GUIDE FOR INOCULATIONS WITH MYCORRHIZAL FUNGI FOR MEDITERRANEAN SILVOPASTURE SYSTEMS

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Guide for inoculations with mycorrhizal fungi for Mediterranean silvopasture systems

What are mycorrhizal fungi?

Mycorrhizal fungi are those that establish mutualistic symbiotic associations with the roots of terrestrial plants. Most of the plant species that exist on the earth's surface are mycorrhized.

Figure 1: Summer truffle mycorrhiza seen in the binocular loupe. Own soruce

Mycorrhization is the process, natural or artificial, by which the root of an actively growing plant comes into contact with a mycorrhizal fungus and the structure resulting from this contact is the **mycorrhiza**. Mycorrhizae are classified according to the way the fungus penetrates the root, by the characteristic structures it develops, and the species of fungi and plants involved. There are two predominant types of mycorrhizae: ectomycorrhizae and endomycorrhizae.

What are the benefits of mycorrhization?

The main benefits of mycorrhization of plants are the following:

- ✓ Improves the assimilation and uptake of nutrients, especially minerals, such as phosphorus, calcium, potassium and even nitrogen.
- ✓ Increases the water absorption capacity of the plant
- ✓ Helps protect the plant against pathogenic organisms.
- ✓ Improves the physiological state and activates the roots.

Therefore, the application of mycorrhizal fungi in the soil of Mediterranean silvopasture systems, such as dehesas and montados, can be beneficial to improve the vigor of these ecosystems while protecting against possible diseases.

What fungi do we inoculate with?

When choosing the species of fungi to use, both the characteristics of the soils and the tree species that dominate the ecosystem must be taken into account. In the case of the Mediterranean silvopasture systems (dehesas and montados), the stands are generally dominated by species of the *Quercus* genus, mainly holm oaks (*Q. ilex*) and cork oaks (*Q. suber*). These trees are associated with ectomycorrhizal fungi.

From among the multitude of ectomycorrhizal fungi that can establish symbiosis with the roots of holm oaks and cork oaks, in this guide we have selected two species that in our opinion may

be more interesting for inoculations in Mediterranean silvopasture systems: *Pisolithus tinctorius* (or *P. arhizus*) , whose capacity to improve the absorption of water and nutrients as well as protection against diseases has been widely studied and scientifically corroborated, and summer truffle or *Tuber aestivum* which has the added advantage that, in case of fruiting, the truffle obtained has a high culinary and economic value.

It is important to highlight that the protective role against diseases of this type of fungi is limited to the prevention of the disease in healthy trees, not being effective as a curative method of a present disease.

Figure 2: Fruiting bodies of P. tinctorius (left) and T. aestivum (right). Own source

How are inoculations done?

Inoculations will be done in the **fall**. The soil needs to be slightly damp.

It is important to select **healthy trees** in **well-drained soils**, since these species of fungus do not withstand waterlogging well.

Detailed steps to follow for inoculations with the two selected fungal species are described below:

1.- *Pisolithus tinctorius* inoculations

- ✓ Manually dig at least five trenches 15 cm deep, radially from the base of the trunk outwards, reaching approximately the edge of the crown projection.
- ✓ Introduce in each trench the commercial spore inoculum of *P. tinctorius* (approximately one liter per trench, with a minimum concentration of 1x10^8 spores / l). For the correct distribution of the inoculum it is recommended to apply it previously mixed in a solid substrate, for example exfoliated vermiculite.
- ✓ Mix the inoculum well with the soil and completely cover the trenches.
- ✓ Water after inoculations.

Figure 3: Radial trenches opened with hoe in the area of influence of the roots (left) and application of P. tinctorius inoculum previously mixed in exfoliated vermiculite (right). Own source

2.-*Tuber aestivum* inoculations

- ✓ Since the soils of dehesas and montados are usually acidic, and summer truffle needs a neutral - basic pH, it will be necessary to correct the pH of the soil by means of limestone amendments (application of calcium carbonate).
    - It is necessary to know the initial pH of the soil from a previous analysis
    - It is advisable to perform the limestone amendment before performing the inoculations
    - Gradual amendments should be made, without raising more than one pH point each year to avoid damaging the soil.
    - The pH target to be achieved is 7.5.
- ✓ Decompact the environment of the tree (root influence area) by means of a rototiller or rotovator.
- ✓ Add the commercial *Tuber aestivum* sporal inoculum. For the correct distribution of the inoculum it is recommended to apply it previously mixed in a solid substrate, for example exfoliated vermiculite.
- ✓ Mix the inoculum well with the soil and smooth the ground.
- ✓ Water after inoculations.
- ✓ Restrict livestock to avoid compaction of the land that prevents truffles from bearing fruit.

Figure 4: Limestone amendment to increase pH (left). Application of T. aestivum inoculum mixed in exfoliated vermiculite on soil removed with a rotovator (right).

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