



Hungarian University of Agriculture and Life Sciences

**The Beneficial Effects of The Microbial Inoculates
Application to Improve Root Vegetables Production**

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LIST OF ABBREVIATIONS

AMF : Arbuscular mycorrhizal fungi

PGPR: Plant growth promoting rhizobacteria

ÖMKi: Research Institute of Organic Agriculture

SOAICE: Solutions for Improving Agroecosystem and Crop Efficiency for Water and Nutrient Use

PAHs: (Polycyclic aromatic compounds)

Caco₃ : Calcium carbonate

IH : Intraradical hyphae

EH: Extraradical hyphae

ePGPR: extracellular Plant Growth Promoting Rhizobacteria

iPGPR: intracellular Plant Growth Promoting Rhizobacteria

N: Nitrogen

P: Phosphorus

MATE : Hungarian University of Agriculture and Life Sciences

SFA: Segmented Flow Analysis

BBCH : Biologische Bundesanstalt, Bundessortenamt and CHemical industry

L+SYM: Latagro peatmoss with Symbivit

(L+SYM).S) : Sterilized Latagro peatmoss with Symbivit

F: Soil from Soroksar experimental research farm, located at the Hungarian University of Agriculture and Life Sciences organic farming Unit

F %: Frequency of mycorrhiza in the root system

M %: Intensity of the mycorrhizal colonization in the root system

m %: Intensity of the mycorrhizal colonization in the root fragments

a %: Arbuscular abundance in mycorrhizal parts in root fragments

A %: Arbuscular abundance in the root system

No₃: Nitrate

NH₄⁺ : Ammonium

1. INTRODUCTION AND OBJECTIVES

Root vegetables are considered an important food around the world and as a rich source of vitamins and minerals. To meet the growing global food demand for root vegetables, it is important to provide consumers with high nutritional value. Due to this increase in root vegetable consumption, the cultivation of root vegetables is also increasing worldwide. The production of root vegetables has also increased in recent years due to the expansion of cultivation. Therefore, more researchers became interested in this subject on how to improve root vegetables quality to achieve high production.

In order to attain better quality of food crops and also to protect the environment, green technologies such as microbial inoculation, which aim to either replace or reduce the use of agrochemicals and preserve a clean environment, are good solutions to current agricultural problems especially in integrated and organic growing. The use of alternatives and eco- friendly solutions is crucial to replace synthetic inputs with organic materials while improving the chemical, physical and biological properties of soils (PAPP et al. 2021). Overuse of chemical fertilizers leads to several environmental problems including groundwater pollution, soil degradation and their impact on crop growth (SAVCI 2012). To reduce these negative effects, alternative ways must be found, such as the inoculation of beneficial microorganisms into the soil (AL ZABEE and ALMALIKI 2019).

Some plant-microbe interactions such as Plant Growth Promoting Rhizobacteria (PGPR), Arbuscular Mycorrhizal Fungi (AMF) and compost have been widely used to enhance plant growth through different mechanisms of action (TAHIRI et al. 2022). Also, microbial inoculants are easy and inexpensive to manufacture compared to chemical pesticides (ELNAHAL et al. 2022). The benefits of co-inoculating phosphate-solubilizing PGPR and/or nitrogen-fixing PGPR with mycorrhiza in plants have been demonstrated (KUMAR et al. 2017). Microbial inoculation of plants is already widely used in many research projects. It is important to study in more details the effects of these microorganisms in plants and their interaction with the host plants, especially PGPR, AMF and *Trichoderma*, as these are key players in plant growth. In order to be able to better assess these effects, it is important to carry out further researches, and in fact, they may be various depending on the different conditions of the experiment.

Based on the previous literatures, our study was divided into two parts; the first part was conducted as pilot research in a glasshouse of the Department of Vegetable and Mushroom Growing located at the Hungarian University of Agriculture and Life Sciences – Budai campus in 2019 to check the

symbiosis between the sweet potatoes and mycorrhiza and the influence of mycorrhiza on the physical parameters of sweet potato seedlings such as, length of roots and shoots (cm), fresh weight of shoots, roots (g) and the length of the stem (cm) of the sweet potato seedlings.

Due to the corona virus in 2020, the research goals had to be changed and be involved in a field experiment by a cooperation and help from ÖMKi (Research Institute of Organic Agriculture) to be able to continue my Ph.D studies and research. This experiment carried out by ÖMKi in two years (2020+2021) at the Soroksár experimental research farm, located at the Hungarian University of Agriculture and Life Sciences. This experiment was part of the SolACE project (Solutions for Improving Agroecosystem and Crop Efficiency for Water and Nutrient Use). This study examines the effects of different microbial inoculations and their combinations on potato tubers yield (kg/m^2), starch content in potato tubers (%), Total phosphorus content in potato tubers (mgP kg^{-1}). Also, it observed the symbiosis and the mycorrhizal colonization parameters (F %: Frequency of mycorrhiza in the root system, M %: Intensity of the mycorrhizal colonization in the root system, m %: Intensity of the mycorrhizal colonization in the root fragments, a %: Arbuscular abundance in mycorrhizal parts in root fragments, A %: Arbuscular abundance in the root system)

OBJECTIVES:

- Highlighting the differences between two sweet potato varieties (orange and purple) in different parallel tests, such as the inoculation method used.
- Studying whether the mycorrhizal inoculum is effective in developing a symbiotic relationship with the roots of sweet potato, affecting the physiological and physical parameters of the plant; in addition, it will be investigated whether the sterilization of the substrate has an influence on mycorrhizal colonization.
- Detecting the effectiveness of mycorrhizal inoculation with a sterilized substrate on the:
 - Mycorrhizal parameters (F %, M %, m %, a %, A %).
 - Physical parameters such as, length of roots and shoots (cm), fresh weight of shoots, roots (g) and the length of the stem (cm) of the sweet potato seedlings.
- Determining the beneficial effects of the microbial inoculates to improve potato productivity with and without irrigation in field conditions.
- Observing which microbial inoculation combination yields the highest performance and improves potato production in organic farming by the measured parameters:
 - Mycorrhizal parameters (F %, M % and A %).
 - Total phosphorus content (mgP kg^{-1}).
 - Starch content (%) in potato tubers.
 - Phenological growth stages of potato plant.
 - Tuber yield (kg/m^2)

2. HYPOTHESIS

Due to the global significance of root vegetables as an important nutrient rich food, recent research and studies have been focused on new knowledge and methods to increase the quality and quantity of root vegetables. According to the traditional cultivation methods and agricultural practices, the production faces many problems and difficulties, for example; the high cost of chemical fertilizers and pesticides, and nutrients depletion. As a result, environmentally friendly methods may be an alternative that can be used to improve vegetable production. Connecting to this field, my PhD research focuses on the efficiency of the beneficial microbial inoculants used and their ability to associate with root vegetables such as sweet potatoes and potatoes in organic cultivation. The research hypothesis

Hypothesis:

- The mycorrhizal inoculation (Symbivit) can be influenced by the orange sweet potato variety (Norangel) and Purple variety (Purple) and by the sterilization of Latagro Basic Substrat KB2 type.
- The sterilized environment can ensure the purity of the substrate to avoid the appearance of pathogens, weed seeds or other microorganisms.
- Organic potato production can be improved by different mixtures of microbial inoculates.
- The applied microbial inoculates (Arbuscular mycorrhizal Fungi, Plant Growth Promoting Rhizobacteria, and *Trichoderma*) efficiency can be affected by the water demand irrigation availability of potato plant (Desiree variety).

3. LITEATURE REVIEW

3.1. Mycorrhizal Inoculation

Arbuscular mycorrhizal inoculated horticultural crops are becoming increasingly common practice, particularly in intensive horticultural production systems, as native soil arbuscular mycorrhizal fungi populations decreased. The efficiency of mycorrhizal inoculation depends on a few factors such as soil phosphorus concentration, ecotype characteristics, cultivar and mycorrhizal species (ORTAS and AKPINAR 2011). A study has shown that the addition of phosphorus during inoculation of arbuscular mycorrhizae could increase uptake of phosphorus and zinc concentrations, however, higher phosphorus intake could also result in low inoculation efficiency of mycorrhizae (ORTAS 2012). The heavy application of nitrogen fertilization and tillage does not result in the arbuscular mycorrhiza showing in beneficial levels, so a milder application should be used to avoid the arbuscular mycorrhiza concentrating, and its activity on nutrient uptake (BORRIELLO et al. 2012).

The arbuscular mycorrhizal association could be influenced directly or indirectly by cultivation practices. The effect could be either good or vice versa, however, farming practices have more negative impacts compared to natural ecosystems, furthermore the composition of soils in agriculture today is rather arbuscular mycorrhizal-unfriendly (ROUPHAEL et al. 2015). Some studies have shown negative effects on arbuscular mycorrhizal spore germination and hyphae elongation as, polycyclic aromatic compounds (PAHs), trace metals in soil, and high concentration of calcium carbonate (CaCO_3) (CALONNE et al. 2010; LENOIR et al. 2016).

3.1.1. Fungi-plant symbiotic association

Plants have developed adaptive strategies for mutualistic microbes in both mycorrhizal and endophytic fungi (YUAN et al. 2007). In nature, it is known that more than 6,000 species of fungi are able to form mycorrhizal associations with about 240,000 species of plants (CHUAN et al. 2019; SINGH 2007). They play an important role in improving nutrient absorption and water absorption. Their strategy is that they create structures in the plant roots and then induce the various functions. Arbuscular mycorrhizal fungi promote plant growth and increases disease resistance and stress tolerance. They also contribute to improving the physical properties of the soil through the aggregate formed by the mycorrhizal mycelium (NADEEM et al. 2013). The importance of

arbuscular mycorrhizal fungi to agricultural and forest plant species lies in their role in plant growth and nutrition. The occurrence of mycorrhizae in tropical forests has a major impact on soil fertility and thus on plant growth and development (SADHANA 2014).

In maize plant, it was shown that *Glomus intraradices* has the ability to promote shoot and root production than non-mycorrhizal inoculated plant (ORTAS and AKPINAR2011). Arbuscular mycorrhiza symbiosis could affect biochemical and physiological processes such as protection towards oxidative damage, improved water usage efficiency, improved gas exchange ratio, enhanced osmotic regulation, water and nutrients uptake (RUIZ-SÁNCHEZ et al. 2010).

A study shows that after being inoculated by arbuscular mycorrhiza, the carbon accumulation in the soil and plant increased, the carbon increased by 585 kg C ha⁻¹ in the soil, and 1897 kg C ha⁻¹ in the plant (SUBRAMANIAN et al. 2011; KRISHNAKUMAR et al. 2013). The root exudates, which has important role to ensure the colonization of arbuscular mycorrhiza may be responsible toward carbon and nitrogen cycling through exhibiting soil organic matter degraders and inhibiting nitrification process. This carbon flow could also affect the soil aggregation (HAICHAR et al.2014). For arbuscular mycorrhiza plants, the plants will allocate labile photosynthates (sugars from photosynthesis), plant-derived carbon to arbuscular mycorrhiza. Arbuscular mycorrhiza has the ability to release plant-derived carbon to microbial community, such as some bacteria and protozoa. Thus, this results in higher arbuscular mycorrhiza density and reduced bacteria, protozoa and nematodes community. Though not all bacteria are affected by enhanced atmospheric carbon dioxide level, for example, *Burkholderia* and *Pseudomonas* community is reduced while *Bacillus* and *Actinomycetes* are not affected. In addition, protozoa and nematodes were a community reduction from indirect grazing (DRIGO et al. 2010).

Arbuscular mycorrhizal fungi (AMF) normally form into a beneficial symbiosis with many plants to modulate the growth of host plants growing under biotic and abiotic conditions through direct and indirect mechanisms such as reactive oxygen species (ROS) and oxidative damage induced alleviation the mediation of phytohormone synthesis (HASHEM et al. 2019). Arbuscular mycorrhizal fungi have the potential to improve soil properties and thereby promote plant growth in both normal and stressed environments (HASHEM et al. 2018). The *Glomus* genus is one of the most abundant genera of arbuscular mycorrhizal fungi. Arbuscular mycorrhizal fungi produce symbiotic signals to stimulate better root growth, and branching (MALHI et al. 2021). Arbuscular mycorrhizal fungi (AMF) provide benefits to most crop species through improved nutrient uptake, increased resistance to drought and abiotic stress, and reduced action of pathogens and pests (SCHAEFER et al.2021). In the field, crop roots are colonized with multiple arbuscular

mycorrhizal fungi species that are difficult to separate and identify. The ability of mycorrhizal roots to take up phosphate in the field is thought to be a mosaic of the different abilities of different AMF (KOBAE 2019).

3.1.2. Arbuscular mycorrhizal fungi (AMF)

The symbiosis of mycorrhizal fungi and host plants has been important in the evolution of terrestrial plants and this relationship can be demonstrated in more than 80 % of vascular species (Santander et al. 2017). There are two main types of mycorrhizae; endomycorrhizae and ectomycorrhizae as shown in Figure1. Endomycorrhizae involve penetration of fungi towards cortical cells, development of arbuscules within the cells, and external mycelium extending from the arbuscules. On the other hand, ectomycorrhizae involves colonization of the area between cells called the Hartig net and development of a thick surface coat known as the fungal sheath (BARMAN et al. 2016).

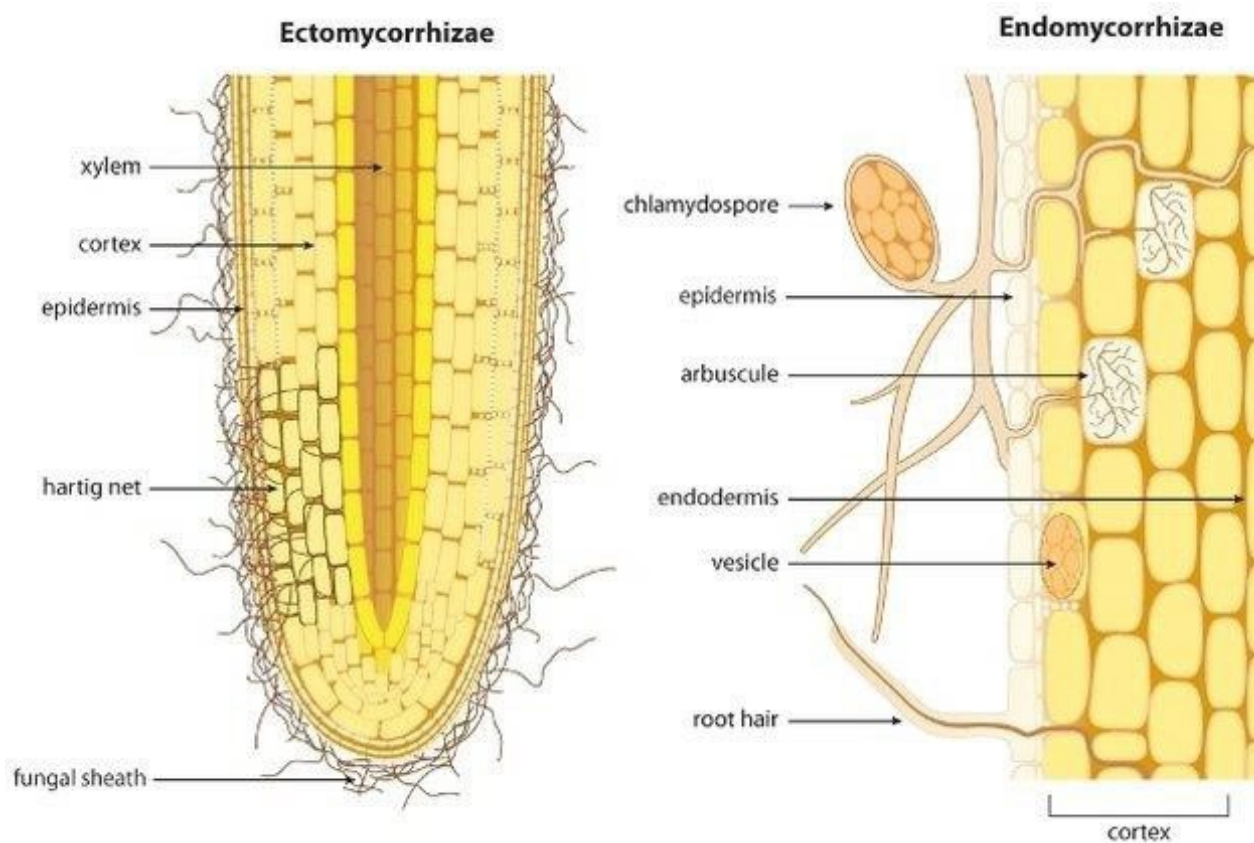


Figure 1. Schematic diagram on association of ectomycorrhizae and endomycorrhizae with the plant root (MCNEAR 2013).

Ectomycorrhizae are generally found among woody plants and have rarely been found in the herbaceous plant families (MARSCHNER 2012). Endomycorrhizae are further classified into arbuscular mycorrhiza, ericoid mycorrhiza, and orchid mycorrhiza, of which arbuscular mycorrhiza is the most common (MCNEAR 2013). To establish the association of the fungus with the root, the root will secrete root exudates that serve as signals for microbial recognition, and an example of these root exudates are strigolactones (LUGINBUEHL et al. 2017). Quercetin and strigolactone are secreted by plants to give signals for spore germination as well as the development of arbuscular mycorrhizal hyphal growth (MARSCHNER 2012; HAICHAR et al. 2014). Hyphal growth is important to ensure a large surface area between arbuscular mycorrhiza and plant, which increases the efficiency of nutrient exchange (SMITH et al. 2011).

Therefore, fungi colonize the root surface, penetrate the root cells to develop arbuscules on the inner cortex with formation of a periarbuscular membrane, and end with senescence and degeneration of the arbuscules (LUGINBUEHL et al. 2017). Because arbuscular mycorrhizal fungi live in symbiosis, they cannot live without the host plant. They have different morphological structures, such as: arbuscules, vesicles, hyphae and spores. These structures are produced in plant roots, but hyphae and spores can sometimes be produced outside the root system (SOUZA 2015).

3.1.3. The main structures of AMF and their functions

Hyphae

Intraradical hyphae (IH), this can transfer nutrients and water from outside to the root cortex of the plant. Furthermore, they also exchange these substances to obtain energy. They can differentiate as arbuscules, vesicles, or spores once they reach the cortical zone (BERBARA et al. 2006; AL-QARAWI et al. 2013). Other type of hyphae which is Extraradical hyphae (EH); there are extraradical hyphae that can grow from the soil to the root surface, others that can make nutrient uptake more important, and extraradical hyphae that can reproduce and give new spores (REDECKER et al. 2013; CRUZ et al. 2008).

Arbuscules

Arbuscules are formed from intraradical hyphae. They look like branching haustoria of a similar shape with small trees. The exchange between fungus and host plant takes place mainly in the arbuscules (SENA et al. 2004). During the symbiotic phase and hyphal growth, root colonization occurs and arbuscules form within 2 days (SOUZA 2015).

Vesicles

Vesicles are also formed from intraradical hyphae. They play an important role in the nutrient storage of the plant. In fact, they have high lipid and glycogen levels. Some species don't have this kind of special structures, but for the species that possess vesicles, they can rapidly increase their numbers after proliferation (BERBARA 2006).

Auxiliary cells

Auxiliary cells are specialized structures found only in some species of the order Diversisporales, formed from extraradical hyphae (REDECKER et al. 2013). The function of these cells is not well defined, but some authors have suggested that they play a role in nutrient storage (MORTON and BENNY 1990).

Spores

They can be of different origin: extraradical hyphae, intraradical hyphae or vesicles, which are asexual spherical structures of the fungi. There can be different places for the formation of the spores, for example in the soil surface for example *Glomus*, in the roots such as *Rhizophagus* and in the soil for example *Funneliformis*. They can be formed in clusters such as *Diversispora* and sporocarps for example *Sclerocystis* (SOUZA 2015).

3.1.4. Application of the arbuscular mycorrhizal fungi in agriculture

The production and application of microbial fertilizers is increasing worldwide due to the negative effects of excessive or improper use of chemical-based fertilizers and increased awareness of the relationship between rhizosphere microorganisms and plants (IGIEHON and BABALOLA 2017). The multiple benefits of arbuscular mycorrhiza have increased the possibilities for its commercial application. As a result, arbuscular mycorrhiza-related markets have grown significantly over the past few decades, with an increasing number of actors, products and market volume (CHEN et al. 2018). Arbuscular mycorrhizal fungi (AMF) are primarily used in the production of bioinoculants as they are known to form symbiotic relationships with more than 85 % of plant species of agricultural interest (BASIRU et al. 2020).

Arbuscular mycorrhizal fungi have historically played a prominent role in the development of plant development and are an extremely important symbiotic organism for arable farming with the promotion of plant growth, soil stability and improvement of soil quality (WILKES 2021).

Since the 1990s, the number of companies selling mycorrhizal products has increased significantly. On a global level, the main producers are located in North America, Europe, Asia and Latin America

(CHEN et al. 2018). Global demand for arbuscular mycorrhizal fungi in mycorrhizal-based industrial market was 268.8 million US \$ in 2019 and is projected to grow to 621.6 million US \$ by 2025, at an estimated growth rate of 14.8 % over the next 5 years (Market Growth Trends for Mycorrhiza-based Biofertilizers and forecast 2020-2025) (SRIVASTAVA et al. 2021).

Organic microbial fertilizers for agricultural purposes are also known as bioinoculants. They can be generally defined as formulations of active or latent strains of microorganisms, mainly bacteria, either alone or in a mixture with algae or fungal components, which actively or passively enhance microbial activity and thereby increase the utilization of nutrients from the soil (SUYAL et al. 2016). Organic fertilizer is still an uncertain term. It is easy to see that Biofertilizers are classified into plant extracts, composted municipal waste and other microbial combinations with undetermined ingredients, and synthetic fertilizer formulations supplemented with organic chemicals (HARI et al. 2005). Biofertilizer use is known to have effects on the diversity of microbial communities, which in turn increases the diverse microbiome of plants. Biofertilizers also affect soil physico-chemical properties, pH, texture and organic matter, all of which have a significant impact on plant growth and development (RAIMI 2021).

3.2. Plant growth promoting rhizobacteria (PGPR)

PGPR are soil microorganisms that play an important role in promoting plant growth. Various mechanisms are included in this function, such as: nitrogen fixation, phosphate solubilization, and potassium solubilization (VERMA et al. 2018). There are two types of PGPR: extracellular PGPR (ePGPRs) and intracellular PGPR (iPGPRs). The ePGPRs are those located in the rhizosphere and/or in the inter root spaces. The iPGPRs are located in specific node structures of root cells. The iPGPRs belong to the *Rhizobiaceae* family, which includes *Allorhizobium*, *Bradyrhizobium*, *Mesorhizobium* and *Rhizobium*, endophytes that can symbiotically fix atmospheric nitrogen (VERMA et al. 2018).

3.2.1. Nitrogen fixation

For nitrogen uptake, PGPR converts the nitrogen into ammonium that can be taken up by the plants. Two types of PGPR can be distinguished in nitrogen fixation: symbiotic, when they live in plants and exchange metabolites with them; and non-symbiotic when they live outside the plant cells (VEJAN 2016). For example, *Paraburkholderia* showed the highest nitrogen fixation activity in root nodules, four weeks after planting of soya bean (PAULITSCH et al. 2021).

3.2.2. Solubilization of phosphate

In the case of phosphorus, most soils are deficient in phosphorus, as this is mostly bound in an insoluble form. PGPR plays a role in solubilizing phosphorus to make it available for plant uptake. PGPR uses different strategies to achieve this, such as: the production of extracellular enzymes, the release of phosphate during substrate degradation and the release of mineral-dissolving compounds (GUPTA et al. 2015). For example, *pseudomonads* are able to solubilize inorganic phosphate through the production organic acids, such as gluconic acid (GISLASON and KIEVIT 2020).

3.2.3. Potassium solubilization

PGPR also affects the uptake of other nutrients such as potassium and zinc, which are also important nutrients for plant growth. For example inoculation of PGPR into agricultural crops is common because of its important role in yield improvement. PGPR is commonly inoculated into sorghum, wheat, potatoes and sugarcane (VERMA et al. 2019).

3.2.4. Siderophore production

In order to absorb the element iron, there are mechanisms that help plants access this element. The microorganisms in the soil develop these mechanisms. They also produce compounds that form iron chelates. These compounds are called siderophores and responsible for the uptake of iron into the plant (JF 2005). For example, *P. brassicacearum* strains harbour NRPS clusters for synthesis of pyoverdines, 48H11 and 38D4 carry siderophore-encoding loci similar to those found in *P. Frederiksbergensis* (GISLASON and KIEVIT 2020)

3.2.5. Phytohormone production

PGPR also plays an important role in the production of phytohormones. These phytohormones such as cytokinins, auxins, gibberellins and ethylene are responsible for the growth

and development of plants (GUPTA et al. 2015) For example, bacteria including endophytes have evolved several pathways such as, siderophore with high affinity to scavenge and transport iron from the environments, the ability of endophytes to produce or capture siderophores, under iron stress condition, is one the most traits which provide iron to host plants (QASSIM et al. 2018).

antibiotic production

This ability is one of the most important properties of the PGPR to compete pathogens. PGPR can produce a very different type of antibiotic. They can be used as bioagents for phytopathogens (GUPTA et al. 2015). For example, *Pseudomonas fluorescens* strain G308 isolated from barley leaves produces a novel antibiotic substance such as N-mercapto-4-formylcarbostyryl (Cbs) mass isotope ratios analysis, these antibiotic compound is effective against many phytopathogenic fungi in vitro (FAKHOURI et al. 2001)

The mechanisms of action of the PGPR are summarized as shown in Figure 2 below:

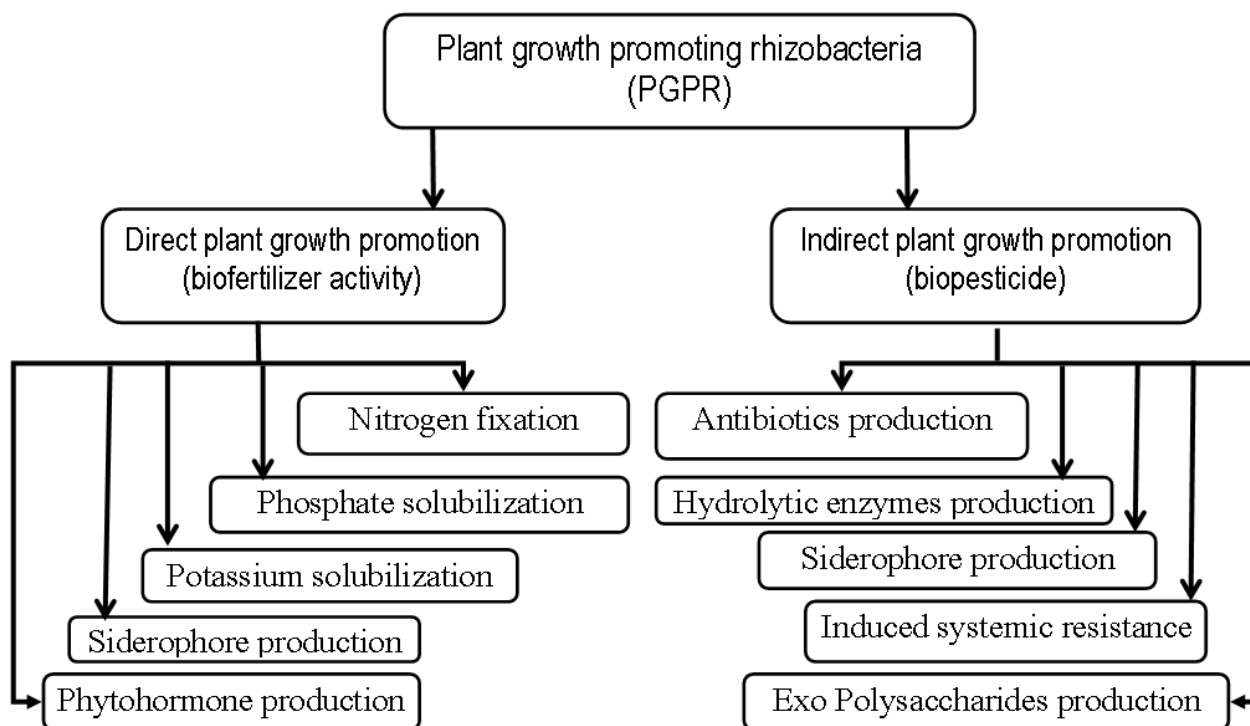


Figure 2. Mechanism of action of PGPR (GUPTA et al. 2015).

Based on the study by NADEEM et al. (2013) here are some examples of strains used in agriculture:

- *Achromobacter piechaudii* increases fresh and dry weight and water use efficiency
- *Pseudomonas fluorescens*, which increases the growth of the plant
- *Pseudomonas spp.* enhances seed germination and seed growth
- *Pseudomonas brassicacearum* enhances root growth and nutrients uptake.

3.3. *Trichoderma* species

Fungi of the genus *Trichoderma* are soil-borne ascomycetes with green spores found all over the world (SCHUSTER and SCHMOLL 2010). It can colonize different types of soils in different climate zones and can be found in soils with different types of crops (ROIGER et al. 1991). *Trichoderma* spp. is a commonly used biocontrol fungi that has been extensively investigated using genomes and metabolomics for example, *Trichoderma asperellum* strain T34 can control *Phytophthora capsici* in pepper (SEGARRA et al. 2013) These fungi have been studied for almost a century for their beneficial effects on plants and soil (LORITO et al. 2010). *Trichoderma* growth generally requires lower soil water content. In fact, too high soil water content can negatively affect spore production and germination as well as hyphal growth. Depending on the species, the soil temperature also affects the growth of *Trichoderma*. There are some species that can live in relatively cold temperatures, but also some species can live in soils with 40°C (CLARKSON et al. 2004). A study conducted by SINGH et al. (2014) observed that the *Trichoderma* species produced sufficient biomass at different temperatures viz. 20°C, 25°C, 30°C and 35°C but they were found to be best grown at a temperature range of 25°C to 30°C.

Colonization of roots by *Trichoderma* spp. frequently stimulates root development and expansion, hence increasing crop productivity, several *Trichoderma* species have the ability to promote plant growth and productivity by utilizing overlapping modes of action including induced systemic resistance, antibiosis, enhanced nutrient efficiency (LEE et al. 2016) Clearly, the most successful strains are rhizosphere-competent. These reactions are typically triggered by direct plant contact, reduced root microbial activity, or inactivated toxic compounds. Additionally, *Trichoderma* spp. can break down minerals in the soil and increase nutrient absorption (HARMAN et al. 2004). Research has shown that *Trichoderma* can be a powerful biological control agent for soil pathogens. For example; *Trichoderma atroviride* can reduce the *Rhizoctonia solani* in the legumes (KANDULA et al. 2015). *Trichoderma* also produces antibiotics useful for biological control of pathogens, *Trichoderma* spp. produce a large number of compounds with antibiotic activity such as aldehydes, ethylene, ketones, and diketopiperazine-like gliovirin and gliotoxin, *Trichoderma harzianum* showed significant antimicrobial activity against the *Bacillus subtilis* (Sharma et al. 2019) In addition to mycoparasitism and antibiotic production, *Trichoderma* can also take advantage of competition.

3.4. Important vegetables with Tuber

Vegetables can be classified according to their botanical origin, their hardiness or temperature, as well as the parts of the plant used, i.e. leaves, fruits or roots. Root vegetables include carrots, radishes, potatoes, yams, ginseng, celery, parsley, and horseradish (KENZ et al. 2022). Vegetables with tubers and roots exhibit different colors that usually depend on the presence of three main classes of compounds, namely flavonoids, betalains and carotenoids, which can determine their visual appearance and consumer perception (PETROPOULOS et al. 2019).

Sweet potato (*Ipomoea batatas* L. Lam.) belongs to the *Convolvulaceae* family (ALHADIDI et al. 2021). A versatile and nutritious crop is cultivated worldwide. Its tuberous root is commonly used as a food source, while the aboveground biomass, known as sweet potato vine, serves as a by-product of farming (ZHANG et al. 2023). According to data from the Food and Agriculture Organization, sweet potato cultivation takes place in 117 countries, with Asia contributing nearly 80% of the annual production (CONTINI et al. 2019). Sweet potato originates from Central America and it has been cultivated widely in tropical and subtropical countries like Malaysia, Indonesia, China, USA and Japan (CHEN et al. 2003; NEDUNCHEZHIAN et al. 2012). In Malaysia, for example, they have long been cultivated and have become one of the country's most important vegetable crops because they are a cost-effective source of energy, carotene, ascorbic acid, niacin, riboflavin, thiamine and minerals (ALHADIDI et al. 2021). Recently, temperate countries like Hungary have started cultivating it (SZARVAS et al. 2017). The increasing demand for sweet potatoes has also encouraged growers and led to an increase in sweet potato acreage across Hungary (SZARVAS et al. 2018). However, little is known about its cultivation in this climate. According to FAOSTAT 2017 81.3% of sweet potatoes are produced in Asia, most of it in China as shown in Figure 3, the top 5 countries for sweet potato production over these nearly two decades are China, Malawi, Nigeria, Uganda and Indonesia as shown in Figure 4. Thus, most of Asia's production is monopoly by China (FAOSTAT 2017)

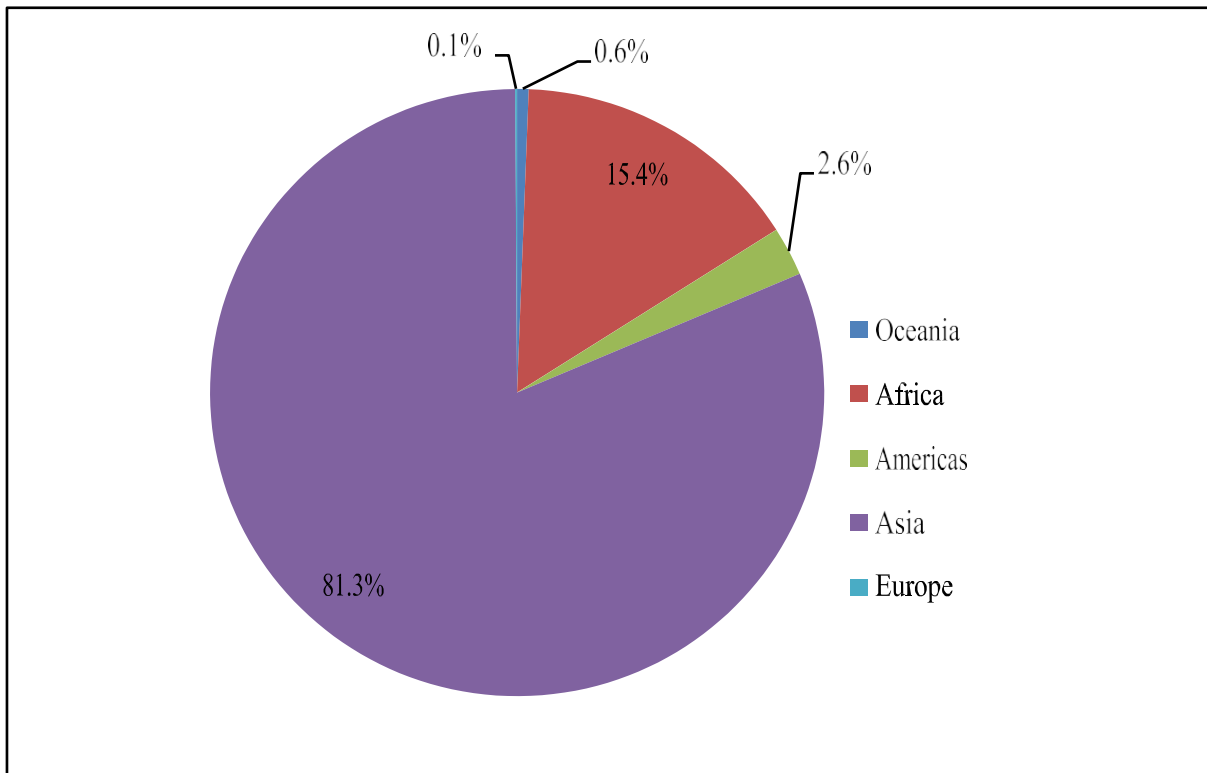


Figure 3. Production share of sweet potato by region at average between 1994-2018 (FAOSTAT, 2017).

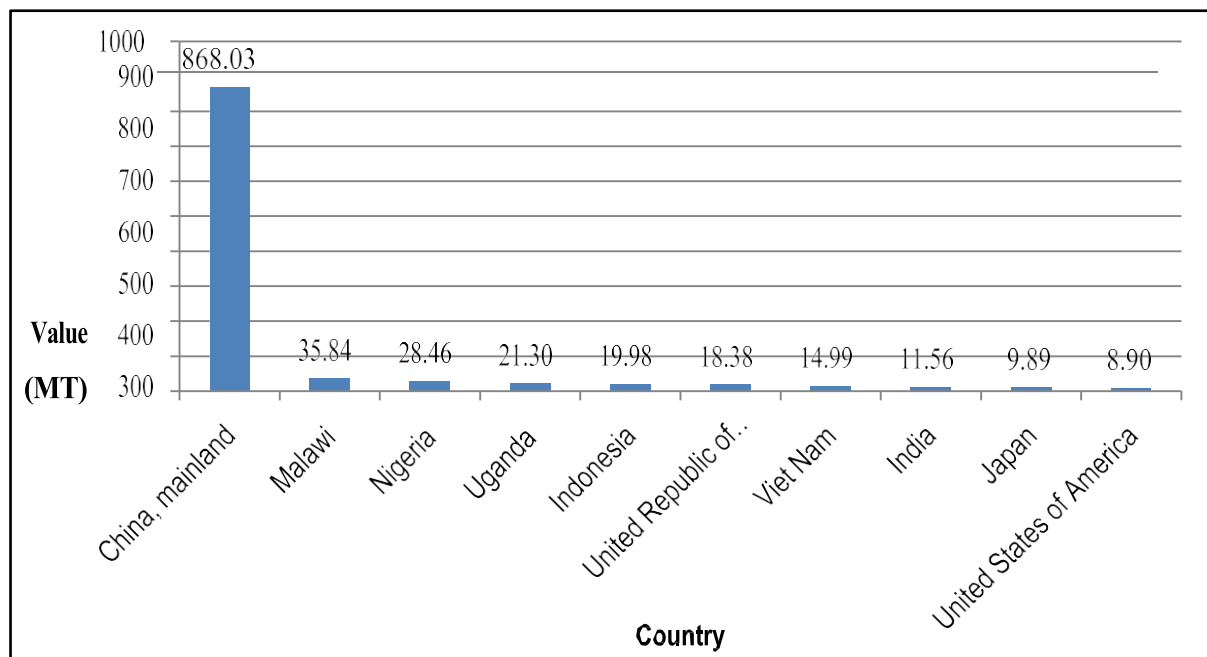


Figure 4. Top countries in production of sweet potato at average between 1994-2018 (FAOSTAT, 2017)

3.4.1. Characteristics and nutritional value of sweet potato

3.4.1.1. Characteristics of sweet potato

The morphology of the flowering plant is generally divided into two parts; shoot system and root system as shown in Figure 5. Sprouting system for sweet potato, which is a dicotyledonous plant, includes apical shoots, leaves, flowers, inflorescences, nodes, internodes and stems. In general, leaves are designed to maximize their exposure to sunlight and photosynthesis, flowers are for pollination, and stems are primarily for the movement of water and food throughout the plant system (TERENCE et al. 2015a). The ability to produce flowers and produce fruit with seeds gives this plant the benefits of enhancing its genetic diversity, particularly for environmental adaptation and survival (ANTONIO et al. 2011). The sweet potato root system includes the fibrous root, pencil root, and storage root. Roots are important for anchoring the plant body, absorbing water and minerals from the soil, storing food and drawing food and water from the soil, and maintaining the scion system (TERENCE et al. 2015b).

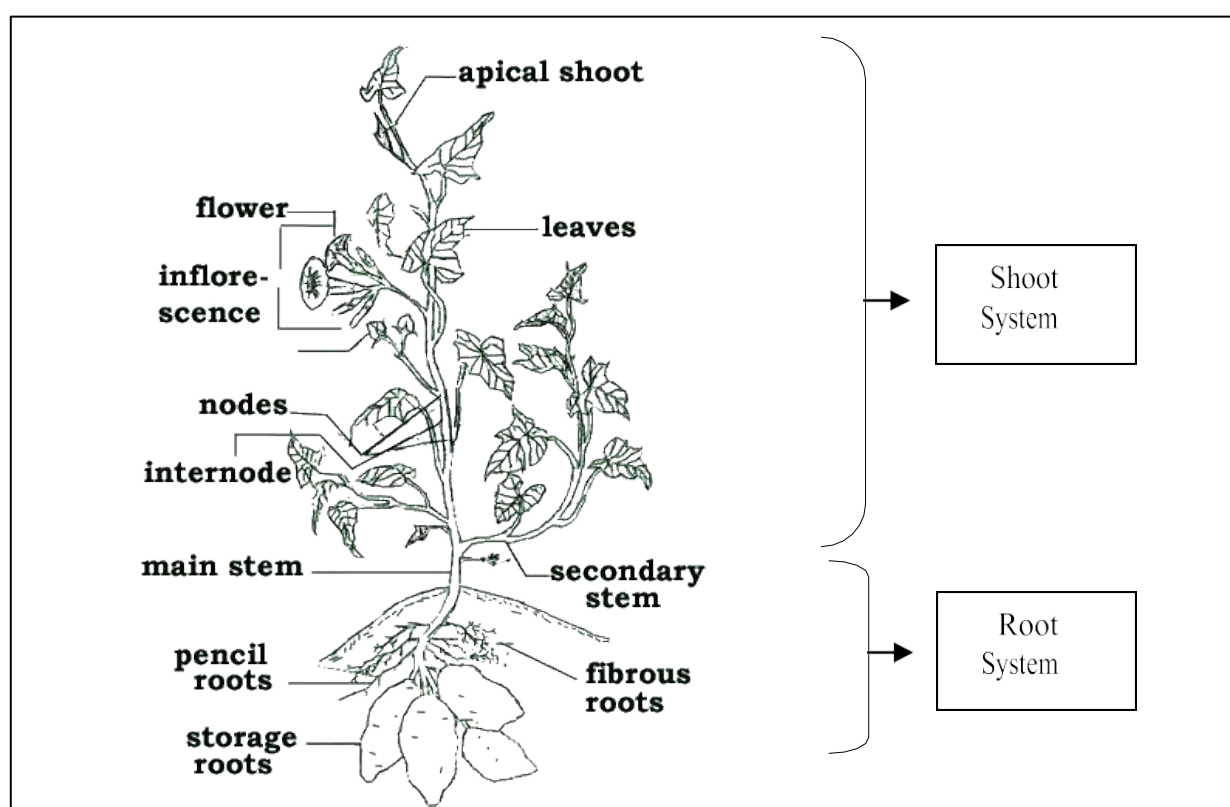


Figure5. Sweet potato plant morphology (HUAMARI 1992)

Sweet potatoes have a moderate twining ability and plant cover with soil could spread up to 151 to 250 cm. The ground cover is high, about 75 to 90 %. The length of the internodes is about 6 to 9 cm while the diameter is about 10 to 12 mm. Mature leaf shapes can be rounded and

triangular. Their mature leaf size is considered large, ranging in size from 16 to 25 cm. The shape of the storage root also differs from variety to variety, they can be round elliptical, elliptical and long elliptical (MBITHE et al. 2016).

3.4.1.2. Nutritional value of sweet potato

Table. Nutritional value of sweet potato

Vitamins (LOEBENSTEIN et al. 2009)	A, C
Sugar (ANTONIO et al. 2011)	Glucose and Fructose
Antioxidants (SZARVAS et al. 2019)	Anthocyanin, Carotenoids
Minerals (AMBIKA et al. 2019)	Potassium, Sodium, Manganese and Iron

3.4.2. Sweet potato varieties

There are many varieties of sweet potatoes around the world and each country has its own varieties. More than 50 cultivars have been found in the United States, including Beauregard, Jewel and Garnet, some of the famous cultivars there (JACKSON et al. 2013). On the other hand, sweet potato was an important crop in China, more in 2000 varieties can be found in China, and some of their typical types are XuShu18, SuShu2 and SuShu8. In China, sweet potatoes can be divided into three groups, a general kind, a high-starch kind and an edible kind (CHEN et al. 2003). Sweet potatoes could also be distinguished by their flesh color. The most common are potato flesh-colored, ranging from white to yellow (NEDUNCHEZHIAN et al. 2012). Aside from the color of the fleshed and the skin of the peel, they have other differences (LOEBENSTEIN et al. 2009). Sweet potatoes with purple flesh have more moisture, protein, starch and fiber than sweet potatoes with orange flesh. On the other hand, total carbohydrates are higher in orange-fleshed sweet potatoes than in purple-fleshed sweet potatoes (RODRIGUES et al. 2016). Under normal conditions, the physical parameters of sweet potatoes of different colors are not much different. However, when stressed at a temperature of 90°C, purple and orange-fleshed potatoes have higher swelling power than white-fleshed sweet potatoes, and purple-fleshed have higher water solubility than orange-fleshed sweet potatoes (LEE et al. 2017). Orange-fleshed cultivars are among the important sources of β -carotene due to their high concentration of provitamin A carotenoid, while purple-fleshed cultivars have been reported to have high concentrations of polyphenolic and anthocyanin components (CHASSY et al. 2008; STEED et al. 2008).

3.4.3. Sweet potato seedling propagation

Seed root selection involves the process of properly selecting, curing, and storing sweet potato roots for making cuttings. Offshoots, also known as vine cuttings (MASABNI et al. 2014) as shown in Figure 6. This is important to ensure high quality cuttings that will ultimately lead to the establishment and production of high-yielding sweet potato crops (BRANDENBERGER et al. 2014). As bedding, sweet potatoes prefer loose, sandy or mixed of them that could help the development of a good root, so rocky and compact soils should be avoided (ANTONIO et al. 2011).



Figure 6. Sweet potato seedling propagation by vine cutting (<https://www.flowerpatchfarmhous.com/propagate-sweet-potato-vine/> last accessed on 2024.02.19).

Another method of sweet potato seedling cultivation, production of slips that involve in the process of preparing the bed for pre-sprouted roots, embedding the pre-germinated roots and caring for them as shown in Figure 7. Bedding preparation is important to ensure that potential diseases and pests are removed from the pre-germinated roots. Dipping the pre-germinated roots with fungicides, helps avoid surface infection with black rot, scab, and root rot (BRANDENBERGER et al. 2014). The knives used should be disinfected (ROOT 2010).



Figure7. Sweet potato propagation by slips (<https://www.sciencebuddies.org/> last accessed on 2024.02.19).

3.4.4. Soil preparation and transplanting

Loose friable soil is important for good production to allow storage roots to develop unhindered (ROOT 2010). A good approach is to mix the fertilizer and soil two weeks before transplanting to allow the soil to settle. The transplanting procedure should be carried out immediately after pulling the cutting from the roots. It is recommended to plant the cutting in moist, soilless growing media first if several days are required before the cutting can be transplanted. Remove weak and thin slips to ensure a good result. The slide depth in the ground should be at least three knots long (BRANDENBERGER et al. 2014).

Another important factor in the success of slip transplant is irrigation. It is important to ensure that watering after transplanting is done at the same time. Cuttings are very sensitive to lack of water, especially in the first month after transplanting (MASABNI et al. 2014). If a transplanter is used, it is recommended that the system be set to water the seedlings immediately after transplanting for each operation. On the other hand, if a drip irrigation system is used, the system should be installed before transplanting and leave the system running while the transplanting process is performed (BRANDENBERGER et al. 2014). For harvest, it is important to note that irrigation should be stopped 2 to 3 months before harvest to avoid damage to the storage root (MASABNI et al. 2014).

In South Africa, it is recommended to start transplanting in mid-November to December in frosty areas, but transplant in January to March in frost-free areas. Sweet potato growth can be affected even by light frosts and they need 4-5 months high temperature period to ensure good production. However, too high temperature could still reduce storage root formation, so it is important to avoid November to February in hot areas (DAFF 2011). For less demanding crops such as green vegetables, crop rotation should be used to increase previous mineral fertilization, since sweet potatoes need time to react with fertilizer (ANTONIO et al. 2011).

3.4.5. Ecological demands

Sweet potatoes are usually planted preferentially in peat or sandy soil (FARZANA et al. 2005). They could also grow on loam and clay loam soils and sandy loam. However, among them, sandy loam with loamy subsoil is best for growing sweet potatoes. Heavy clay soil restricts root development, while sandy soil encourages root expansion into deep soil (NEDUNCHEZHIAN et al. 2010). In terms of soil pH, the optimal soil pH best for growing sweet potatoes is between

5.5 and 6.5. Alkaline soils favor scab diseases, while acidic soils make them suffer from aluminum toxicity (NEDUNCHEZHIAN et al. 2012). Concerning to the heat demands, the Amylose content, enzymatic digestibility and the structure of amylopectin were strongly influenced by soil temperature. As shown in Table 2, low amylose content of starch was observed in sweet potatoes grown at lower soil temperatures compared to higher temperature soils. These results indicate that the influence of developmental temperature on amylose synthesis varies with plant species (NODA et al. 2001).

Table 2. Starch content and amylase content in Ayamurasaki (Purple fleshed cultivar) and Sunnyred (Orange fleshed cultivar) cultivated in a temperature-controlled greenhouse at four different soil temperatures (NODA et al. 2001).

Cultivar	Soil temperature (°C)	Starch content (%) ^a	Amylose content (%) ^b
Ayamurasaki	15	20.4	12.8
	21	26.9	14.6
	27	31.8	15.8
	33	21.9	17.3
Sunnyred	15	20.9	15.6
	21	31.5	16.8
	27	27.4	19.9
	33	26.8	20.6

^a Values are means of four determinations, Standard deviation ± 1.4 %

^b Values are means of three determinations, Standard deviation ± 0.6 %

Sweet potatoes are drought tolerant but sensitive to waterlogging (NEDUNCHEZHIAN et al. 2010). Water irrigation is not required during the early season of sweet potato as generally effective rain is higher than crop evaporation, except in August as late season is approaching; therefore, a little irrigation is required. High irrigation is required in the post-season as effective rain is very low (OPAFOLA et al. 2018). As for nutrient demands, Nitrogen (N) is important for plant growth and plays an important role in yield and nutrient composition of tubers, including sweet potatoes. The supplied nitrogen correlates positively with the carotenoid, dry matter and protein content of the sweet potato (UKOM et al. 2009). A study showed that increasing nitrogen supply from 50-100 kg ha⁻¹ increased lateral roots by 32 % compared to increasing nitrogen supply from 0 -50 kg ha⁻¹. Aside from that, this supplied N rate also increases adventitious roots by 65 %. However, further increases would not bring benefits for lateral and adventitious root development (VILLORDON et al. 2013). Also, phosphorus (P) is needed by sweet potatoes because it helps in the activity of cell division, respiratory mechanism, transport of ions across cell membranes, photosynthesis, energy production, transfer and storage of nutrients in plants, protein and nucleic acid synthesis. Phosphorus is also an important agricultural factor to ensure that sweet potatoes

grow faster, crop maturity increases, starch synthesis activity increases and high root development occurs (Abdel-Naby et al. 2018). The phosphorus content correlates positively with the swelling capacity of the starch, but not with the solubility (KARIM et al. 2007). Potassium (K) contributes to promoting good development of plant growth and tuber yield. In addition, it also helps in the activity of energy transport, water, photosynthesis, translocation of assimilates and protein uptake (Abdel-Naby et al. 2018). When potassium and zinc were applied at the highest concentration 150kg K₂O/fed and 30 ppm zinc had shown an increase in vegetative growth, yield, and root quality (El-BAKY et al. 2010).

3.4.6. Potato cultivation

Potato (*Solanum tuberosum* L.) is a perennial plant belongs to *Solanaceae* family (MULETA and AGA 2019). A short day vegetatively propagated C₃ plant cultivated in temperate, subtropical and tropical regions (MALLICK et al. 2021). Potatoes are originally from the southwestern United States to central –Argentina (BRADSHAW and RAMSAY 2009). They were first domesticated in the Andes. In the 1990s, Europe, North America and some countries of the Soviet Union were the largest consumers of potatoes. In Europe, the potato was introduced from the Andean highlands of Bolivia, Peru and northern Argentina. Later there were also potatoes from Chile (MACHIDA- HIRANO 2015). Potato production worldwide was about 365 million tons in 2018 and about 370 million tons in 2019 as shown in Figure 8. The Asian continent was the first potato producer in 2019 with 189,800,000 tons of potatoes produced (FAOSTAT 2019).

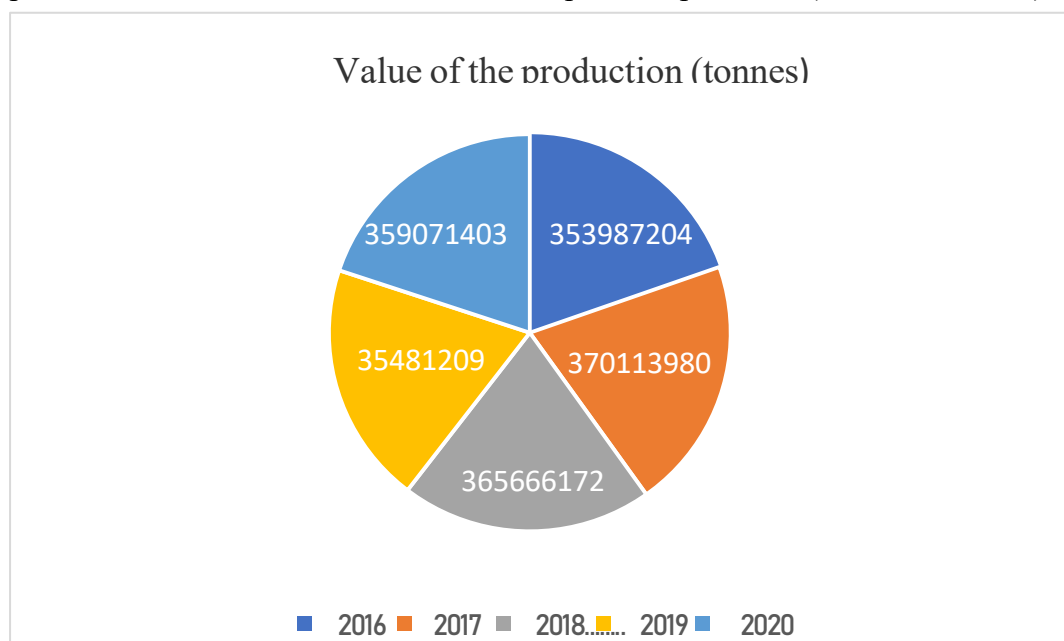


Figure8. world production of potato in the world for 5 years (FAOSTAT, 2022)

3.4.7. Taxonomy of potato and botanical characterization of potato

Taxonomy of potato plant

Table 3. Taxonomy classification of cultivated potato (BRADEEN et al. 2011).

Family	Solanaceae
Subfamily	Solanoideae
Tribe	Solaneae
Genus	Solanum L.
Subgenus	Potatoe (G.Don) D' Acry
Section	Petota Dumortier
Subsection	Potatoe G. Don
Superseries	Rotata Hawkes
Series	Tuberosa (Rydb.) Hawkes
Species	Solanum tuberosum L.
Subspecies	Tuberosum

Potato plant botany characteristics

The potato leaves are compound; consist of a petiole continued by a midrib bearing leaflets as shown in Figure 9. There are three distinct leaflets; the terminal leaflet at the outermost part of the leaf and the primary leaflets interspersed with secondary leaflets (DE JONG et al. 2011). Potatoes have bisexual flowers, including pistils (female) and stamens (male). They are naturally pollinated by insects especially bumblebees. The color of the corolla can vary between white, red, blue and purple (DE JONG et al. 2011). In some varieties, fruits also grow in the plants in addition to tubers. These fruits contain about 25 to 200 seeds. When the fruit is ripe, the seeds can be extracted and dried and used as planting material (DE JONG et al. 2011). Potato tubers are underground stem, stolon grows horizontally, and the tubers are formed at the terminal end of the stolon. The uncovered runners develop into vertical stems. It has a color that varies from white to yellow. The shape of the tuber generally varies from oblong, oval, kidney-shaped or spherical (WERNER 2020).

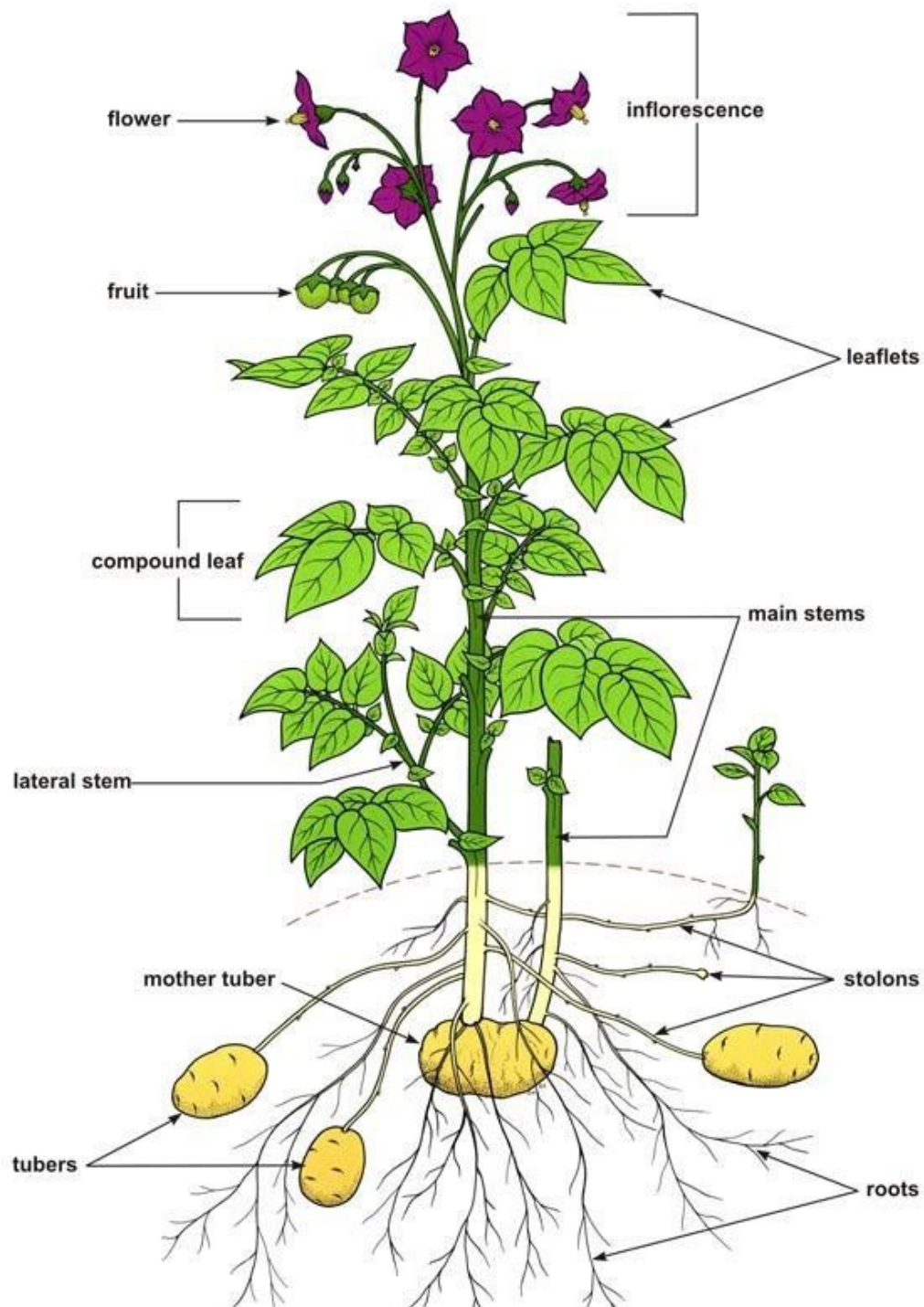


Figure9. Potato plant parts (<https://cipotato.org/potato/how-potato-grows/> - international potato center CIP, last accessed on 2024.02.18)

3.4.8. Phenology of potato plant according to BBCH scale (MEIER 2018)

Tabla 4. Potato plant phenology according to BBCH scale(MEIER 2018)

Description	Code
Sprouting/ Germination	00-09
Leaf development	10-19
Formation of basal side shoots- below and above soil surface (main stem)	20-29
Main stem elongation	30-39
Tuber formation	40-49
Inflorances emergence	50-59
Flowering	60-69
Development of fruit	70-79
Ripening of fruit and seed	80-89
Senescence	90-99

The plant types of different potato cultivars can vary greatly, when compared to indeterminate types, determinate types are typically shorter, have fewer flower clusters, and mature earlier. Conversely, indeterminate types require a longer growing season, which could result in a higher yield (DE JONG et al. 2011). An "inflorescence," or flower, is typically produced by potato plants after they have formed 10–13 leaves. There are big differences between varieties in terms of flowering time and amount (STARK et al. 2020). Potatoes are known to develop a sympodial shoot, which permits the development of multiple levels following the termination of the lower-order levels, or main stems, into inflorescences, for potato (*Solanum tuberosum* L.) different plant phenological scales originating from seed tubers have been established (KACHEYO et al. 2020). Berries from potato plants can contain from 100 to 400 seeds. It is currently not possible to propagate genetically identical plants from seeds for agricultural use because these seeds are heterogenic, meaning that their progeny differ genetically from their mother plant (SIBERT et al. 2019).

3.4.9. Propagation materials

Most often, potatoes are grown by vegetative propagation using the whole tuber or a cut piece of tuber. But using seeds (from the fruit) can also be a method of growing potatoes (DE JONG et al. 2011). It is also possible to grow potatoes by planting real seeds. The ripe fruits contain seeds, and when dried, these seeds can be used as planting material. The use of real seeds as planting material requires certification. By using real seeds as planting material, the quality of the parents for the next generation will not be the same but will be poorer, which is a disadvantage of planting with real seeds. Even if the seeds come from the same fruit, the characteristics of the cultivated plant are not the same (DE JONG et al. 2011). Growing with tubers is the most commonly used method as it is easier to practice. Planting true seeds can take a long time to obtain new plants as it requires several months of initial growth. This method is commonly used to create new varieties (DE JONG et al. 2011).

3.4.10. Potato varieties

Potatoes have a very large variety of varieties. Many parameters can be considered to distinguish the varieties. These varieties have characteristics that vary according to skin and flesh color (white skin, white flesh; yellow to white skin, yellow flesh, red skin, white flesh, etc.) or dormancy or germination (very early, long dormancy, etc.) (DE JONG et al. 2011). In culinary terms, there are three different categories of potatoes: Starch potatoes have less water content and more starch. More are used for frying, boiling or baking; Waxy potatoes, which have higher water content and are harder compared to starch potatoes, are typically used in cooking and salad preparation because they become difficult to crisp when fried. The third potato category is the all-purpose potato, which is a cross between the starchy potato and the waxy potato. This species is easy to cook as it can be used for all types of cooking.

3.4.11. Nutritional values and inner contents

Potatoes contain various nutrients that are important for health. The potato contains high-quality protein (lysine content), which makes the potato one of the vegetables with the best protein quality. In addition, it is a good source of various minerals and vitamins, making it a health-promoting vegetable (NAVARRE et al. 2014). However, some studies have shown that potatoes contribute to obesity when consumed in excessive amounts. The reason for this is the high content of starch, a complex carbohydrate. Starch contributes to energy transport and the dry matter of potatoes makes up about 80 % starch. The glycemic index value, which is a measure of the increase

in blood sugar, is high in potatoes. Therefore, potatoes may contain various nutrients important for health, but eating them in moderation is recommended to prevent the risk of diabetes (BRADSHAW AND RAMSEY 2009, NAVARRE et al. 2014).

Table 5. Inner contents in potato tuber.

Starch	Amylose and amylopectin	13-20 % (NGOBESE et al. 2017)
Sugar	Glucose and Fructose	1- 5 % (NGOBESE et al. 2017)
Lipids	Phospholipids	0.1-0.5 g/100g (GIBSON and KURILICH 2013).
Vitamins	C B6	50mg/100g 0.45-675mg/100g (NAIDU2003)
Protein	Aminoacids such as glutamic and aspartic acid	30-50% (WICHROWSKA et al. 2020).
Minerals	Potassium Phosphorus Magnesium Calcium	150-1386 mg/100g 42-120mg/100g 16-40mg/100g 2-20mg/100g (CAMPOS and ORTIZ 2020).

3.4.12. The ecological demands of potato

The potato prefers cool temperatures. A temperature between 18-20°C is optimal for the growth of the tubers. According to studies, the high temperature reduces yield by inhibiting starch synthesis (HOEKSTRA 2008). The water requirement of the potato increases with the development of the plants. After planting, the plant does not require much water. When the maximum canopy development is reached, the water requirement stabilizes. The optimal amount of rainfall for potatoes is 650–800 mm annually; with 60–70% of this amount falling during vegetation (DVOŘÁK et al. 2016). The ideal soil pH for growing potatoes is a slightly acidic pH around 6. At this pH most nutrients are available. Below pH 5.5 some macronutrients may not be available to plants, and above pH 7 the availability of phosphorus, zinc and magnesium will decrease (DE JONG et al. 2011). The nutrients which should be incorporated in the soil depend

on the nutrient need of the plant. For the potato, the approximate amount of nutrients which the plant uptake for 10 tons of tubers were; 40-50kg nitrogen, 8.8kg phosphorus, 22kg potassium, and 8.4kg magnesium (DVOŘÁK 2016).

3.5. Organic cultivation of tuber type vegetables

Due to increasing consumer awareness of health and environmental issues, the demand for safe organic food has increased significantly worldwide in recent years, offering producers and exporters in developing countries opportunities to improve their incomes and living conditions (FAROUQUE and SARKER 2018). For instance, production of organic potato has increased in the European and non-European countries from 2007 to 2008 as shown in Table 6. Agricultural producers must switch to organic farming, this applies in particular to the technologies for the production of vegetables, fruits and dairy products. Scientists have proven that the human body development can improve by 30-40 % by eating organic vegetables grown in compliance with environmental standards (EZOV et al. 2020). Industry interest in organically grown sweet potatoes has increased in recent years; however, organic sweet potato production is believed to be well below demand (NWOSISI et al. 2017).

The high commercial value of the product prompts potato growers to adopt extensive agricultural measures, which have undoubtedly been responsible for higher early potato yields in recent decades. Furthermore, with conventional cultivation, undesirable residues can occur both in the tubers and in the soil. This has increased the crop share of new potatoes from organic producers, also because organic new potatoes can be sold at a significantly higher price than the conventionally grown equivalents, because their quality is considered higher.

However, the improved qualitative value of organic compared to conventional products could not be determined. Controversial and opposite data differences between organically and conventionally grown nutrients and antioxidant compounds were found in the main crop potato tubers (IERNA et al. 2022). The inner content of the potato is strongly influenced by the cultivation technique. Organic farming helps the soil meet the plant's nutritional needs and improves soil properties. For example, using compost helps improve soil properties while nutrients by increasing the exchangeable amount of potassium and phosphorus availability (RAMLI et al. 2020). Compared to conventional production, the yield in organic cultivation is lower (MOURO et al. 2008). However, the potato quality is better in organic cultivation, for example, the culinary properties of the organic potato are better (IERNA et al. 2022). According to some studies, the dry

matter content is also higher in organic potatoes than in conventional cultivation (DANGOUR et al. 2009).

Various methods are used for plant protection in organic farming, such as cultivation techniques; crop rotation, organic soil improvement, use of sanitized seeds, choice of variety, and plant extract and inoculation with microorganisms (SOUZA 2015). . Some studies showed that the application of bio fertilizers and bio pesticides to potatoes has positive effects on their production (IERNA et al. 2022).

Table 6. Production of organic potato in some European and non-European countries from 2007-2008 (CANALI et al. 2012).

Country	Surface of organic potato (ha)	Organic potato in total organic production (%)	Organic potato in total potato production (%)
Denmark	1268	0.84	3.08
Germany	8150	0.9	2.96
UK	3270	0.44	2.35
Netherlands	1271	2.52	0.79
Poland	1861	0.59	0.33
USA	3348	0.17	0.73
South Africa	398	0.91	0.69
Canada	447	0.07	0.28
Turkey	16	0.12	0.09
Morocco	50	1.45	0.08

4. MATERIALS AND METHODS

4.1. Sweet potato pilot study

4.1.1. Origin, growing of the experimental seedlings

The experiment carried out from 6th march until the 23th of July 2019 at Buda Campus of the Hungarian University of Agriculture and Life Sciences (MATE) in the experimental glasshouse of the Department of Vegetable and Mushroom Growing (47.28° N, 19. 04° E). At the beginning of March 2019, two sweet potato varieties, orange "Norangel" and purple "Purple" (characterized by mature tuber and no flowers or buds were appeared on these tubers), these tubers were provided from Soroksár experimental research farm, located at the Hungarian University of Agriculture and Life Sciences Vegetable farming unit, they were propagated by tuber for seedlings production as shown in Figure 10, and planted in individual pots, the irrigation were carried out around once a week. However, the frequency of irrigation also depended on humidity and climate avoiding to be infected by some kind of pathogens. The sprouting was waited until they reached the length of 30-40 cm. This stage was reached in the 21th of May 2019, so the seedlings were ready to be transplanted. The seedlings were grown in a plastic pot (1.5 L, 11.5 cm× 12 cm×15cm). For seedling production, non-treated ground/minced peat) was used as a growing medium from Latagro Basic Substrat KB2 type (white peat 100% (0–10 mm) with specification: pH value (H₂O)

6.4; soluble nutrients available to the plants: Nitrogen<7 mg/L; Phosphate <7.8 mg/L; Potassium oxide<40 mg/L).

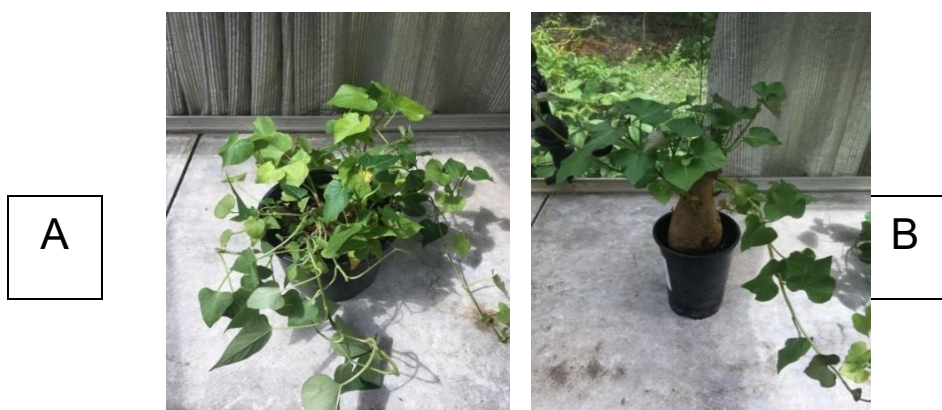


Figure 10. Sweet potato varieties orange; {A} "Norangel" and {B} purple "Purple" Department of Vegetable and Mushroom Growing experimental glasshouse in Buda campus (2019)).

4.1.2. Mycorrhizal inoculation

Mycorrhizal inoculation carried out at 21th of May 2019 on the transplanted seedlings of sweet potato varieties orange and purple. It was performed with Symbivit (Symbiom Ltd. Product from Sazava 170, 563 01 Lanskroun, Czech Republic), mycorrhizal, composition: five species of mycorrhizal fungi (*Claroideoglomus etunicatum*, *Claroideoglomus claroideum*, *Rhizophagus irregularis*, *Funneliformis geosporus* and *Funneliformis mosseae*), in addition to natural minerals, sapropel, extracts from sea organisms, natural keratin, humates and powdered biodegradable water-storing polymer granules. The bioadditive part represents 110 g in each kg of product. For 1 L of the substrate, 15 g of Symbivit was mixed. The experiment was made with three treatments for each variety in three replications; in each treatment seedlings were used with a total of 45 as shown in Figure 10.



Figure 11. Department of Vegetable and Mushroom Growing experimental glasshouse in Buda campus (2019)

In each pot, one seedling was used. As a control, the soil from Soroksar experimental research farm, located at the Hungarian University of Agriculture and Life Sciences organic farming units was used in which previous research experiments had already shown mycorrhizal colonization (FEKETE et al. 2020), (pH (KCl) 7.45; NO₂+NO₃-N (mg/kg) 71.6; P₂O₅ (mg/kg) 577; K₂O(mg/kg) 166). Table 7 shows the code of the treatments. There was no additional nutrient replenishment during seedling cultivation. For the inoculation experiment, half of the peat moss was sterilized to detect if the sterilization has any influence on the mycorrhizal inoculation efficiency. Peat moss was sterilized at the mushroom lab in the Department of Vegetable and Mushroom Growing by a trade Raypa steam sterilizer at a temperature of 121 °C for 20 min. For the sterilized treatments hygiene procedure was carried out during the transplanting by wearing rubber gloves to decrease the risk of contamination with the pathogens in the sterilized media.

Table 7. Treatments used during the experiment per variety at the Department of Vegetable and Mushroom Growing experimental glasshouse in Buda campus 30th May 2019

Substrate	Number	Treatment Code	Treatment Description
Non-Sterilized peat moss	1	(L+SYM)	Non sterilized peat moss with mycorrhizal inoculum
Sterilized peat moss	2	[(L+SYM). S]	Sterilized peat moss with mycorrhizal inoculum
Soil from experimental farm (Control)	3	F	Soil from from Soroksar experimental research farm, located at the Hungarian University of Agriculture and Life Sciences organic farming units

Temperature and humidity were measured during the experiment by Voltcraft co. DL-181THP data logger. The maximum temperature was 33.12 °C, and the minimum was 16.14 °C. Maximum relative humidity was 88.44 % as shown in Figure 12.

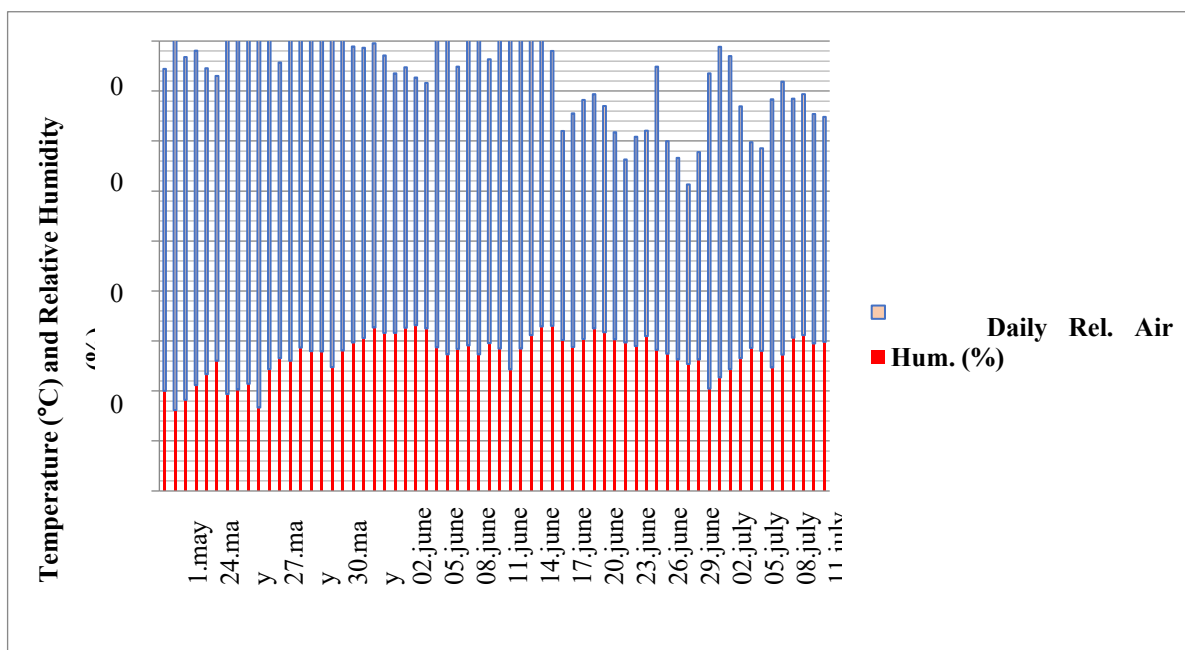


Figure 12. Temperature and relative humidity during the sweet potato pilot study

4.1.3. Roots sampling and staining

The roots were sampled on the 8th week after transplanting (end of July 2019). After that, the roots were washed by tap water to remove the substrate by immersing the roots in the water for 2- 3 seconds and shake the roots to ensure that there is no substrate in the all parts of the root system. At the end of the pilot study, several physical parameters were measured for sweet potato seedlings for both varieties manually by ruler such as, length of roots and shoots (cm), and the length of the stem (cm). In addition to the fresh weight of shoots and roots (g) of seedlings for both varieties were measured by electronic balance. The samples were stained at the laboratory of the Department of Vegetable and Mushroom Growing in the days following the sampling.

Roots staining were carried out according to the method of PHILLIPS and HAYMAN (1970). Preparation for roots staining was done separately for each root sample. Samples were placed in 10% KOH for 1 hour and 15 minutes at 65°C in the dryer. Drying in solution, make the roots lighter, more colorless, and easier to examine. After clarification, washing the roots with distilled water was needed to remove the KOH solution. Then, the roots were acidified in 10% lactic acid and left overnight. The acidified root pieces were stained with aniline blue then poured 3 times for 30 seconds in 10% lactic acid to wash off excess dye. For long term storage, stained roots were also added to lactic acid.

4.1.4. Evaluating of the arbuscular mycorrhizal colonization

Colonization of AMF was evaluated according to TROUVELOT et al. (1986) method as shown in Table 7. The root sample was placed in a 7x7 grid with a small amount of lactic acid and the root pieces were randomly distributed there. The root fragments distributed on the 49 lattice points were examined using a light microscope in 2 replicates and the degree of colonization and arbuscular abundance of the root fragments reaching a lattice point were graded. In cases where the grid points no root was dropped, we moved on and recorded no value. This was done because the target was not the examination of 49 roots per replicate, but the examination of a randomly distributed root mass based on 49 different spatial points, so the points that did not reach the root fragment were simply not examined. Between the two repetitions, the root pieces were randomly shuffled and redistributed, giving us the opportunity to examine the sample more comprehensively. When examining the root fragments, they were classified using the following point system (Table 8).

Table 8. Classification of mycorrhizal colonization and arbuscular abundance according to TROUVELOT et al. (1986).

Development of the point system determining the level of colonization	0	No colonization
	1	The colonization rate is less than 1%.
	2	Colonization rate less than 10%
	3	The colonization rate is less than 50%.
	4	The colonization rate is greater than 50% but less than 90%.
	5	The colonization rate is greater than 90%.
Classification of arbuscular abundance	A0	There is no arbuscule in the root.
	A1	Only a few arbuscules are visible.
	A2	Arbuscules are common.
	A3	Extremely high arbuscular abundance.

Based on the equation proposed by TROUVELOT et al. (1986), all mycorrhizal parameters of colonization were calculated and expressed as a percentage by using MycoCalc software as shown in Figure13 (developed by Biro Zsombor: frequency calculations were performed using a Windows forms application written in C # and developed to facilitate the process, based on the equations of TROUVELOT et al. (1986)) (ALHADIDI et al. 2021). (F %: Frequency of mycorrhiza in the root system((nb of fragments myco/total nb)*100), M %: Intensity of the mycorrhizal colonization in the root system((95n5+70n4+30n3+5n2+n1)/(nb total) where n5 = number of fragments rated 5; n4 = number of fragments 4 , m %: Intensity of the mycorrhizal colonization in the root fragments(M*(nb total)/(nb myco), a %: Arbuscular abundance in mycorrhizal parts of roots fragments(100mA3+50mA2+10mA1)/100) where mA3, mA2, mA1 are the % of m,rated A3,A2,A1,respectively,with mA3=((95 n5A3+70n4A3+30n3A3+5n2A3+n1A3)/nb myco)*100/m and the same for A2 and A1, A %: Arbuscular abundance in the root system (A% = a*(M/100). Slides were prepared to check the hyphal and the arbuscular development by Zeiss AxioCAM Hr3 microscope camera (Jena, Germany).

MycoCalc2.0

	0	1	2	3	4	5
A0						
A1						
A2						
A3						

Calc

F% = -
M% = -
m% = -
a% = -
A% = -

Reset

More Less

Frequency of mycorrhiza in the root system
 $F\% = (\text{nb of fragments myco} / \text{total nb}) * 100$

Intensity of the mycorrhizal colonisation in the root system
 $M\% = (95n5 + 70n4 + 30n3 + 5n2 + n1) / (\text{nb total})$
 (where n5 = number of fragments rated 5; n4 = number of fragments 4 etc.)

Intensity of the mycorrhizal colonisation in the root fragments
 $m\% = M * (\text{nb total}) / (\text{nb myco})$

Arbuscule abundance in mycorrhizal parts of root fragments
 $a\% = (100mA3 + 50mA2 + 10mA1) / 100$
 (where mA3, mA2, mA1 are the % of m, rated A3, A2, A1, respectively, with
 $mA3 = ((95n5A3 + 70n4A3 + 30n3A3 + 5n2A3 + n1A3) / \text{nb myco}) * 100 / m$
 and the same for A2 and A1.)

Arbuscule abundance in the root system
 $A\% = a * (M / 100)$

Total Number = -
 Nb of Fragments Myco = -
 n1 = -
 n2 = -
 n3 = -
 n4 = -
 n5 = -
 a1 = -
 a2 = -
 a3 = -
 ma1 = -
 ma2 = -
 ma3 = -
 a = -

Developed by: B. Zs
According to Trouvelot et al (1986)

Figure 13. Calculation of the mycorrhizal parameters by MycoCalc software (developed by Biro Zsombor according to TROUVELOT et al. (1986)).

4.2. Organic potato experiment

4.2.1. Time and place of the experiment

This experiment was designed by Agroscope (the Swiss Confederation's center of excellence for agricultural research) and applied by ÖMKi (Research Institute of Organic Agriculture) as part of the SolACE project (Solutions for improving Agroecosystem and Crop Efficiency for water and nutrient use) at the Soroksár experimental research farm, located at the Hungarian University of Agriculture and Life Sciences organic farming units between April till August of 2020 and 2021 as shown in Figure 14. Organic farming methods have been practiced at the trial site for more than a decade. The site was previously planted with rye. The experiment area was open field with 1316.25 m² area divided into 64 experimental plots. A randomized complete block design was selected in the two years experiment. The size of the experimental plot was 864 m² plot with inter spacing 22.5 m². A total of 32 experimental plots were created, in which the irrigated area was 432 m².



Figure 14. Potato cultivation in Soroksár experimental research farm, located at the Hungarian University of Agriculture and Life Sciences organic farming units (ALHADIDI 2021).

4.2.2. Soil characteristics of the experimental field

The soil type on the experimental site at Soroksar experimental research farm, located at the Hungarian University of Agriculture and Life Sciences organic farming units is sandy soil with pH (H₂O) 8.5, CaCO₃ 9 % and humus 2.3 %. Soil analysis study conducted in 2020 and 2021 gave the tabulated results as shown in Table 9. The contents of nitrate, nitrite, and ammonium (mg/kg) (dry matter) in the soil samples were measured according to ISO 14256-2:2005E International standard, the nitrate is measured by segmented flow analysis (SFA) system, the nitrate in the extracts is reduced to nitrite by passing copperized cadmium powder (4.30). Nitrite originally present in the soil and those produced by reduction form a diazo compound in acid medium after the addition of sulfanilamide and N-(1-naphthyl) ethylenediamine dihydrochloride (4.14) (Griess-Ilosvy reagent). Its absorbance is measured at a wavelength of 543nm. The determination of ammonium ions is carried out using an SFA system and is based on the Berthelot reaction, in which a phenol derivative (Salicylate, 4.8) forms an indophenol in the presence of ammonium and sodium dichloroisocyanurate (4.16) under catalytic action of sodium nitroprusside (4.15). Its absorbance is measured at a wavelength of 660nm. The applied fertilizers in the experiment in Kg/ha were nitrogen fertilizer 120 (Viano Blood Meal 13%, 372 potassium (Patentkali) and there was no phosphorus fertilizer due to the high amount of phosphorus in the soil.

Table 9. Soil characteristics for experimental area in soil depth (0-30 cm).

Parameters	2020		2021	
	Irrigated	Control	Irrigated	Control
NO ₃ -N mg/kg	No data	No data	10.7	8.7
NH ₄ -N mg/kg	6.8	7.3	6.6	7.2
EDTA-P ₂ O ₅ mg/kg	528	476	524	475

4.2.3. Meteorological data during the experiment period

Precipitation means per month (mm), temperature (°C), relative humidity (%), soil temperature (°C) and Leaf Wetness (%) of the experiment area during two years of growing season were recorded in Table 10 and 11. The total precipitation was 443.4 mm and 310 mm in 2020 and 2021, respectively. The meteorological station was set up by the University of Debrecen, using

plant production information system called Metagro (ÖMKi - Research with the participation of Hungarian farmers (www.biokutatas.hu) at the trial site to monitor rainfall and soil moisture, and to provide a precise calculation of necessary water quantities for optimal potato irrigation. Drip irrigation system was used in the experimental site. The amount of irrigation water was measured based on the irrigation system and its capability. In 2020, the amount of applied water is 15mm/hr while in 2021, 7mm of water /hour was applied. Therefore, the amount of irrigation water depended on the actual amount of precipitation.

Table 10. Means of monthly temperature (°C), relative humidity (%), soil temperature (°C), precipitation (mm), and Leaf Wetness (%) in the experimental site in 2020 (Soroksár, Hungary) (Metagro system).

Month	T (°C)	RH (%)	Soil T (°C)	Precipitation (mm)	Leaf Wetness (%)
April	11.09	54.35	14.13	13.8	21.83
May	14.94	62.98	18.58	16.2	31.26
June	20.58	74.14	23.26	77.6	40.24
July	21.92	69.93	24.56	58.0	45.81
August	23.12	68.28	24.75	42.8	42.93
September	17.78	72.46	20.082	30.4	48.57

Table 11. Means of monthly precipitation (mm), temperature (°C), relative humidity (%) and soil temperature (°C) and Leaf Wetness (%) in the experimental site in 2021 (Soroksár, Hungary) (Metagro system).

Month	T (°C)	RH (%)	Soil T (C)	Precipitation (mm)	Leaf Wetness (%)
April	8.93	67.16	10.04	26.6	30.01
May	14.12	71.25	15.07	52.2	36.53
June	22.9	61.42	26.11	10.2	28.90
July	24.69	62.96	26.47	43.4	34.11
August	20.57	72.2	22.34	56.4	48.57
September	17.19	70.31	18.56	19.4	38.71

4.2.4. Plant materials and cultivation of the tubers

One variety of potato ‘Desiree’ was used in the two years experimental field (Figure 15). The tubers were supplied from Primag Kft (Hungary-9022 Győr, Czuczor Gergely u 18-24), its originated from Székelyföld. The Desire variety has a pink to red skin with an elongated to oval shape. It is a resistant variety for drought and viruses as well as powdery scab, in addition to good yield under water shortage. The foliage for Desiree variety is Medium-sized, medium-dark grey-green leaves. Its leaves are stiff, slightly curved. Its dry matter content is moderately high (21.4

%), a versatile, rather waxy variety, it has a long dormancy period and can be stored well (<https://www. www.primag.hu>, last accessed on 2024.02.18). The potato field was grown according to the EU regulation (EC No. 2018/848), which corresponds to the usual organic practices on the farm. The tubers were planted in 20th of April in 2020 and in 26th of April in 2021 at a soil depth of 10 cm, with plant density (3, 70 cm (between rows) * 30 cm (interrow)). After emergence, 20-30 cm high ridges were laid along the rows. Weed control was done manually and regular crop protection treatments against *Phytophthora infestans* and *Leptinotarsa decemlineata* with *Bacillus thuringiensis* (Novodor FC) which is manufactured by Valent Biosciences (Libertyville, Illinois, United States), copper (Cuproxat) which is manufactured by Nufarm (Melbourne, Australia) , and Spinosad (Laser Duplo) manufactured by Laser Duplo (United States) were applied. The tubers harvest took place at the 2nd of September.



Figure 15 Potato Desiree cultivar (ALHADIDI 2021)

4.2.5. Applied treatments of microbial inoculates

Seven different treatments were used with arbuscular mycorrhizal fungi (AMF), plant growth promoting rhizobacteria (PGPR) and *Trichoderma*. Each treatment had four replicates with a total of 64 plots. Each plot was planted with 12 potato tubers. The plots were separated and surrounded by at least two rows of buffers in each direction. The selected strains of the microbial inoculates were kindly supplied from other partners of SOAICE project, they already gone under selection in previous pots experiments in greenhouse and they performed the best results. The isolates were selected for testing in the open field in previous laboratory experiments conducted within the framework of the SolACE project.

Several strains of microbial inoculants were prepared and mixtures of them were used as shown in Table 12. The treatments were conducted under both irrigated condition and another plots without irrigation. Three mixtures of inoculants were tested on potato compared to the untreated (control). The inoculants were prepared by Minigran technology (<https://dcm.green/en/minigran-technology>, last accessed on 2024.02.17). Minigran technology is a unique granulation process used to create fertilizers. It was developed by the Belgian company DCM and is used in their organic and organo-mineral fertilizers. The process involves combining a variety of organic and mineral nutrients into small, uniform granules. These granules are then coated with a thin layer of a slow-release polymer. This coating helps to control the release of nutrients into the soil, making them more available to plants over a longer period of time. The inoculates were sensitive to heat and UV light. As soon as the inoculants were applied to the tubers in the planting date in 20th of April 2020 and in 26th of April 2021 in the opened furrows as shown in Figure 16 these were manually covered as quickly as possible. The tubers were not treated with chemicals (bactericide, fungicide). Treatments were applied once in both years at planting time for each growing season.

Table 12. Treatments and types of microorganisms of inoculum mixtures used in the potato field trial.

Microbial inoculates strains	Application rate/ Biological material need in (g)	(CFU/tuber (for AMF: g/tuber)	Concentration of microbial product (CFU/g)	Quantity of granule per tuber (g)
<i>Pseudomonas brassicacearum</i> 3Re2-7	7.20	2.00E+08	1.60E+10	0.75
<i>Paraburkholderia phytofirmans</i> PsJN	6.40	1.00E+08	9.00E+09	0.75
<i>Trichoderma asperelloides</i> A	0.86	1.50E+06	1.00E+09	0.75
<i>Rhizophagus irregularis</i> MUCL41833	0.3456	6.00E-04	n.d	0.75
<i>Rhizophagus irregularis</i> MUCL41833+ <i>Pseudomonas brassicacearum</i> 3Re2-7	0.3456 7.20	6.00E-04 2.00E+08	n.d 1,60E+10	0.75
<i>Rhizophagus irregularis</i> MUCL41833+ <i>Paraburkholderia phytofirmans</i> PsJN	0.3456 6.40	6.00E-04 1.00E+08	n.d 9.00E+09	0.75
<i>Rhizophagus irregularis</i> MUCL41833+ <i>Paraburkholderia phytofirmans</i> PsJN+ <i>Trichoderma asperelloides</i> A	0.3456 6.40 0.86	6.00E-04 1.00E+08 1.50E+06	n.d 9.00E+09 1.00E+09	0.75
Control treatment	n.d	n.d	n.d	n.d



Figure 16. Applying microbial inoculates to the potato tubers in the planting date in 20th of April 2020 and in 26th of April 2021 at Soroksár experimental research farm, located at the Hungarian University of Agriculture and Life Sciences organic farming unit.

4.3. Field measurements of the harvested potato tubers

4.3.1. Yield of harvested potato tubers

For each plot and for each treatment, the number of plants was counted just before harvest. Yield was measured per row once the potato tubers were harvested five months after transplanting. They were bagged and measured immediately as shown in Figure 17.



Figure 17. Harvested tubers bags from the experimental field (ALHADIDI 2020).

4.3.2. Starch measurement of harvested potato tubers

The measurement of the starch was made according to EU-direction (International Starch Institute: Determination of Starch in Potatoes). One measurement was made for the starch content in 2020 and 2021. The results are represented in a graph and a table will be determining the amount of starch content (International Starch Institute: Determination of Starch in Potatoes, starch content of potato versus density according to EU-direction: 1999 release,

<http://www.starch.dk/isi/methods/starchct.htm>- last accessed on 20th of May, 2022

4.3.3. Phenological growth stages of potato plants

The rows were scaled according to the BBCH scale in 17th of June 2020 and in 24th of June 2021. The BBCH scale is a scale that allows the phenological stage of plants to be observed by developmental stage (MEIER 2018).

BBCH scale code Stage (00-09)Sprouting (tuber germination) (10-19)Emergence of the shoots (20-23)Tuberization (30-39)Bulking (40-49) Tuber maturation

4.4. Lab measurements

4.4.1. Total phosphorus content in the potato tubers

The Total phosphorus content of the potato tubers measured at the soil lab in Godollo campus located at the Hungarian University of Agriculture and Life Sciences, Godollo. Potato samples were cut into small pieces, placed in 1-1 Petri dishes and dried in a drying oven (LP-321 (200 L) (Labor- Mix Laborszerviz, Hungary) . After drying, the samples were ground, homogenized and filled into small paper packets. The total phosphorus content was determined according to the MSZ 21470- 50:2006 standard (Environmental testing of soils- Determination of soil toxic element, heavy metal and chromium(VI) content, Hungarian standards institution, Budapest Hungary) (HEGEDUS et al. 2016)after digestion in a microwave-assisted ($\text{HNO}_3 + \text{H}_2\text{O}_2$) mixture, for which a CEM MARS 5(United states and Magne-Chem Kft (1195 Budapest Jókai utca 22. fsz. 2), the domestic distributor of CEM products in Hungary) closed-chamber microwave device equipped with temperature and pressure sensors used. Samples weighing 0.4000 (± 0.0500) g were placed in teflon tubes and add the mixture of 5 mL 65(m/m) % HNO_3 (Honeywell, Seelze) and 2 mL 30(m/m) % H_2O_2 (Thomasker, Budapest). Finally, the walls of the

tubes were washed with a small amount of Milli Q water. The caps were put on the teflon tubes and locked with the key. The tubes put into the rotor digester on the standard programme. The digested samples were filtered into 25 ml volumetric flasks and filled with Milli Q water as shown in Figure 18. The filtrates were analyzed on UV-Vis spectrophotometer (Spekol 221, Carl Zeiss, Jena) for total phosphorus content as shown in Figure 19.

The spectrophotometrical filtrate analysis showed as the following; pipette 5-5 ml of the digested samples into 50 ml volumetric flasks and add 10-10 ml of vanadate-molybdenate solution. Fill up to 50 ml with distilled water and shake. The solution should be yellow color. Wait 30 minutes before measuring. The color stays stable for 4-5 hours. Measure at 410 nm for light solutions and at 430 nm for dark solutions. Before the measurement, the system was washed with distilled water. The absorbance of the standard solutions was measured first, then that of the samples. The phosphorus content of the samples was read from the standard curve (calibration curve).



Figure 18. Prepared samples with Milli Q water after filtration (ALHADIDI 2020).



Figure 19. Samples measuring by spectrophotometer (ALHADIDI 2020).

4.4.2. Mycorrhizal colonization of the roots

Roots sampling and staining

Two potato plant roots were sampled from each treatment per replicate in the two years of the experiment after four months of transplanting. Ink based staining was carried out following the method suggested by PHILLIPS and HAYMAN (1970). The method used to evaluate roots colonization is the method of TROUVELOT et al. (1986). Thirty root fragments approximately 1cm in length are randomly selected and placed in two slides (each slide has 15 root fragments) for a first replicate observation under the light microscope. After observation, the scores of the hyphae and arbuscules written on the lab sheets is selected individually. The scoring is transferred individually to MycoCalc software and the software automatically calculates the percentage of mycorrhizal parameters (F %: Frequency of mycorrhiza in the root system, M %: Intensity of the mycorrhizal colonization in the root system, m %: Intensity of the mycorrhizal colonization in the root fragments, a %: Arbuscular abundance in mycorrhizal parts in root fragments, A %: Arbuscular abundance in the root system). .

4.5. Statistical analysis

4.5.1. Sweet potato pilot study data

Data analysis was performed with IBM SPSS 25 software version 25.0. Armonk, NY: IBM Corp (2017). Shoot and root fresh weight (g) and root and stem lengths were analyzed using the

two-way MANOVA model with the factor's diversity (orange and purple) and treatment levels [(L+SYM). S], F, (L +SYM). The normality of the residuals was tested using the ShapiroWilk method ($K(74) > 0.95$; $p > 0.05$). After having an overall significant MANOVA result, we performed subsequent univariate ANOVA tests with Bonferroni's correction. In some cases, the homogeneity of variances was slightly violated ($Levenes\ 0.05 > p > 0.02$), so pairwise comparisons of treatments were performed using the Games-Howell post hoc test.

4.5.2. Organic potato experiment

Data analysis was performed using SAS software version 9.4 (2013). Starch, total phosphorus and yield were analyzed using the one-way three-factor Anova model; year, microbial inoculation treatment and water treatment. Prior to ANOVA, descriptive statistics were generated for all measurements to monitor the distribution of the data and normality using a general linear model ($p\text{-value} > 0.05$). Means were separated at a significance level of 0.05 using Tukey's test.

5. RESULTS

5.1. Sweet potato pilot study

5.1.1. Arbuscular mycorrhizal developmental shapes

After checking roots samples under the microscope, the samples of which was treated in sterilized peat moss with the presence of the mycorrhizal inoculum (Symbivit) has shown a symbiotic relationship with sweet potato seedlings for both varieties, orange and purple. In case of control seedlings, we could observe hyphal and arbuscular development in as well by the light microscope (Figure 20-21).

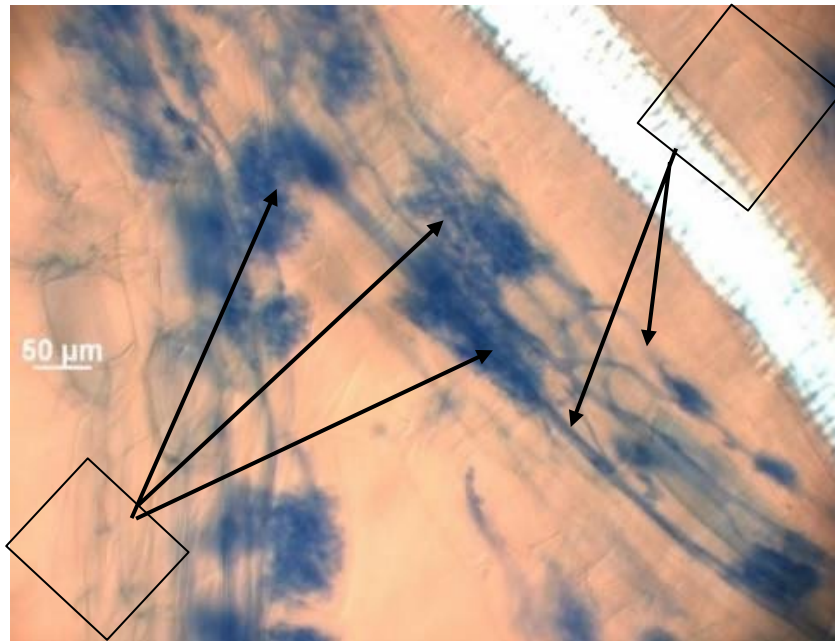


Figure 20. Root of purple sweet potato seedlings treated with mycorrhizal inoculum in sterilized peat moss showing **(H)** hyphal and **(A)** arbuscular development.

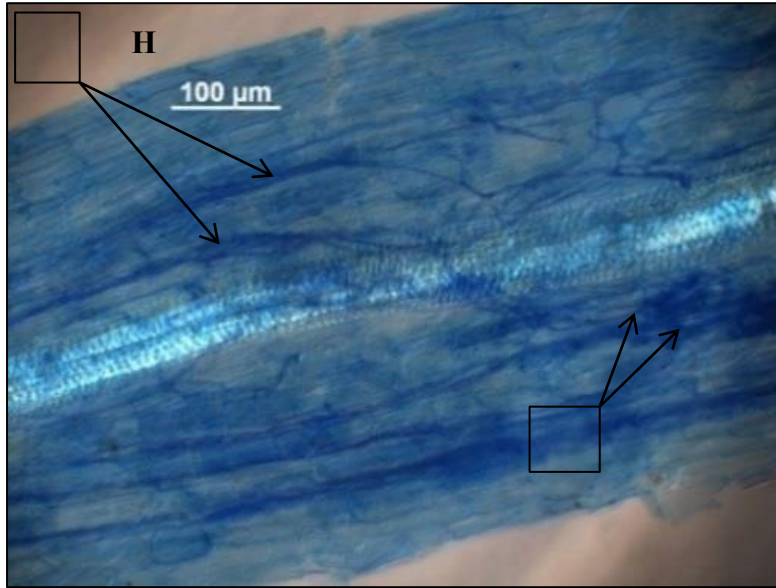


Figure21. Dense arbuscular formation on root in control sweet potato seedlings.

Both varieties were also able to develop arbuscular mycorrhiza when treated with soil from Soroksár experimental research farm, located at the Hungarian University of Agriculture and Life Sciences organic farming units. **(H)** Hyphal development and **(A)** arbuscular formation can also be observed in the orange sweet potato seedling

5.1.2. Mycorrhizal parameters

The high intensity of mycorrhizal colonization in the root system (M %) was measured of the purple variety in (L+SYM) as shown in Table 13. The root fragments (m %), the arbuscular frequency in the mycorrhizal parts (a %) showed that in the purple variety a significantly higher percentage was recorded with (L+SYM) than with [(L+SYM). S] based on the calculations by Mycocalc software according to Trouvelot et al. 1986 method. However, no arbuscules were found in the (F) control treatment, although in the orange variety, arbuscules were found only in the [(L+SYM).

S] treatment, while no arbuscules were found in the (L+SYM) and (F) treatments. Regarding the arbuscular frequency in the root system (A %), in the purple variety, the highest arbuscular frequency was recorded at (L+SYM), followed by [(L+SYM).S], while no arbuscules were found with the (F) control treatment. In the orange variety, arbuscules were only found in the [(L+SYM).S] treatment, while no arbuscules were detected in (L+SYM) and the control treatment.

Table 13. Mycorrhizal parameters (F%, A%, a%, M% and m %) within two varieties of sweet potato roots samples with different treatments.

Variety	Treatment	F %	Std.Dev	M %	Std.Dev	m %	Std.Dev	a %	Std.Dev	A %	Std.Dev
Purple	L+SYM	20.22	3.33	11.49	1.18	58.07	15.45	86.82	0.20	9.97	1.04
	[(L+SYM).S]	84.86	1.20	42.73	9.41	50.29	10.37	5.38	2.11	2.40	1.40
	F	4.52	1.68	1.25	1.52	36.50	47.37	no arbuscule	No data	no arbuscule	No data
Orange	L+SYM	21.63	6.44	0.32	0.06	1.47	0.14	no arbuscule	No data	no arbuscule	No data
	[(L+SYM).S]	67.19	7.11	21.44	0.57	32.13	4.24	35.28	7.86	7.59	1.88
	F	15.89	1.77	2.82	2.24	18.67	16.26	no arbuscule	No data	no arbuscule	No data

F %: Frequency of mycorrhiza in the root system, M %: Intensity of the mycorrhizal colonization in the root system, m %: Intensity of the mycorrhizal colonization in the root fragments, a %: Arbuscular abundance in mycorrhizal parts in root fragments, A %: Arbuscular abundance in the root system

5.1.3. Physical parameters of sweet potato seedlings

Two-way MANOVA resulted in significant diversity and treatment effects (Wilks lambda = 0.48, $p < 0.001$; Wilks lambda = 0.18, $p < 0.001$) with significant interaction (Wilks lambda = 0.64, $p < 0.05$). Subsequent univariate ANOVA was for both shoot fresh weight and stem length for both cultivar and treatment ($F(1,38) > 6.48$; $p < 0.05$) and in the case of total root fresh weight for the treatment effect ($F(4,38) = 7.90$; $p < 0.01$) as well as for their interaction in the case of shoot fresh weight and stem length ($F(4,38) > 4$) significant. 88; $p < 0.01$). The variety influence was not significant in the case of fresh weight of total roots and length of roots ($F(4,38) = 0.08$) and the treatment effect was not significant in the case of length of roots ($F(4,38) = 3.13$; $p = 0.06$). Means and standard deviations of the four physical parameters and post-hoc test results are summarized in Table 14.

The highest mean of shoot fresh weight for orange sweet potato seedlings was found with the (L+SYM) treatment (11.43 g). There was no significant difference between treatment [(L+SYM). S] and (F), therefore we concluded that mycorrhizal inoculum could increase shoot weight over time, while sterilization had no increased effect on shoot weight (g) in treated orange sweet potato seedlings. The highest fresh shoot weight in purple sweet potato seedlings was under treatment (L+SYM) with a mean of (15.81 g) while [(L+SYM).S] treatment gave the lowest mean (3.80 g), the same conclusion previously, the mycorrhizal inoculation also increased shoot fresh weight in the purple sweet potato seedlings, whereas sterilization had no effect on shoot fresh weight. The highest root weight was found in orange sweet potato seedlings, with the highest mean at [(L+SYM). S] treatment (4.89 g), while the lowest mean was found in the control treatment (F) with a mean of (1.09 g). This means that mycorrhizal inoculation and sterilization had a great impact on the root weight of orange sweet potato seedlings. For the root weight of purple sweet potato seedlings, the highest mean was with the (L+SYM) treatment (5.89 g), while the lowest mean was with the (F) treatment with a mean (3.44 g). This is because mycorrhizal inoculation had a stimulatory effect on fresh root weight of purple sweet potato seedlings. PATI et al. (2024) described the phenological growth stages of sweet potato for the first time using the extended BBCH scale. From the pervious results, it can be refer to the extended BBCH scale stage 4 which describe the development of the tubers. According to the BBCH scale, code (400) Tuber initiation: swelling of first stolon tips to twice the diameter of

subtending stolen. At (BBCH 405), half of the last tuber mass comes during the inflorescence, and 95% of the last tuber development is completed after the plant reaches maturity (BBCH 408). The highest root length of the orange sweet potato variety was measured in seedlings treated with Symbivit in sterilized substrate (peat moss) [(L+SYM).S] treatment gave a mean of (35.52 cm). However, there was no significant difference between the mean values when comparing the three stages of treated orange sweet potato seedlings. Therefore, mycorrhizal inoculation with sterilization had no effect on root length of orange sweet potato seedlings. The highest mean root length (36.13 cm) was measured in the purple sweet potato seedlings propagated in a sterilized Latagro peat moss with Symbivit [(L+SYM).S] treatment. However, the differences between the treated seedlings were not significant. The highest mean stem length in orange sweet potato seedlings was measured with the (L+SYM) treatment (75.32 cm), while the lowest mean occurred with the (F) treatment (20.00 cm). Accordingly, the mycorrhizal inoculation could increase the length of the stems in the orange sweet potato seedlings.

Table 14. Sweet potato seedlings means and standard deviations of the four physical parameters fresh weight of shoots (g), fresh weight of total roots (g), length of roots (cm) and length of stem (cm) together with the post hoc tests results (Games–Howell’s, $p < 0.05$). The different letters are for significantly different groups (lower case: comparison of treatments for fixed varieties-read vertically), upper case: comparison of varieties for fixed treatments-read horizontally)

Variety		Orange				Purple			
parameters	treatment	Mean	Std. Dev	comparison of treatments ¹	comparison of varieties ²	Mean	Std. Dev	comparison of treatments	comparison of varieties
FW of shoots (g)	[(L+SYM).S]	4.76	0.88	A	A	3.80	2.99	a ¹	A2
	F	2.83	1.73	A	A	9.42	3.04	b ¹	B2
	L+SYM	11.43	3.85	B	A	15.81	4.32	c ¹	A2
FW total roots (g)	[(L+SYM).S]	4.89	1.14	B	A	3.88	2.38	a ¹	A2
	F	1.09	0.89	A	A	3.44	1.87	a ¹	B2
	L+SYM	4.79	1.87	B	A	5.89	2.97	a ¹	A2
Length of roots (cm)	[(L+SYM).S]	35.52	6.26	A	A	36.13	13.58	a ¹	A2
	F	23.17	8.69	A	A	32.74	6.54	a ¹	B2
	L+SYM	25.90	7.44	A	A	30.83	8.30	a ¹	A2
Length of stem (cm)	[(L+SYM).S]	35.56	12.89	A	B	14.69	5.80	a ¹	A2
	F	20.00	11.21	A	A	29.37	7.83	b ¹	A2
	L+SYM	75.32	36.27	B	A	47.69	14.72	c ¹	A2

¹ comparison of treatments for fixed varieties – read vertically; ² comparison of varieties for fixed treatments – read horizontally.

On the other hand, sterilization had no increasing effect on stem length in orange sweet potato seedlings. In the purple sweet potato seedlings, the highest stem length was measured in L+SYM (47.69 cm), while the lowest mean was in treatment [(L+SYM).S] (14.69cm). Mycorrhizal inoculation had a positive effect on root length. However, sterilization had no stimulating effect on root length of purple sweet potato seedling

5.2. Field organic potato experiment 2020 and 2021

5.2.1. Mycorrhizal parameters

In the first season (2020), the highest mycorrhizal colonization frequency (F %) and mycorrhizal colonization intensity (M %) were recorded on non-irrigated treated areas with a combination of *Rhizophagus Irregularis*MucL41833+ *Paraburkholderia phytofirmans*PSJN, namely 96.6 7% and 28.56 %. In the irrigation treatments, it was higher than F % and M%, but there were no significant differences based on the calculations by Mycocalc software according to Trouvelot et al. 1986 method. With irrigation, the F% percentage was higher in *Rhizophagus Irregularis*MucL41833, *Pseudomonas Brassicacearum*3Re2-7, *Paraburkholderia phytofirmans*PSJN, *Trichoderma asperelloides*A and *Rhizophagus Irregularis*MucL41833+ *Paraburkholderia phytofirmans*PSJN + *Trichoderma asperelloides*A combination than treatments without irrigation. This is confirmed by the second year results where all treatments under irrigated conditions gave a lower F % percentage than the control (no irrigation), achieving a mycorrhizal colonization frequency of 100 %. For the arbuscular abundance (A %), Most treatments showed no arbuscular frequency in both years, with the exception of the mixture of *Rhizophagus irregularis*MucL41833 + *Pseudomonas brassicacearum*3Re2-7 (41 %), *Rhizophagus irregularis*MucL41833 + *Paraburkholderia phytofirmans*PSJN (24 %) and *Trichoderma asperelloides*A (33 %). All these parameters during the two years are presented in Table15.

Table 15. Mycorrhizal parameters (F %, M %, A %,) in irrigated and non-irrigated potato treatments within 2020 and 2021 experiments.

Treatment	Mycorrhizal colonization Freuency				Mycorrhizal colonization Intensity				Arbuscular abundance			
	F %				M %				A %			
	2020		2021		2020		2021		2020		2021	
	I	C	I	C	I	C	I	C	I	C	I	C
Control	26	80	93	100	1.3	24	2.3	16	no arbuscular	no arbuscular	no arbuscular	no arbuscular
Rhizophagus irregularis MucL41833	56	32	100	100	3	1.6	7.2	26	no arbuscular	no arbuscular	no arbuscular	no arbuscular
Pseudomonas brassicacearum 3Re2-7	89	57	97	100	13	5.3	6.2	22	no arbuscular	no arbuscular	no arbuscular	no arbuscular
Rhizophagus irregularis MucL41833+Pseudomonas brassicacearum3Re2-7	90	90	100	100	15	11	20	17	41	41	41	41
Paraburkholderia phytofirmansPSJN	90	37	94	100	16	3.1	3.1	22	no arbuscular	no arbuscular	no arbuscular	no arbuscular
Rhizophagus irregularis MucL41833+Paraburkholderia phytofirmansPSJN	96	97	98	100	18	29	5.7	17	24	24	24	24
Trichoderma asperelloidesA	100	49	100	100	26	1	4.1	16	33	33	33	33
Rhizophagus irregularis MucL41833+Paraburkholderia phytofirmansPSJN+Trichoderma asperelloidesA	78	40	100	100	8.2	2.2	18	18	no arbuscular	no arbuscular	no arbuscular	no arbuscular

5.2.2. Potato tubers yield

As an overall result of the effect of various treatments on potato yield, as shown in Table 16, yield was not significantly affected by any of the treatments in the two test seasons. It was found that the yield of irrigated treatments is generally higher than that of non-irrigated treatments in both seasons. In terms of inoculation effect, *Paraburkholderia phytofirmans*PSJN yielded the highest yield (15.21kg/m²) under irrigation in the first season, but *Rhizophagus irregularis* MucL41833 was highest in the second season (16.72kg/ m²). Among non-irrigated treatments, control treatment was the highest (12.03 kg/ m²), followed by *Paraburkholderia phytofirmans* PSJN treatment (12.02 kg/ m²) in 2020, and in the second season, inoculation with *Rhizophagus irregularis* MucL41833 gave the highest yield (16.72kg/ m²) in irrigated treatments followed by the *Rhizophagus irregularis*MucL41833+ *Paraburkholderia phytofirmans*PSJN+ *Trichoderma asperelloides*A (16.66 kg/ m²). In non-irrigated treatments, *Rhizophagus irregularis*MucL41833 presented the highest yield (11.11 kg/ m²) while there is no significance difference among other treatments.

Table 16. Means of potato tubers yield (kg/m²) by microbial inoculants in irrigated and non-irrigated conditions within two years experiment.

Yield of potato 2020			Yield of potato 2021	
I		C	I	C
<i>Pseudomonas brassicacearum</i> 3Re2-7	12.81 ±0.822	11.81 ±0.629	16.24 b±1.857	10.49 ±0.489
<i>Paraburkholderia phytofirmans</i> PSJN	15.21 ±0.708	12.02 ±0.503	14.48 ±1.729	10.11 ±0.624
<i>Trichoderma asperelloides</i> A	14.05 ±1.050	11.58 ±1.251	15.18 ±1.170	10.31 ±0.839
<i>Rhizoglyphus irregularis</i> MucL41833	13.37 ±0.724	10.81b±0.563	16.72 ±0.861	11.11 ±1.035
<i>Rhizoglyphus irregularis</i> MucL41833+ <i>Pseudomonas brassicacearum</i> 3Re2-7	14.00 ±0.478	11.66 ±0.560	14.20 c±1.075	9.83 ±0.968
<i>Rhizoglyphus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN	13.25 ±1.078	11.54 ±0.541	15.07 ±1.183	9.83 ±0.997
<i>Rhizoglyphus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN+ <i>Trichoderma asperelloides</i> A	12.61 ±1.202	10.78 ±0.668	16.66 ±1.359	10.43 ±1.057
C (control)	14.45 ±0.959	12.03 ±0.552	15.09 ±1.488	10.97 ±0.849

Correlation and regression between the yield of potato tubers (kg/m²) and the water supply (mm) for the two years, 2020d 2021 were checked by the analysis of the data. According to the results, the R (values of the response variable made by the model) = 0.0476 and the R² (the proportion of the variance in the response variable that can be explained by predictor variables in the regression model) = 0.002. Both values are < 0.05, so there is no correlation between the yield of potato tubers (kg/m²) and the water supply (mm).

5.2.3. Total phosphorus in potato tubers

Table 17. Means of phosphorus content (mgP kg⁻¹) in potato tubers by microbial inoculants in irrigated and non-irrigated conditions within two years experiment.

	Total phosphorus in the tubers 2020		Total phosphorus in the tubers 2021	
	I	C	I	C
<i>Pseudomonas brassicacearum</i> 3Re2-7	0.32 ±0.012	0.32 ±0.010	0.62 ±0.044	0.56 ±0.053
<i>Paraburkholderiaphyt ofirmans</i> PSJN	0.32 ±0.012	0.34 ±0.008	0.68 ±0.036	0.58 ±0.022
<i>Trichoderma asperelloides</i> A	0.33 ±0.010	0.32 ±0.006	0.64 ±0.037	0.64 ±0.045
<i>Rhizophagus irregularis</i> MucL41833	0.31 ±0.014	0.32 ±0.013	0.54 ±0.051	0.57 ±0.034
<i>Rhizophagusirregular is</i> MucL41833+ <i>Pseudomonas brassicacearum</i> PSJN	0.31 ±0.007	0.30 ±0.016	0.63 ±0.029	0.54 ±0.018
<i>Rhizophagusirregular is</i> MucL41833+ <i>Paraburkholderiaphytofirm ans</i> PSJN	0.31 ±0.012	0.32 ±0.005	0.68 ±0.053	0.66 ±0.038
<i>Rhizophagusirregular is</i> MucL41833+ <i>Paraburkholderiaphytofirm ans</i> PSJN+ <i>Trichoderma asperelloides</i> A	0.32 ±0.014	0.32 ±0.011	0.63 ±0.014	0.54 ±0.025
C (control)	0.32 ±0.018	0.35 ±0.010	0.69 ±0.031	0.50 ±0.030

The total phosphorus content in the tubers is shown in Table 17. The results show non-significant differences in both years under both irrigated and non-irrigated conditions. The highest value was recorded for the *Trichoderma asperelloides*A treatment under irrigation conditions and

for the control treatment without irrigation treatment. In the second year (2021) there was an apparent increase in phosphorus levels in almost all irrigated treatments, however there was no significance difference within microbial inoculates in the irrigated and non-irrigated treatments separately. The highest was seen in the control treatment with irrigated treatment and in the *Rhizophagus irregularis*MucL41833+ *Paraburkholderia phytofirmans*PSJN treatment without irrigation.

5.2.4. Starch content by microbial inoculants treatments

Starch content was similar between different treatments in both years, with no significant differences. In 2020, the highest mean value was found in *Paraburkholderia phytofirmans*PSJN treatment with a mean of (17.16 %) and (16.37 %) under irrigated and non-irrigated conditions, respectively, followed by *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans* PSJN treatment with a mean of (16.73 %) under irrigated conditions. The lowest starch content was found in the control treatment with irrigation, while the lowest starch content in the no-irrigation treatment was found in the *Rhizophagus Irregularis*MucL41833+ *Paraburkholderia phytofirmans* PSJN treatment (14.90%). In the second season, the highest starch content in *RhizophagusIrregularis*MucL41833 treated tubers, there were no significant differences in both irrigated and non-irrigated treatments. The highest strength value in irrigation treatment was found in *Rhizophagus Irregularis*MucL41833 treatment (12.29 %), followed by *Rhizophagus Irregularis*MucL41833+ *Paraburkholderia phytofirmans*PSJN + *Trichoderma asperelloides* A (11.48%). The lowest starch content was recorded in the *Pseudomonas brassicacearum*3Re2-7 treatment (10.11 %). With the unirrigated treatment, the highest starch content was found in *Pseudomonas brassicacearum*3Re2-7(11.61 %), followed by the *Rhizophagus irregularis* MucL41833 treatment (11.60 %). The lowest starch content in the *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans* PSJN+ *Trichoderma asperelloides*A treatment (10.37 %) was also calculated, all results are presented in Table 18.

Table 18. Means of the starch content (%) by microbial inoculants treatments in irrigated ad non-irrigated conditions within two years' experiment.

	Distribution of starch by inoculant and irrigation 2020		Distribution of starch by inoculant and irrigation 2021	
	I	C	I	C
<i>Pseudomonas brassicacearum</i> 3Re2-7	16.30± 0.542	15.73 ± 0.441	10.11 ± 0.448	11.61±0.666
<i>Paraburkholderia phytofirmans</i> PSJN	17.16 ± 0.331	16.37 ± 0.201	11.06 ± 0.611	11.55 ± 0.672
<i>Trichoderma asperelloides</i> A	15.99 ± 1.308	16.09 ± 0.490	11.32 ± 1.037	10.83 ± 0.679
<i>Rhizophagusirregular is</i> MucL41833	16.42 ± 0.570	15.13 ± 0.674	12.29 ± 0.372	11.60 ± 0.836
<i>Rhizophagusirregular is</i> <i>MucL41833+Pseudomona s</i> <i>brassicacearum</i> 3Re2-7	16.69 ± 0.826	16.33 ± 0.352	10.85 ± 0.540	11.49 ±0.494
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderiaphytofir</i> <i>mans</i> PSJN	16.73 ± 0.599	14.90 ± 1.134	11.31 ± 0.602	10.67 ± 0.757
<i>Rhizophagusirregular is</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN+ <i>Trichoderma asperelloides</i> A	15.86 ± 1.392	16.20 ± 0.804	11.48 ± 0.691	10.37 ± 0.362
C (control)	15.77 ± 0.832	16.32 ± 0.452	11.08 ± 0.712	11.04 ± 0.568

The correlation and regression between the starch content of potato tubers (%) and the water supply (mm) for the two years , 2020d 2021 were checked by the analysis of the data. According to the results, the R (values of the response variable made by the model) = 0.520 and

the R^2 (the proportion of the variance in the response variable that can be explained by predictor variables in the regression model) =0.270. Both values are > 0.05 , so there is a correlation between the yield of potato tubers (kg/m^2) and the water supply (mm). The water supply (mm) has a positive effect to increase the starch content in the harvested potato tubers.

5.2.5. Phenological growth stages of potato plant

5.2.5.1. Phenological growth stages 2020

For the non-irrigated treatments, not all microbial inoculation treatments showed results in terms of main stem leaf folding in the tested plants. The control treatment showed the highest mean value 15, followed by the mixture of microbial inoculations *Rhizophagus irregularis* MUCL41833+ *Pseudomonas Brassicacearum* 3Re2-7 (8.00), *Trichoderma asperelloides* A (5.00), *Rhizophagus irregularis* MUCL41833+*Paraburkholderia phytofirmans* PsJN and *Rhizophagus irregularis* MUCL41833+*Paraburkholderia phytofirmans* PsJN+ *Trichoderma asperelloides* A show the same mean value(3.00). For the first leaf of the 2nd order branch, the control treatment gave the highest mean value (38.00), followed by the mixture of microbial inoculations *Rhizophagus irregularis* MUCL41833 + *Paraburkholderia phytofirmans* PsJN + *Trichoderma asperelloides* A and *Trichoderma asperelloides* A treatment 5.00, as well as other treatments which shows no results. For the first individual buds of the first inflorescence (main stem), all treatments gave a mean value. The mixture of microbial inoculates yielded the highest mean value of *Rhizophagus irregularis* MUCL41833 + *Paraburkholderia phytofirmans* PsJN + *Trichoderma asperelloides* A (35.83), followed by *Rhizophagus irregularis* MUCL41833 (26.88). *Pseudomonas Brassicacearum* 3Re2-7 and *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans* PSJN gave the same mean value (19.38).

The buds of the first inflorescences extended to 55 mm, an increase occurred in almost all treatments. For *Pseudomonas Brassicacearum* 3Re2-7 and *Rhizophagus irregularis* MUCL41833+*Paraburkholderia phytofirmans* PsJN it is the same (71.88). *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans* PSJN+ *Trichoderma asperelloides* A shows a percentage of (57.50). Not all treatments gave a result for flower appearance. The mixture of microbial inoculations *Rhizophagus irregularis* MUCL41833+*Paraburkholderia phytofirmans* PsJN+*Trichoderma asperelloides* A has the highest percentage at (40.00), followed by *Rhizophagus irregularis* MucL41833 and *Rhizophagus irregularis* MucL41833+*Paraburkholderia phytofirmans* PSJN treatment at (3.00). When the flower first opened, all treatments gave the same mean value of (5.00). However, at the beginning of flowering

the mean value varies between (5.00 and 10.00), with the exception of *Trichoderma asperelloides* A, which shows no results. At the full bloom stage, only *Paraburkholderia phytofirmans* PSJN treatment gave a mean value (50.00) result, while other treatments showed no results.

In the irrigated treatments, control and *Rhizophagus irregularis* MUCL41833+*Paraburkholderia phytofirmans* PSJN and *Rhizophagus irregularis* MUCL41833+*Pseudomonas Brassicacearum* 3Re2-7 showed a (5.00) result, while other treatments showed no results. Only the control treatment showed that the first leaf of the 2nd order branch unfolded by (5.00) above the first inflorescence, while other treatments showed no results. For the first individual buds, *Trichoderma asperelloides* A gave the highest mean value of (29.7), followed by the control treatment at 22.92%. For the buds of the first visible inflorescence, which extended to 5 mm, *Pseudomonas Brassicacearum* 3Re2-7 has the mean value (78.75), followed by *Rhizophagus irregularis* MUCL4183 (76.25).

Rhizophagus irregularis MUCL41833+*Paraburkholderia phytofirmans* PSJN+*Trichoderma asperelloides* A and *Trichoderma asperelloides* A only treatments gave a result for the first visible petals of the first inflorescence of (3.00). For the first flower opening in the population, all treatments gave results varying between (3.00 and 5.00). *Paraburkholderia phytofirmans* PSJN and control treatments gave the highest flowering initiation (20.00), while other treatments gave (10.00). For the full flowering phase, *Trichoderma asperelloides* A gave the highest mean value (50.00), followed by *Paraburkholderia phytofirmans* PSJN and control treatments at (25.00), while other treatments showed no results. The end of flowering phase shows 3.00 results in *Paraburkholderia phytofirmans* PSJN treatment while other treatments show no results. At the first visible berry stage, only *Rhizophagus irregularis* MUCL41833 and *Trichoderma asperelloides* A show results varying between (3.00 and 5.00), while other treatments show no results. All results are shown in Table 19.

Table19. Phenological growth stages of potato plant during 2020 (Budapest, 2022).

Treatment	Irrigation	phenological growth stage stage 2020											
		119	SD	121	SD	501	SD	505	SD	509	SD	600	SD
C (control)	Control	15	0	38	53	18.12	0.88	70.21	5.00	0	0	5	0
<i>Paraburkholderia phytofirmans</i> PSJN		0	0	0	3	11.88	3.53	70.00	0.00	0	0	5	0
<i>Pseudomonas brassicacearum</i> 3Re2-7		0	0	0	0	19.38	2.65	71.88	2.65	0	0	5	4
<i>Rhizophagus irregularis</i> MucL41833		0	0	0	0	26.88	4.41	67.50	3.53	3	4	5	0
<i>Rhizophagusirregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN		3	4	0	0	19.38	6.18	71.88	4.41	3	4	5	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN+ <i>Trichoderma asperelloides</i> A		3	4	5	7	35.83	3.53	57.50	17.67	40	66	5	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Pseudomonas brassicacearum</i> 3Re2-7		8	11	0	0	21.25	3.53	69.38	4.41	0	0		0
<i>Trichoderma asperelloides</i> A		5	7	5	0	26.25	7.07	64.38	4.41	0	0	5	0
C (control)	Irrigated	5	7	5	1	22.92	12.37	71.25	0.00	0	4	3	14
<i>Paraburkholderia phytofirmans</i> PSJN		0	0	0	0	17.50	7.07	68.75	15.90	0	0	3	4
<i>Pseudomonas brassicacearum</i> 3Re2-7		0	0	0	0	13.13	4.41	78.75	5.30	0	0	5	0
<i>Rhizophagus irregularis</i> MucL41833		0	0	0	0	16.88	9.72	76.25	5.30	0	0	5	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN		5	0	0	0	17.50	8.83	72.50	7.07	0	0	3	4
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN+ <i>Trichoderma asperelloides</i> A		0	0	0	0	19.75	3.88	73.75	5.30	3	4	5	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Pseudomonas brassicacearum</i> 3Re2-7		3	4	0	0	18.75	3.53	73.13	6.18	0	0	5	0
<i>Trichoderma asperelloides</i> A		0	0	0	0	29.17	3.53	58.13	0.88	3	4	3	4

Table 19. contd. Phenological growth stages of potato plant during 2020 (Budapest, 2022).

Treatment	Irrigation	phenological growth stage stage 2020							
		601	SD	605	SD	639	SD	700	SD
C (control)	Control	10	0	0	0	0	0	0	0
<i>Paraburkholderia phytofirmans</i> PSJN		10	0	50	0	0	0	0	0
<i>Pseudomonas brassicacearum</i> 3Re2-7		10	0	0	0	0	0	0	0
<i>Rhizophagus irregularis</i> MucL41833		5	7	0	0	0	0	0	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN		10	0	0	0	0	0	0	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN+ <i>Trichoderma asperelloides</i> A		5	7	0	0	0	0	0	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Pseudomonas brassicacearum</i> 3Re2-7		10	0	0	0	0	0	0	0
<i>Trichoderma asperelloides</i> A		0	0	0	0	0	0	3	4
C (control)	Irrigated	20	35	0	0	0	0	0	0
<i>Paraburkholderia phytofirmans</i> PSJN		20	14	25	35	3	4	0	0
<i>Pseudomonas brassicacearum</i> 3Re2-7		10	0	25	0	0	0	0	0
<i>Rhizophagus irregularis</i> MucL41833		10	0	0	0	0	0	3	4
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN		10	0	0	0	0	0	0	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN+ <i>Trichoderma asperelloides</i> A		10	0	0	0	0	0	0	0
<i>Rhizophagus gularis</i> MucL41833+ <i>Pseudomonas brassicacearum</i> 3Re2-7		10	0	0	0	0	0	0	0
<i>Trichoderma asperelloides</i> A		10	0	50	0	0	0	0	0

Phenological stage: 119: 19th leaf of main stem unfoled (>4cm), 121: First leaf of 2nd order branch above first inflorescence unfolded (> 4 cm), 501: First individual buds (1-2mm) of first inflorescence visible (main stem), 505: Buds of first inflorescence extended to 5 mm, 509: First flower petals of first inflorescence visible, 600: First open flowers in population, 601:Beginning of flowering: 10 % of flowers in the first inflorescence open (main stem), 605: Fullflowering: 50 % of flowers in the first inflorescence open, 639: End of flowering in the 3rd inflorescence, 700: First berries visible.

The correlation and regression between the phenological growth stages of potato plants and the temperature (°C) was checked by the analysis of the data for each BBCH scale code . According to the results, the R value of **119**= 65535, $R^2 = -1.52217E-16$, R value of **121**= 3.15011E-08, $R^2 = 9.92317E-16$, R value of **501**= 2.09844E-08, $R^2 = 4.40345E-16$, R value of **505**= 65535, $R^2 = -1.14377E-16$, R value of **509**= 0, $R^2 = 0$, R value of **600**= 2.32155E-08, $R^2 = 5.3896E-16$, R value of **601**= 0, $R^2 = 0$, R value of **605**= 65535, $R^2 = -1.4925E-16$, R value of **639**= 2.0978E-08, $R^2 = 4.40078E-16$, R value of **700**= 65535, $R^2 = -4.87229E-16$. From these values, there is no correlation and regression between the phenological growth stages of potato plants and the temperature (°C).

5.2.5.2. Phenological growth stages 2021

The beginning of the flowering phase in the non-irrigated treatment shows the highest mean value in the mixture of microbial inoculations *Rhizophagus irregularis* MucL41833 + *Paraburkholderia phytofirmans* PSJN + *Trichoderma asperelloides* A n (39.06), other treatments show a result, with the exception of *Rhizophagus irregularis* MucL41833 + *Pseudomonas Brassicacearum* PSJN, which shows no results. For full flowering, *Trichoderma asperelloides* A treatment has the mean value (76.56), followed by *Rhizophagus irregularis* MucL41833+*Pseudomonas Brassicacearum* PSJN (56.25). For the end of flowering, *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans* PSJN, control and *Rhizophagus irregularis* MucL41833+*Pseudomonas Brassicacearum* PSJN shows the same percentage of (6.25), *Paraburkholderia phytofirmans* PSJN and *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans* PSJN+ *Trichoderma asperelloides* A, which shows no results.

In the irrigated treatment, *Paraburkholderia phytofirmans* PSJN treatment shows the highest mean value of beginning of flowering (15.62), *Pseudomonas brassicacearum* 3Re2-7, *Rhizophagus irregularis* MucL41833, *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans* PSJN and *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans* PSJN+ *Trichoderma asperelloides* A shows the same mean value (1.56). The control treatment shows a results (6.25) while *Rhizophagus irregularis* MucL41833+*Pseudomonas brassicacearum* PSJN and *Trichoderma asperelloides* A which shows no results. For the full flowering, all treatments shows a results, *Trichoderma asperelloides* A shows (100.00) followed by *Rhizophagus irregularis* Muc L41833 (87.5). For the end of flowering phase only *Rhizophagus irregularis* MucL41833+*Pseudomonas brassicacearum* PSJN treatment shows a results (1.56). Table 20 shows all mean values.

Table 20. Phenological growth stages of plant during 2021 (Budapest, 2022).

Treatment	Irrigation	601	SD.	605	SD.	609	SD.
CO (control)	Control	1.56	2.2	39.06	11.04	6.25	0
<i>Paraburkholderia phytofirmans</i> PSJN		37.5	0	36.45	7.36	0	0
<i>Pseudomonas brassicacearum</i> 3Re2-7		12.5	17.67	14.06	6.62	14.06	6.62
<i>Rhizophagus irregularis</i> MucL41833		37.5	0	25	17.67	1.56	2.2
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN		6.25	0	25	0	6.25	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN+ <i>Trichoderma asperelloides</i> A		39.06	11.04	17.18	11.04	0	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Pseudomonas brassicacearum</i> 3Re2-7		0	0	56.25	0	6.25	0
<i>Trichoderma asperelloides</i> A		1.56	2.2	76.56	15.46	0	0
CO (control)	Irrigated	6.25	0	75	0	0	0
<i>Paraburkholderia phytofirmans</i> PSJN		15.62	13.25	56.25	0	0	0
<i>Pseudomonas brassicacearum</i> 3Re2-7		1.56	2.2	76.56	15.47	0	0
<i>Rhizophagus irregularis</i> MucL41833		1.56	2.2	87.5	0	0	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN		1.56	2.2	76.56	15.46	0	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Paraburkholderia phytofirmans</i> PSJN+ <i>Trichoderma asperelloides</i> A		1.56	2.2	76.56	15.46	0	0
<i>Rhizophagus irregularis</i> MucL41833+ <i>Pseudomonas brassicacearum</i> 3Re2-7		0	0	76.56	15.46	1.56	2.2
<i>Trichoderma asperelloides</i> A		0	0	100	0	0	0

Phenological stage: 601: Beginning of flowering: 10 % of flowers in the first inflorescence open (main stem), **605:** Full flowering: 50 % of flowers in the first inflorescence open, **609:** End of flowering in the first inflorescence.

The regression between the phenological growth stages of potato plants and the temperature (°C) was checked by the analysis of the data for each BBCH scale code . According to the results, the R value of **601**= 7.29545E-08, R^2 = 5.32236E-15, R value of **605**= 65535, R^2 = -2.92337E-15, R value of **609**= 3.96267E-08, R^2 = 1.57028E-15. From these values, there is no correlation and regression between the phenological growth stages of potato plants and the temperature (°C).

6. DISCUSSION

6.1. Sweet potato pilot study

The present study demonstrated that the used mycorrhizal inoculum could establish a symbiotic relationship with the treated and controlled sweet potato seedlings. Confirmation of establishment can be demonstrated using the calculated mycorrhizal parameters (Table 13). GAI et al. (2006) also showed that several mycorrhizal fungal species can colonize sweet potatoes to varying degrees. More than 90% of plant species can form symbiosis with arbuscular mycorrhizal fungi, indicating their common abundance (ALIZADEH 2011). This is also visible in our results as sweet potatoes grown on control soil also showed signs of colonization. Arbuscular mycorrhizal fungi have effectively inoculated cotton, tomato, pepper, faba bean, garlic, soybean, cucumber, melon, watermelon, corn, and eggplant plants (ORTAS 2012).

To ensure colonization, some prerequisites must be met, specifically, the simultaneous use of a variety of arbuscular mycorrhizal fungal species, a high amount of infective propagating fungi, the absence of pathogens and pests, the availability of useful bacterial additives, and the use of dry solid inoculum (ROUPHAEL et al. 2015). These requirements were also observed in our results, as colonization was highest when the commercial inoculant was used with multiple arbuscular mycorrhizal fungi species and a controlled propagation number, while the growth medium was sterilized and free of pests and pathogens. Arbuscular mycorrhizal fungi can promote plant growth directly and indirectly. It can directly promote the growth of the root system through the modulation of host phytohormones, leading to its indirect growth promotion through the increased availability of some immobile nutrients through the enlarged root zone (MARSCHNER 2012; PONS et al. 2020; PILIAROVÁ et al. 2019). In addition to immobile nutrients, mycorrhiza can also help accumulate nitrogen from its various forms such as nitrate (NO_3), ammonium (NH_4^+) and amino acids using their extra-radical hyphae (JIN et al. 2012). Some studies have demonstrated the positive effect of arbuscular mycorrhizal fungi on the absorption of phosphorus, nitrogen, potassium, magnesium, copper, zinc, calcium, iron, cadmium and nickel (WANG et al. 2017; KRISHNAKUMAR et al. 2013).

Arbuscular mycorrhizal symbiosis can influence biochemical and physiological processes such as protection against oxidative damage, improve water use efficiency, increase shoot weight, enhance gas exchange rate, and promote osmotic regulation (RUIZCHEN et al. 2017).

From our results on fresh shoots and root weights, it appears that mycorrhizal inoculation could improve the physical parameters of sweet potato seedlings. Several other studies have already shown the positive effect of arbuscular mycorrhizal fungi on physical parameters. In maize, it has been demonstrated that *Glomus intraradices* can enhance dry weight of shoots and roots (ORTAS 2011). KAKABOUKI et al. (2021) examined the effect of *Rhizophagus irregularis* on cannabis seedlings and found significantly increased root length, significantly increased stem dry weight, and an improvement in survival rate and phosphorus content. Another study also confirmed that arbuscular mycorrhizal fungi increased the fresh weight of sweet potato sprouts and roots in sweet potato varieties PROC 65-3 (white-fleshed) and Tainung 57 (orange-fleshed) (NEUMANN et al 2009). In addition, in the study of SAKHA and JEFWA (2019), two sweet potato cultivars, Kemb- 10 and Bungoma, were examined with and without arbuscular mycorrhizal fungi inoculation, focused on physical parameters, namely branch number, vine length and yield; They found that mycorrhizal inoculation improved yield and growth (REDDY et al 2018).

The yield of storage roots correlates positively with the vegetative properties and the correlation with the number of leaves per plant is significant (MARTIN 2013). Regarding the length of the roots, we did not find any positive effect of mycorrhizal inoculation, while the length of the stem increased. The effect of the sterilization treatment was small; the reason for this could be that the microbial population in the autoclaved growth medium was lower than in the non-autoclaved growth media. This is consistent with the results of KÖHL et al. (2016). Nevertheless, the defined influence of arbuscular mycorrhizal fungi on plant growth and development is not stable due to the complex relationship between arbuscular mycorrhizal fungi, the inoculation method and environmental conditions (PERNER et al. 2006). Our results showed that although the highest colonization rates were observed in sweet potatoes grown on sterilized peat, the treatment did not perform better in terms of physical parameters compared to non-sterilized inoculated treatments. Furthermore, this can be confirmed by other studies showing that plants with high mycorrhizal colonization rates can be maintained on peat-based substrate, but that plants may not consistently benefit from mycorrhizal symbiosis growth under these conditions (BAUM et al. 2015). However, the interaction between arbuscular mycorrhizal species may differ between sweet potato varieties (WIPF et al. 2019). In our experiment, the mycorrhizal inoculum (Symbivit) enhanced the fresh weight of shoots (cm), fresh weight of total roots(g). Also, the length of roots and stem(cm) in both sweet potato varieties, orange and purple.

6.2. Organic potato experiment

Our recent study showed that arbuscular mycorrhiza can form a symbiotic relationship with potato tubers in both growing seasons under irrigated and non-irrigated conditions. ZHU et al. 2022 also showed that the combination of arbuscular mycorrhizal fungi with other compounds can further promote the establishment and growth of arbuscular mycorrhizal fungi, improve the nutrient utilization rate of the host plant, and thus strengthen the symbiotic connection between plant and mycorrhizal fungi. LARANJEIRA et al. (2022) found that inoculation with beneficial microorganisms and supplemental irrigation during critical stages promotes chickpea growth and should be considered to increase crop productivity and promote agricultural sustainability. Our results show that mycorrhizal colonization frequency and mycorrhizal intensity increased over the two years under non-irrigated conditions, demonstrating that the applied mycorrhizal inoculants were successful in establishing a symbiotic relationship with the treated potato tubers. This can be confirmed by AUGÉ (2004) that arbuscular mycorrhizal fungi help plants absorb water, and numerous mechanisms have been postulated to explain these effects. These include improved stomatal regulation, higher hydraulic conductivity of roots and increased interaction with soil particles. Most treatments showed no arbuscular abundance in both years, except for the mixture of *Rhizophagus irregularis* MucL41833 + *Pseudomonas brassicacearum* 3Re2-7 (41 %), *Rhizophagus irregularis* MucL41833 + *Paraburkholderia phytofirmans* PSJN (24 %) and *Trichoderma asperelloides* A (33 %).

There was no significant difference in starch content in the two seasons under different treatments and irrigation conditions. However, the potato tubers treated with mycorrhizal inoculants and the microbial inoculants mixture also yielded the highest starch content. In 2020, the highest starch content was observed in *Paraburkholderia phytofirmans* PSJN treatment with a mean of (17.16 %) under irrigated and non-irrigated conditions. while in 2021, the highest starch content among the irrigated plots was found in the *Rhizophagus irregularis* MucL41833 treatment (12.29

%). A study by BERTA et al. (2014) showed that inoculation with plant growth promoting bacteria and arbuscular mycorrhizal fungi increased starch content. Since the development of arbuscular mycorrhizal fungi can also increase with time, the increase in starch content can be explained by the improvement in the development of arbuscular mycorrhizal fungi over time.

According to 2020 phenology measurements, the mean value was the highest in nearly every growth stage in the non-irrigated treatment than in the irrigated treatment. Higher values were also obtained by the combination of the microbial inoculants *Rhizophagus irregularis*

MUCL41833+*Paraburkholderia phytofirmans* PSJN+ *Trichoderma aspeloides* A and *Trichoderma* A. With the exception of full flowering, the irrigated treatments had the highest percentage in the second season (2021) of planting. There was an increase in the percentage with relation to the applied microbial treatment in the mixture of the applied microbial inoculates. This can be confirmed by WU et al. (2013) that the beneficial microorganisms mainly play a protective role against biotic and abiotic stresses, which frequently leads to enhanced host plant growth, fitness, and eco-system health. The total phosphorus content in potato tubers increases with time and with the microbial inoculants used. Nevertheless, there is no significant difference between the treatments and irrigation conditions in our study. The control treatment in the second year (2021) gave the highest total phosphorus content under irrigation conditions (0.69 mg kg⁻¹), followed by the mixed treatment of the microbial inoculated plants *Rhizophagus irregularis* MucL41833 and *Paraburkholderia phytofirmans* PSJN (0.68 mg kg⁻¹). This can correspond with a study carried out by MA et al. (2021) showed that the mycorrhizal colonization and activity can be affected by several factors which may lead to decrease the arbuscular mycorrhizal colonization such as; high nutrient content in the soil, high temperature and precipitation can also affect negatively the development of mycorrhiza, high phosphorus supply decreases root colonization, and root cadmium content decreases the root mycorrhizal colonization. Furthermore; other study by AVIO et al. (2013) showed that intensive soil-tillage also affects negatively the development of arbuscular mycorrhizal fungi spores. Among other possible reasons; the interaction between arbuscular mycorrhizal fungi and other microorganisms might have an antagonistic effect on the mycorrhizal colonization and development, there was no soil solarization during the experiment, therefore there was a variation of the microorganisms which might affect negatively the arbuscular mycorrhizal fungi activity, and the influence of the soil and environmental conditions during the experiment application.

Research by ADAVI and TADAYOUN (2014) concluded that tuber size; number of tubers per plant, tuber yield and starch yield are significantly affected by mycorrhizal inoculation because this biofertilizer can improve the plant's uptake of phosphorus. An increase over time was also observed as an overall result of the effect of different treatments on potato yield. In the first year, *Paraburkholderia phytofirmans* PSJN gave the highest yield (15.21 kg/ m²) under irrigation conditions, while *Rhizophagus irregularis* MucL41833 gave the highest yield (16.72 kg/ m²) in the second year also under irrigation conditions. This is shown by a study by SZCZABA et al. (2019), which shows that the combination of arbuscular mycorrhizal fungi and *Trichoderma* has a positive effect on plant yield.

Mixing the inoculation with different species could have an antagonistic effect or no effect, according to studies. For the mixture of plant growth promoting rhizobacteria and arbuscular mycorrhizal fungi, inoculation of a mixture of the microbial inoculates such as plant growth promoting rhizobacteria species; *Azospirillum* with *Pseudomonas* had no effect on maize plant growth (VAZQUEZ et al. 2000). Furthermore, inoculation of *Pseudomonas* and *Trichoderma* reduced the activity of other inoculated microorganisms. Colonization with arbuscular mycorrhizal fungi can eliminate the effect of *Trichoderma* on grapevine growth (WASCHKISET al. 1994). Inoculation of just one microorganism in the plant can have a significant positive effect on the plant. However, inoculation with other microorganisms, especially arbuscular mycorrhizal fungi, can lead to a weakening of the effect of other inoculations. This could be explained by the qualitative change in root exudate caused by arbuscular mycorrhizal fungi colonization (COX et al.1975). In our research, the results show that the treatments showed no significant difference in most measurements in both years of study. There was no significant difference between the 2020 and 2021 results for both inoculation treatments. The non-irrigated plants showed better results in terms of arbuscular mycorrhizal fungi colonization, higher starch content and higher total phosphorus content in the non-irrigated samples compared to the irrigated ones.

7. CONCLUSIONS AND RECOMMENDATIONS

Based on the results from the sweet potato pilot study, it can be concluded that the symbiotic relationship was successfully developed; we could observe this by the scoring of the mycorrhizal parameters (F %, M %, m %, a %, A %) in the stained roots under microscope. We could also detect different mycorrhizal developmental structures such as hyphae and arbuscules, especially in the seedlings that were treated with Symbivit in sterilized peat moss. According to our observations, substrate sterilization may influence microbial inoculates that improve nutrient uptake, protect plants from pests and diseases, and promote plant growth can replace agrochemicals in food production. The high intensity of mycorrhizal colonization in the root system (M %) of the purple variety in the treatment [(L+SYM).S] compared to the treatments (L+SYM) with control (F) with low mycorrhizal intensity. For the orange variety, the sterilization treatment [(L+SYM).S] also had the highest mycorrhizal intensity, but the (L+SYM) without sterilization had the lowest intensity of mycorrhizal colonization in the root fragments (m %). The arbuscular abundance in the mycorrhizal parts (a %) showed that in the purple variety, a significantly higher percentage was recorded in (L+SYM) than in [(L+SYM). S]. However, no arbuscules were found in the (F) treatment, although in the orange variety, arbuscules were found only in the [(L+SYM).S] treatment, while no arbuscules were found in the (L+SYM) and (F) treatments. For the arbuscule frequency in the root system (A %), in the purple variety, the highest arbuscule frequency was recorded in (L+SYM) followed by [(L+SYM).S], while no arbuscule were found in the (F) treatment. In the orange variety, arbuscules were found only in the [(L+SYM).S] treatment, while no arbuscules were detected in (L+SYM) and the control treatment (F). The highest mean shoot fresh weight for orange sweet potato seedlings was found in the L+SYM treatment. There was no significant difference between treatments [(L+SYM). S] and (F), therefore, it can be concluded that mycorrhizal inoculum could increase shoot weight over time, while sterilization had no increasing effect on shoot weight (g) in treated orange sweet potato seedlings. The highest fresh shoot weight in Purple sweet potato seedlings was achieved under (L+SYM) treatment, mycorrhizal inoculation also increased shoot fresh weight in the purple sweet potato seedlings, whereas sterilization had no effect on shoot fresh weight. The highest root weight was observed in orange sweet potato seedlings in [(L+SYM). S] treatment. This means that mycorrhizal inoculation and sterilization had a large impact on the root weight of orange sweet potato seedlings. For the weight of the roots of purple sweet potato seedlings, the mycorrhizal inoculation had a stimulating effect on the fresh root weight of purple sweet potato seedlings. The

highest root length of the orange sweet potato variety was measured in seedlings treated with Symbivit in sterilized substrate (peat moss) [(L+SYM). S]. However, there was no significant difference between the average values when comparing the three levels of treated orange sweet potato seedlings. Therefore, mycorrhizal inoculation with sterilization had no effect on the length of the roots of the orange sweet potato seedlings. The root length increased in the purple sweet potato seedlings propagated in a sterilized Latagro peat moss with Symbivit[(L+SYM).S] treatment. However, the differences between the treated seedlings were not significant. Mycorrhizal inoculation could increase the length of stems in orange sweet potato seedlings. On the other hand, sterilization had no increasing effect on stem length in orange sweet potato seedlings. For the purple sweet potato seedlings, mycorrhizal inoculation had a positive effect on root length. However, sterilization had no stimulating effect on the root length of purple sweet potato seedlings.

In the two-year experiment, we examined the effect of different microbial inoculations applied in combination of different treatments under irrigated and non-irrigated conditions in two seasons. The result shows that the mycorrhizal colonization developed successfully and established a symbiotic relationship with potato tubers. This can be observed in the measured mycorrhizal parameters (F %, M %, m %, a %, A %) in both seasons. Starch content was similar among different treatments in both years, with no significant differences. In 2020, the highest starch value was observed in *Paraburkholderia phytofirmans*PSJN treatment on average under irrigated and non-irrigated conditions, respectively, followed by *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans*PSJN treatment under irrigated conditions. The lowest starch content was found in the irrigation control treatment, while the lowest starch content in the non-irrigation treatment was found in the *Rhizophagus irregularis*MucL41833+ *Paraburkholderia phytofirmans* treatment. Also, in the second season (2021) with the highest starch content in *Rhizophagus irregularis*MucL41833 treated tubers, there were no significant differences in both irrigated and non-irrigated conditions. In 2021, the highest starch value among the irrigated plots was found in *Rhizophagus irregularis* MucL41833 treatment. The lowest starch content was recorded in the *Pseudomonas brassicacearum*3Re2-7 treatment. Under non-irrigated conditions, the highest starch content was found in *Pseudomonas brassicacearum* 3Re2-7treatment followed by *Rhizophagus irregularis* MucL41833 treatment. The lowest starch content was apparent in the *Rhizophagus irregularis*MucL41833+ *Paraburkholderia phytofirmans*PSJN+ *Trichoderma asperelloids*A treatment. The results show non-significant differences in both years under both irrigated and non-irrigated conditions. The non-irrigated treatment during the two years of the

experiment showed the highest mean value of the phenological stages. Additionally, the various phenological stages of the potato plants increased as a result of the combination of microbial inoculants. The highest phosphorus content was measured for the *Trichoderma asperelloides* A treatment under irrigation conditions and for the control treatment without irrigation. In the second year (2021), there was an obvious increase in phosphorus content, but the highest value was measured in the control treatment under irrigation conditions and in the *Rhizophagus irregularis* MucL41833+ *Paraburkholderia phytofirmans* PSJN treatment without irrigation. Tubers yield was not significantly affected by any of the treatments in the two test seasons. The yield of the irrigated treatments was higher than that of the non-irrigated treatments in both seasons. As for the inoculation effect, *Paraburkholderia phytofirmans* PSJN achieved the highest yield under irrigation in the first season, but *Rhizophagus irregularis* MucL41833 was the highest in the second season. Based on the irrigation treatment, it can be concluded that the microbial inoculations achieved better results under non-irrigated conditions than under irrigated conditions. It can be concluded that there is no positive effect with any of the inoculates, in almost all measurements we found that there is no significance difference between microbial inoculates treatments. Even with the non-irrigated treatment, no significant benefit from inoculates was measurable.

8. NEW SCIENTIFIC RESULTS

1. Our work proved that the sweet potato seedlings for both varieties, purple (Purple) and orange (Norangel) can establish a symbiotic relationship with the Symbivit mycorrhizal inoculum. Mycorrhizal inoculation with a sterilized substrate performed better results on root length (cm) among the studied sweet potato varieties.
2. We demonstrated that the inoculation of tested sweet potato seedlings with Symbivit inoculum can be functional, which presumably improves plant growth.
3. Colonization of arbuscular mycorrhiza is higher in arid climate in organic potato cultivation in case of Desiree variety, under non-irrigated environment.
4. Starch content in potato tubers increasing by the water supply not by the applied microbial inoculates (Arbuscular mycorrhizal fungi, plant growth promoting rhizobacteria and Trichoderma).
5. There is no influence of the water supply and the microbial inoculates applied together (Arbuscular mycorrhizal fungi, plant growth promoting rhizobacteria and Trichoderma) on the yield of potato tubers

9. SUMMARY

9.1. Sweet potato pilot research

A sweet potato is an increasingly important crop and growing them effectively and sustainably has gained importance in temperate countries. The purpose of this pilot study was to investigate the effects of mycorrhizal inoculum, Symbivit, and whether it can establish a symbiotic relationship with the seedlings of two sweet potato varieties (orange and purple). The effectiveness of mycorrhizal inoculation with a sterilized substrate on mycorrhizal parameters (F %, M %, m %, a %, A %) and physical parameters (length of roots and shoots (cm), fresh weight of shoots and roots (g) and the stem length (cm)) of the sweet potato seedlings were also examined. The results show that sterilization treatment with Symbivit increased the incidence of mycorrhiza in the root system in both cultivars. There was a difference between the varieties, mycorrhizal inoculum and the sterilization treatment with regard to the intensity of mycorrhizal colonization in the root fragments and the arbuscular frequency. Overall, the preliminary results provided remarkable information on mycorrhizal inoculation, substrate sterilization and mycorrhizal development, as well as changes in physical parameters between sweet potato seedlings. Our results could serve as a practical strategy for further research to give meaning to the effect of beneficial soil microbes on sweet potatoes.

9.2. Organic potato experiment

Green technologies such as microbial inoculation to replace or reduce the use of agrochemicals and maintain a clean environment are good solutions to current agricultural problems. It is known that many microorganisms have benefits for plants and can represent alternatives to chemical products that are suitable for environmental protection and plant value. These microorganisms include arbuscular mycorrhizal fungi, plant growth promoting rhizobacteria and *Trichoderma* spp. that can live in symbiosis with plants. Seven different treatments of microbial inoculum strains were applied under irrigated and non-irrigated conditions in two cultivation periods to observe whether they had a positive effect on potato tuber improvement under irrigated and non-irrigated conditions. Several parameters were measured during the study, such as: Mycorrhizal parameters (F %, M % and A %), total phosphorus in potato tubers, total starch content, phenological measurements and potato tuber yield. The results indicate

that non-irrigated plots all performed better for arbuscular mycorrhizal fungi colonization, but no impact on yield or quality was seen because there was no significance difference between microbial inoculates treatments. Our results could prove to be a practical strategy for further researches and field experiments into microbial inoculation on potatoes.

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13. APPENDIX



Symbivit (Mycorrhizal inoculum)



Mixing mycorrhizal inoculum (Symbivit) with the substrate



Sweet potato seedling sweet potato seedlings transplanting



Transplanting of the sweet potato seedlings in the sterilized peat moss