants to survive in drought conditions [52]. This can make the plant less attractive to pests, as well as making it more resistant to damage from pests that do attack. Additionally, AMF can help to improve the overall health of the soil, which can make it more difficult for pests to establish themselves and thrive. AMF can also help to protect plants from pests by promoting biodiversity in the soil. This can include increasing the populations of beneficial organisms, such as predators or pathogens, that can help to control pests [53].

For instance, some studies have demonstrated that AMF can enhance the populations of nematodes that feed on insects, which can help to reduce pest populations. So, AMF helps to improve overall health and crops productivity, which can make them more resilient to pest damage [54]. This can include upgrading growth, yield of crops and improving the quality of the crops. One of the ways that AMF protects plants from pests is by activating the plant's defense mechanisms. AMF can activate the plant's immune system by releasing signaling chemicals that trigger production of the defense-related compounds, like phytohormones [55]. These compounds can then help the plant to fend off attacks from pests and pathogens. AMF can also protect plants from pests by completely changing the mutualistic environment of plant roots by releasing certain compounds which will alter the pH. Also change the microbiota present in soil, making it less favorable for pests and pathogens [56].

Table 6: AMF species and their effect on pest

AMF species	Pest	Host	References
Rhizophagus irregularis, Glomus aggregatum, Funneliformis mosseae, Claroideoglomus etunicatum	*Lissorhoptrus oryzophilu*	Rice *(Oryza sativa)*	[39]
Glomus mosseae *Gigaspora sp.*	*Spodoptera litura*	Brassica junica (Indian Mustard)	[57]
Glomus intraradices	*Spodoptera litura*	Blackgram *(Vigna mungo)*	[58]

Glomus mosseae	*Tetranychus urticae*	*Phaseolus vulgaris*	[46]
Glomus intraradices	*Aphidius rhopalosiphi*	*Phleum pratense*	[41]

3. Activation of Plant Root Defense Mechanism:

The AMF acts as a biocontrol agent to protect the plants from biotic stresses as mentioned above. The diagram below demonstrates the mechanism of plant defense against biotic stress [59]. Firstly, the colonization between AMF and plant roots further leads to the cascade of events, signal transduction pathways and chemical toxins release [60]. The surface of AMF has small molecules known as microbe associated molecular patterns (MAMP), also special proteins that are secreted in the plants. The receptor protein complexes in the plants i.e., pattern reorganization receptors recognize the MAMP and leads to the invigoration of MAMP triggered immunity or immunity triggered by the effectors [61]. The induced immunity leads to activation of effector molecules that were produced by AMF. The R proteins recognize the effectors which will show the hypersensitive response by ethylene biosynthesis, reactive oxygen species production, antimicrobial compounds production, PR-1 protein synthesis, callose deposition and cell wall thickening. The compounds after activation produce certain hormones to defend the plant [62]. Moreover, the transcriptional changes control the release of toxins by pathogens. The dense induced by AMF also include chitinases, phytoalexins, proteins related to pathogenesis, β-1,3-glucanases, phenolic compounds, hydroxyproline-rich glycoprotein as well as the enzymes of phenylpropanoid pathway. Moreover, there are few effectors that suppress the triggered immunity to facilitate the AMF colonization [58]. The effectiveness of AMF to protect the crop plant against biotic stresses not only depends on the AMF but also on the certain factors including microorganisms that are present in the rhizosphere, agroclimatic conditions and rhizosphere chemistry. However, AMF plays a wide role in defense against fungi, virus bacteria and herbivores. They not only control single disease but perform a significant role in multi disease protection [63].

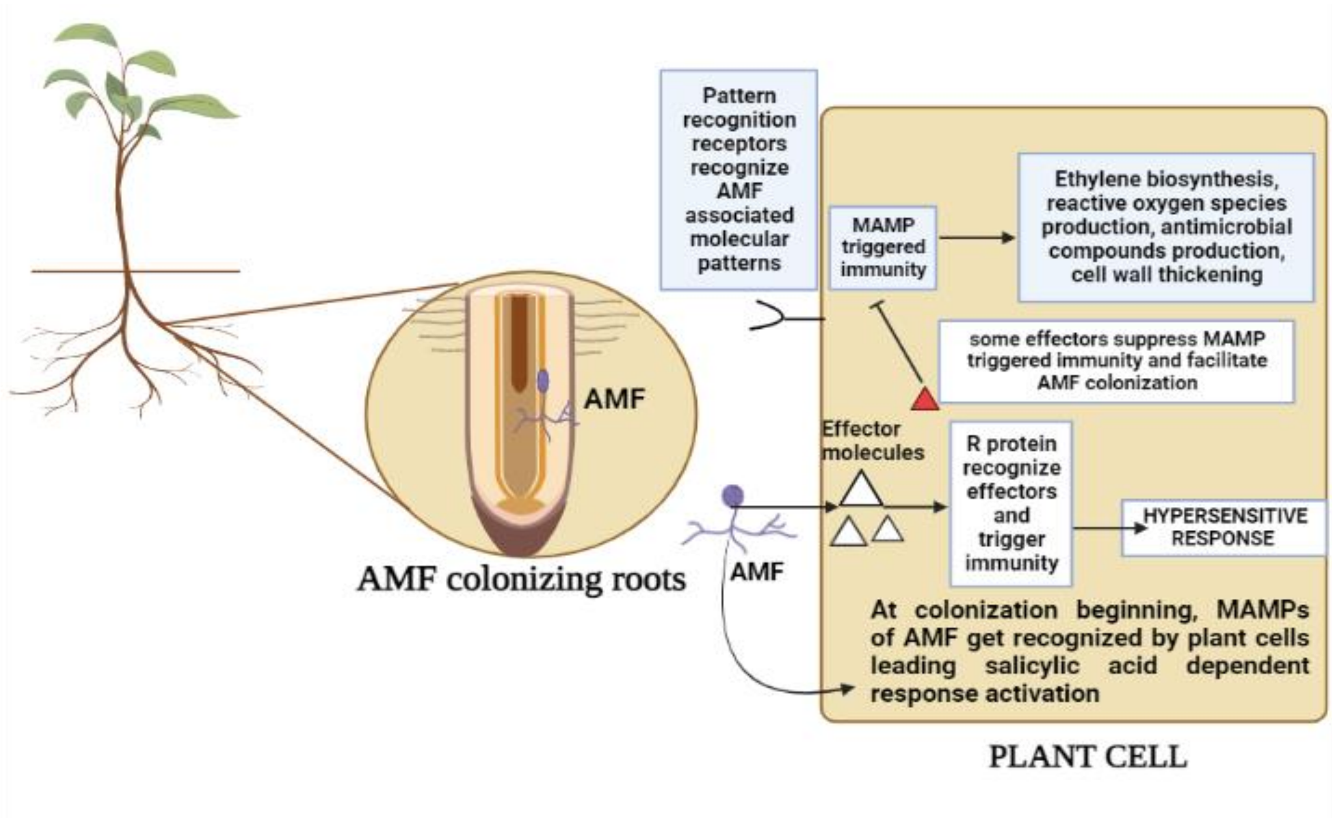

Figure 3: Activation of MAMP triggered immunity (MTI) after the AMF colonization association with plants

4. Factors that limit the use of AMF:

With all the studies and researches conducted towards the beneficial role of arbuscular mycorrhizal fungi in plant development, growth and protection against biotic and the abiotic stresses there is still some limitations and hindrances in use of AMF in certain areas of world and it also questions the sustainability of AMF as biofertilizers [64]. Many factors can limit the benefits of AMF and hence cause reduction in efficiency of these symbiotic organisms. Use of chemicals such as pesticides, fungicides and herbicides, tillage practices, use of rotational crops with no AMF associations may not only hamper the activity and productive effects of these fungi but also destroy their colonies [65].

4.1: Use of Fungicides and other Agrochemicals:

Use of phosphorus rich fertilizers can reduce the activity of AMF as when enough phosphorus will be available to plant it will not trade back carbohydrates. This lack of carbon source will affect the root colonization of plants hence leading to inactivity of arbuscule formation [48]. Likewise, in case of nitrogen and potassium, excessive use of fertilizers can also limit signal transduction to AMF which in turn decreases diversity of these fungal species [66].

Other agrochemicals such as fungicides and pesticides that are used to control certain diseases and pathogens of plants can also affect growth of AMF badly. These chemicals can block the uptake of nutrients by the fungi. Certain developmental processes of fungi such as spore germination, transfer of nutrients, synthetic pathway of sterol etc. have been reported to be affected by use of certain harmful fungicides [38].

4.2: Tillage and Agricultural practices:

Tillage which can be defined as preparation of soil for cultivation of crops can destroy the normal colonization of root mycorrhiza. These practices lead to crashing and physical destruction of mycelium reducing their variety and hampering their beneficial role. Moderate and balanced tillage has shown to improve quality as well as quantity of these microbes which in turn nourishes soil and improves its fertility [37].

The practice of crop rotation is another problem that limits the benefits obtained by AMF. The varieties of crop that lack the root colonization of these mycorrhiza when grown can disturb the abundance of these microbes, hence the following crop may not get desired benefits as one would have expected [36]. Paddy fields are also problematic as AMF cannot grow in damp fields. Another problem is preparation of inoculum of these microbes as they are obligate microbes so they require host for their propagation and reproduction which make production of their cultures time consuming and tiresome. These problems limit exploitation of benefits of these microbes but research is needed to overcome these problems [57].

Conclusion:

Many different types of microorganisms inhabit the plants during their life cycle. Some impart beneficial effects while others could be hazardous to plants causing certain diseases. Arbuscular mycorrhizal fungi form symbiotic relationships with the plants and not only improve its development but also confers resistance against many biotic stresses including fungi, bacteria, viruses and some insects. This protection is mediated by induction of specific proteins activated by plant fungus interaction leading to regulation of different metabolic pathways such as JA and salicylic acid pathways. In this review AMF plant symbiotic relationship under biotic stresses has been discussed. Use of AMF as a biofertilizer for enhanced productivity and stress tolerance in plants has been established. Many researchers have documented the role of AMF as a natural friend of plants helping it against pathogens. Regulation of metabolic pathways and activation of receptors involved in this symbiosis is the area that needs to be further explored in order to exploit this microorganism for improving agricultural practices. Despite their beneficial aspects certain limitations are needed to be overcome to gain full advantage of these natural fertilizers and utilize them in place of conventional procedures.

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