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Research Article

Meta-Analysis: Impact of AMF Colonization on Allium Species

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Abstract

With available data, this meta-analysis assessed the effects of Arbuscular Mycorrhizal Fungi (AMF) inoculation on well-researched Allium species related to the under-researched *Allium tricocum* species. The overarching goal was to synthesize findings from published research to develop hypotheses for future investigation into the role of AMF inoculation on *Allium tricocum*. Three Allium species were evaluated across ten studies. Four parameter categories were assessed. Parameter categories included

- plant mass, plant size, and yield
- mineral composition
- elemental composition
- phosphorus uptake

Two treatment comparison categories were constructed for each parameter category:

- Allium species inoculated with AMF
- uninoculated (control).

Data were presented for each parameter in % increase with control treatment. Allium species inoculated with AMF increased most parameter categories compared to uninoculated controls, except for a few element composition parameters (in a saline environment). They also had the highest average percent increase compared to uninoculated controls.

The meta-analysis indicated that the inoculation of AMF on Allium species is closely related to *Allium tricocum* and generally increases growth, yield, and mineral composition, but it is dependent on the Allium species and AMF species used. Areas for future research of *Allium tricocum* identified through this analysis include whether AMF inoculation could help better growth of Allium tricocum? if AMF inoculation helps in early maturation of Allium tricocum? is the impact of AMF changes with the annual cycle for the growth and survival of Allium tricocum? And does AMF inoculation help transplanted Allium tricocum for adaptation to a new environment?

Introduction

Arbuscular Mycorrhizal Fungi (AMF) belong to *Phylum Glomeromycota* (Redecker et al., 2000; Schußler et al., 2001). They have an obligate symbiotic association with 90% of all vascular plant roots. AMF improves plant growth profoundly through increased uptake of Phosphorus, nitrogen, and other [1]. Because of the higher cost and hazardous effects of heavy doses of chemical fertilizers, interest in using AMF to enhance plant growth and yield has increased in recent years. AMF improves plant growth because extra metrical hyphae act as extensions of plant roots and increase the surface area of the root system and the absorption efficiency of water and diffusion-limited nutrients. The effect is even more pronounced in P-deficient soils (Bagyaraj and Reddy, 2000). Other beneficial effects are biological control of root pathogens, biological nitrogen fixation, and increased ability to withstand abiotic stresses.

The benefits of AMF under field conditions have been reported for annuals and perennials inoculated in nurseries (Bagyaraj, 1984; Chandrashekar et al., 1995). Many forest herbaceous plants such as *Erythronium americanum* (trout lily), *Podophyllum peltatum* (mayapple), and *Allium tricocum* (wild leek) can maintain mutually beneficial relationships with AMF [2] (Brundrett and Kendrick 1990ab, Lapointe and Molard 1997; Watson et al. 2002). Compared to conditions without colonization, AMF colonization benefits forest herbs through increased biomass, growth, and Phosphorus (P) acquisition (Helgason et al. 2002). The AMF species, plant genotype, soil nutrient availability to plants, and environmental and stress factors determine the efficiency of AMF utilization (Chen et al., 2018). Because they have less developed root systems, Allium species are more sensitive to AMF inoculation compared to other species with larger root masses (Deressa et al., 2008; Bowling et al., 1980; Greenwood et al., 1982; Sanders et al., 1992; Bever et al., 1996). With more than 900 species residing predominantly in the Northern Hemisphere, Allium is considered one of the genera containing some of the largest numbers of species in the plant kingdom. Many Allium species also are widely used in human nutrition, such as garlic (*Allium sativum*), onion (*A. cepa*), leek (*A. porrum*), shallot (*A. cepa* L. *Aggregatum* group), chive (*A. schoenoprasum*), and bunching onion (*A. fistulosum*) (FAOSTAT 2015).

The perennial forest herb *A. tricocum* (Ramps), also called wild leek, is a spring ephemeral that persists for only brief periods in the forest understory before the leaf emergence of canopy trees when light availability on the forest floor is highest [3]. The bulbs persist throughout the year and are the main storage organ for the plant's nutrients. New roots form on the rhizome at the bulb's base in autumn and have an annual life cycle [4,5]. The resource capture for ramps throughout the year can be influenced by mycorrhizal colonization as the roots of *A. tricocum* are known to be colonized by AMF, with as much as 60 % of the root length possessing mycorrhizal structures [2].

Despite known natural AMF colonization, the potential effect of AMF inoculation on the growth of ramps is understudied. To improve insights into the potential role of AMF inoculation on-ramp performance and propose hypotheses for future research, we conducted a meta-analysis of the impact of AMF inoculation on the growth of the following closely related and better-studied Allium species: *Allium cepa* (Onion), *Allium sativum* (Garlic), and *Allium porrum* (Leek). Given that these Allium species are annual, we account for potential differences related to the perennial nature of ramps.



Methodology

Database and Selection Criteria

Journal articles selected for inclusion in this study were obtained from Virginia Tech's library database. The keywords "Arbuscular mycorrhizal fungi" and "Allium" were entered into the database search engine, and the search was narrowed using the database's search tools to find articles containing these words in either the title or the abstract. The search engine presented 689 articles with matching keywords in the title or abstract. Of these 689 articles, some were duplicates, and others were written in a language other than English. Such articles were rejected for the search. The remaining articles were then selected for the study using predetermined selection criteria or rejected if they did not meet the selection criteria. To be selected for inclusion in the study, articles had to meet the following three criteria:

- Data include measures of growth, yield, or nutrient/mineral content
- Data is presented numerically in a table.
- Allium species inoculated with AMF or uninoculated controls.

In addition, some of the articles not found in Virginia Tech's library database were downloaded from Google Scholar since they match the selection criteria.

Data Organization

After selecting articles for the meta-analysis, data tables were transcribed into Microsoft Excel version 2010 and organized into four measuring categories: plant growth, Mineral composition, Elemental composition, and phosphorus uptake. There are different measurement parameters within each category. Because different studies measured different growth parameters, yield, and mineral and nutrient content, it was necessary to group data into these broader categories. These four measures of Allium species growth, mineral and elemental content, and phosphorus uptake will represent parameter categories throughout this paper.

Within each parameter category, there was a variety of different kinds of measurements. For example, measures of plant growth included leaf area, plant height, bulb diameter, fresh and dry weight of root, shoot and bulb, shoot height, and root length. Since there was not enough data for the yield of all Allium species, we included yield parameters within the measure of plant growth category. Some studies presented data on different growth parameters of Allium species and compared them with the growth of other species and the percentage colonization of AMF, but they were not included in this meta-analysis because they did not fit our purpose of the study. Some studies have shown the data on growth parameters within different species of AMF inoculated in the study. In that case, we selected the 2-3 species of AMF that are almost similar in all 10 papers and took the average of growth data within different (2-3) species of AMF. Not having enough data on the growth and mineral content parameters in all 10 papers, we could not have enough data to compare the effect of AMF on the growth of different Allium species. Therefore, we showed the data differently for all 3 species.

Calculations

After organizing data into the four categories, calculations were made to determine the percent change between experimental treatment and controls for each parameter/category tested.

Result

Measures of plant growth

The average of every growth parameter increased by a certain percentage in the inoculated Allium species studied in this paper compared to the uninoculated ones. The highest % increase in bulb dry weight can be found in mycorrhizal onion and garlic (nearly 75% increase in both) (Tables 1 & 2). The plant growth data on onion showed that the highest % increase was found with Dry and fresh weight root, fresh weight shoot, and leaf area in mycorrhizal treatment (more than 100% increase) (Table 1). Inoculation with mycorrhiza also showed an increase in bulb fresh weight and bulb diameter in onion under fumigated soil conditions that were not so much different than the increased growth under normal soil conditions (Figure 1). Surprisingly, the fresh weight of the shoot and root of inoculated onion was less than uninoculated in

soil fertilized with Nitrate. Under Ammonia fertilization, both parameters were higher than in normal soil conditions in mycorrhizal onions (Figure 2). Inoculation with mycorrhiza showed higher root and shoot growth under the soil when applied with phosphom (Figure 2). Mycorrhizal onion and garlic showed a higher yield increase than controlled ones, with around 150% increase in yield with inoculation (Table 1 & Table 2). Plant shoot height increases with a higher percentage of mycorrhizal inoculation in leeks than onion and garlic (Tables 1-3).

Table 1: Impact of AMF on growth of Onion (Data presented in % increase with control).

Growth parameter (Onion) % increase with control	
Parameter	% Increase with control
No of leaves	30
Leaf area	142.86
Plant height	38.24
Bulb diameter	23.72
fresh weight shoot	103.35
Fresh weight root	145.99
Fresh weight bulb	53.06
Dry weight g/root	110
Dry weight g/bulb	73.93
shoot dry wt	70.19
shoot height (cm)	20.83
Root length	15.79
Yield	145
Neck Diameter	28.57

Table 2: Impact of AMF on growth and yield of Garlic (Data presented in % increase with control)

Growth parameter (Garlic)% increase with control	
Parameter	%increase with control
No of leaves	4.5
Leaf area	14.37
Plant height	16.29
Bulb diameter	5.75
Fresh weight	44.75
Dry weight g/bulb	78.13
Yield	149

Table 3: Impact of AMF on growth of Leek (Data presented in % increase with control).

Growth parameter (Leek) % increase with control	
Parameter	%increase with control
shoot dry wt.	46.67
shoot height (cm)	25
Root length	30.77

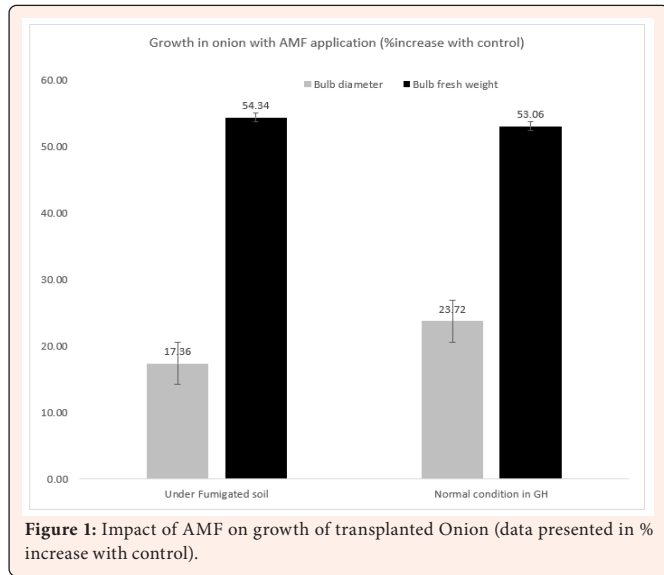


Figure 1: Impact of AMF on growth of transplanted Onion (data presented in % increase with control).

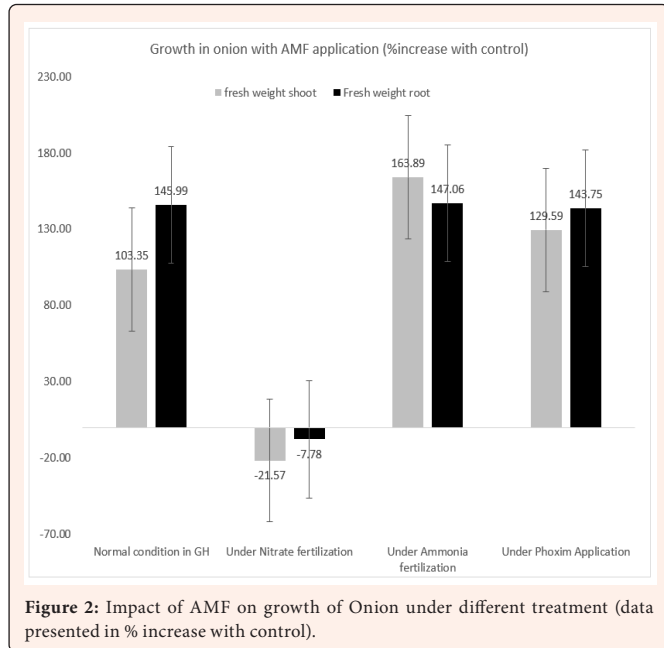


Figure 2: Impact of AMF on growth of Onion under different treatment (data presented in % increase with control).

Mineral content

Mycorrhizal inoculation showed an increase in the % of mineral content in onion and garlic. For example, there was a higher % increase of Selenium (488 %) among available data of mineral content in mycorrhizal garlic than with controlled (Table 4). Both onion and garlic showed a pretty good amount of % increase in Ca with mycorrhizal inoculation (around 150% increase than in controlled) (Figure 3 & Table 4). Inoculation with mycorrhizae also showed an increase in some mineral content of onions grown under high saline conditions. Phosphorus and copper content in mycorrhizal onion were highly increased with controlled under saline conditions than in normal conditioned soil (Figure 3). Some minerals like B, Ca, and Iron in mycorrhizal onion were lower than uninoculated onion under highly saline conditions (Figure 3). Most minerals in mycorrhizal garlic increased by more (more than 100%) than in controlled ones (Table 4).

Table 4: Impact of AMF on mineral content of Garlic (Data presented in % increase with control).

Minerals parameter (Garlic)% increase with control	
Parameters	% Increase with control
Se	488
P	110
K	100
Ca	145
Mg	191
Na	206
B	100
Fe	100
Cu	100
Mo	140
Zn	100
Si	68

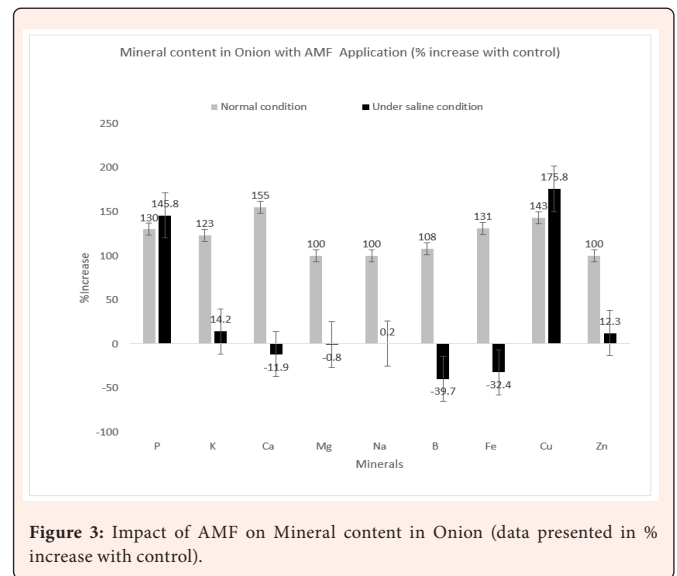


Figure 3: Impact of AMF on Mineral content in Onion (data presented in % increase with control).

Elemental composition

Most of the biochemical elements in onion and garlic increase by a reasonably high percentage (more than 100%) with mycorrhizal inoculation. For instance, one health-promoting phytochemical, flavonoid content, is 100% higher in both mycorrhizal garlic and onions than in controlled ones (Tables 5 & 6). Other biochemical elements like monosaccharides, Ascorbic acid, total sugar, etc., are also increased by more than 100% in mycorrhizal onion and garlic than in controlled ones. Phenolics, one of the best chemicals for plant defense mechanisms, also seem to be increased in onions with mycorrhizal inoculation (Table 5).

Table 5: Impact of AMF on elemental composition of Onion (Data presented in % increase with control).

Biochemical parameter (Onion)% increase with control	
Elemental composition	% Increase with control
monosaccharides	168
Total sugar	100
AOA	115
Flavonoids	100
Ascorbic acid	100
TA	145
Pyruvate	37.15
Total phenolic	27.67

Table 6: Impact of AMF on elemental composition of Garlic (Data presented in % increase with control).

Biochemical parameter (Garlic)% increase with control	
Elemental composition	% Increase with control
monosaccharides	100
Total sugar	119
AOA	100
Flavonoids	100
Ascorbic acid	100
TA	88

Phosphorus Uptake

Mycorrhizal inoculation seems to help in phosphorus uptake in onions and leeks. That helps to increase phosphorus concentration in mycorrhizal onions and leeks compared to controlled ones. Phosphorus uptake seems to increase highly (more than 100%) in both onions and leeks with mycorrhizal inoculation compared to uninoculated ones (Figure 4). Compared to the increase in phosphorus uptake, the increase in phosphorus concentration seems to be higher in inoculated leeks than in inoculated onions (Figure 4).

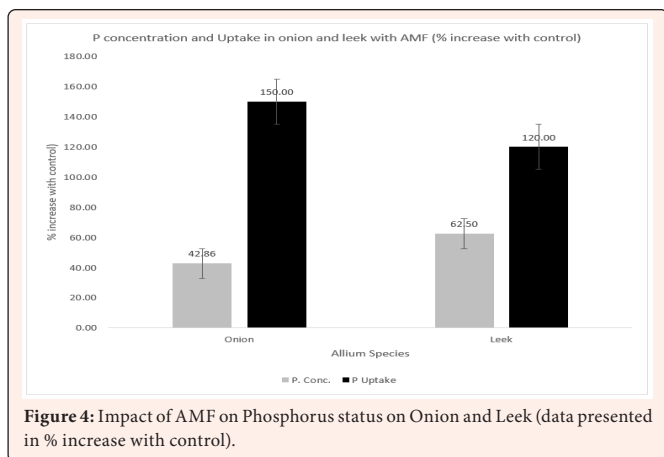


Figure 4: Impact of AMF on Phosphorus status on Onion and Leek (data presented in % increase with control).

Discussion

The results of this study showed that the inoculation with mycorrhizae generally causes higher plant growth in Allium species studied for this paper (Onion, Garlic, and Leek). Borde et al. [6] mentioned that AM fungal inoculation increases the level of

AMF root colonization of garlic, which is very important for plant growth and nutrient uptake. Many researchers have observed the enhanced growth of onion plants and leek plants inoculated with AMF, and the study done by Tinker [7] mentioned that the increase in shoot growth of onion and leek plants inoculated with AMF could be due to an increase in the uptake of nutrients, especially P [7]. El-Seoud [8] in his study also mentioned that the increase in shoot growth of onion and leek plants inoculated with AMF could be due to an increase in the uptake of nutrients, especially Phosphorus. Several other studies have also stated that the increase in plant growth with AMF application may be due to increased nutrient uptake and tolerance towards drought stress [9]. This study found that the percentage increase in onion root and shoot weight was unexpectedly higher with AMF inoculation, even under phoxim-applied soil.

The reason behind this may be because the applied AM fungi are not sensitive to phoxim [10]. Inoculation with AMF has also been shown to benefit yield, carbohydrates, TA, and antioxidant content in the mentioned Allium species in this study. However, the effect of AMF on the mentioned Allium species greatly depends on plant genotype, AMF single species or species consortium, farming management, and soil and environmental conditions [11]. This study also showed an increased percentage of mineral content in onion plants grown under high saline conditions, indicating that AM fungi reduced the detrimental effects of salt on the soil. This alleviation of salt stress is not fully known but seems to be due to several potential metabolic processes that could be facilitated by P nutrition [12].

Phosphorus and Sulphur (S) are two necessary nutrients that increase the growth and yield of plants. Applying mycorrhizal fungi increases this study's special phosphorus uptake to host plants. The increased water and nutrient absorption with AMF inoculation may have transmitted special Phosphorus to host plant cells. That may enhance photosynthesis, which produces more assimilation (Mohamed, 2012). That may have ultimately improved the growth of the host plant. This study also demonstrated how mycorrhizal inoculation affected antioxidant enzymes and enhanced flavonoid concentration. That may also be due to increased nutrient uptake or induction of the plant defense system [13].

Although mycorrhizal inoculation has notable impacts on the physiology and biochemistry of the entire plant, the root is where most symbiotic symbiosis-related alterations occur—the mycorrhizal root acts as a more “active” organ than the root itself. The influence of the mycorrhiza (root+AMF), also known as the “mycorrhizosphere,” was wider and more obvious than that of the non-symbiotic roots in the soil [14]. Finally, looking at how AMF can influence the growth and nutrient content in Allium species studied in this paper, we can see that if inoculated with AMF, we can positively impact ramp plants. However, it may depend on the AMF species and the ramp plant phenology [15,16].

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