

TECHNICAL DATA SHEET Copper nanoparticles application

Atypical practice to reduce GTDs incidence in vineyard



This document is the result of interviews made on field with the principal objective to highlight the diversity of techniques used in field. To date, no assessment, no validation or checking efficacy of this practice was made. In the absence of any assessment, its success in different conditions of the ones exposed is not guarantee and the responsibility of Winetwork partners can't be involved.

Network for the exchange and transfer of innovative knowledge between European wine growing regions



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Introduction

Nanotechnology is already being used in agriculture for the treatment of some plant diseases. Their use may increase the efficacy of commercial pesticides and insecticides by reducing their amount of application at significantly lower doses required for crops with the environmental enhancement entailed.

The application of copper nanoparticles to fight against grapevine trunk diseases (GTDs) is a practice that has been identified in a vineyard from Galicia, in Spain. They are carrying out periodic applications (via trunk injection) of aqueous solution of copper nanoparticles in 26 years old Albariño vines damaged by Esca. They also perform foliar treatments, spraying of a mixture of copper nanoparticles and an elicitor based on amino acids from porcine blood. Results have not been observed yet, since 2016 has been the first year of application. They will perform the applications at least two years more

Description

The application of **copper nanoparticles** (Cu NPs) to fight against GTDs is a practice that has been identified as innovative, since it was detected in only one interview out of the 219 realized throughout the 10 regions that participate in the Winetwork project.

The application of Cu NPs is being carried out in vines of Albariño **affected by Esca complex**, and located in a vineyard at Galicia (Spain) (fig. 1)

Characteristics of the vineyard in which the practice is being carried out:

- Rootstock: R110 y 196-17 CL
- Cultivar: Albariño
- Age: 26 years (in 2016)
- Planting density: 1200 plants/ha
- Yield: 8000-9000 kg/ha
- Training system: parral (arbour)
- Pruning system: Guyot. Pruning is started in November and ended in February. Manual scissor is used and each pruner carries a sprayer with diluted bleach (50:50). It is strictly ordered to disinfect the scissors after pruning every vine. Pruning debris is grinded and removed. Pruning wounds are protected with mastic immediately



Figure 1: Foliar symptoms in vines cv. Albariño affected by Esca (FEUGA, Spain)

All plants showing Esca symptoms are marked annually with rigorous control about their evolution or possible uproot and renewal.

The practice is carried out as follows:

 Two injections of copper nanoparticles. The first one at the **bud break** (April) and and the second one at **veraison** (end of August): An aqueous solution of Cu NPs (15 mg/l) is directly injected (using a syringe) in the trunk, doing previously a hole with a drill (bit 8). Then, the hole is covered with cotton (fig. 2)



Figure 2: Injection of Cu NPs solution into a vine affected by Esca and hole cover with cotton (FEUGA, Spain)

During these 4 months, between April and August, 4 to 5 foliar treatments are performed by spraying a mixture of Cu NPs (ratio 10:1000) and an elicitor based on amino acids from porcine blood (3:1000).

This practice was applied for the first time in 2016, and it is planned to continue for another two years, so **no results can be yet assessed.**

Application area

The treated vineyard is located in Condado county, southwest of Galicia, in D.O. Rías Baixas, Spain. The region is characterized by loam-sand soil with clay and river stones, coastal oceanic climate with mild temperatures and abundant rainfall and no snow or frost.



Figure 3: Area of application of copper nanoparticles. This technique was noticed in one region, Galicia, within the 219 interviews from Winetwork project.

Scientific data

Copper has a special interest because, unlike other antimicrobial metals, it has a **broad spectrum of action against bacteria and fungi**. It has been used extensively in the agricultural sector as a pesticide for thousands of years. The efficacy of copper **depends on environmental conditions**, the concentration of copper ions and the type of microorganisms. More in general, agrochemicals are usually applied by spraying. And sometimes, only a low quantity of the active ingredient reaches the target, due to leaching of chemicals, photolytic, hydrolytic and microbial degradation. Therefore, to have an effective control, it is necessary to use a bigger quantity of the active ingredient, with unfavorable effects such as water and soil contamination.

The use of nanotechnology in agriculture can **increase the effectiveness** of commercial pesticides and insecticides leading to an environmental improvement. To date it is already used in the control of some plant diseases and for the assimilation improvement fundamental nutrients by plants. In fact, nanoparticles can be used as **novel formulations** of pesticides, insecticides and insect repellents by nanoemulsion or nanoencapsulation techniques.

Nanoparticles of silica, polyethylene glycol, silver, aluminum, zinc oxide and titanium dioxide have been tested with promising results. For example, double-layer zinc-aluminum hydroxide compounds have been used for the controlled release of chemical compounds that regulate plant growth. Yields have been improved by the use of fertilizers incorpo-

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rated in nanotubes and the release of nitrogen caused by the hydrolysis of urea has been controlled through the insertion of urease enzymes into nanoporous silica particles. Some publications indicate that zinc oxide nanoparticles used in a wide variety of crops such as cucumber (Zhao et al., 2013), peanuts (Prasard et al., 2012), cauliflower, tomato (Singh et al.2013) helped to increase the efficiency of zinc use in crops.

Cu NPs have attracted more attention in recent years, because of its physical, chemical, antimicrobial properties, as well as for its abundance (Betancourt et al., 2013). In addition, copper has an important biological role in the plant photosynthesis, although it not take part in chlorophyll composition. Therefore, copper has great potential in the sustainable technological development. In relation to plant diseases, **Cu NPs were effective, in vitro, towards several fungi and yeasts** pathogens such as *Staphylococcus aureus, Pseudomonas aeruginosa, Salmonella choleraesuis* and *Candida albicans*, associated to serious diseases (Ren et al., 2009) (Rupareli et al.,2009) (Ramyadevi et al., 2012).

The **antimicrobial properties of copper** is linked to its ability in accepting or donate electrons, that is to have both high catalytic oxidation and reduction potential levels. In its oxidation state (Cu²⁺), it is highly effective as **antimicrobial**, due to the interaction with microbial nucleic acids, enzymatic active sites and cell membrane components .

Background

This practice is being carried out within the framework of a research project funded by national public funding. The beneficiaries are the winery in which the practice is being carried out and a nanotechnology company, which produces the Cu NPs and the elicitor under study.

Outcomes

The project started in 2016, so the trials have only been carried out for a year. The project will last for 3 years, so **it is still early to have results and make conclusions**.

Key points for success / risks

The **key point** of this practice is the **putative ability of Cu NPs for moving through the plant vessels to stop the GTDs**.

In this practice there is a clear environmental benefit, due to the drastic **reduction of copper quantity with the use of nanoparticles**.

Disadvantages:

- The current high price of the Cu NPs.

- Foliar treatments are not effective against other fungal diseases such as Powdery mildew and Downy mildew.

The potential risks that this practice could entail are the typical ones linked to any phytosanitary application, since appropriate product dressing and proper application are required (dosage and cleaning of barrel). To date, no adverse reactions have been observed in the plant after application, but it is convenient to be careful with the dosage, as copper have phytotoxic effects.

Innovative aspects

This practice is considered innovative because of the 219 interviews that were performed in the 10 European wine regions, this type of practice was identified only in one winery. The use of copper sulfate as a fungicide in viticulture is well known and widespread, but as we pointed out in previous sections, the application of nanotechnology in agriculture is a more environmentally friendly alternative for the specific case of insect and pest control than methods with synthetic agrochemicals, which have produced many environmental problems. Their use can increase the effectiveness of commercial pesticides and insecticides by reducing their amount of application to the soil at significantly lower doses required for the crops with the environmental improvement that implies. In addition to the lower environmental impact, there are studies that have shown that MIC (minimum inhibitory concentration) of bactericidal agent is lower for Cu NPs than for Cu²⁺ ions (Mallick et al., 2012), thus displaying a higher antimicrobial efficacy of the Cu NPs against copper sulphate.

Other requirements

This practice is **equally applicable to other types of vineyards**. There are no differences between varieties and vineyards. The protocol is de same.

In terms of cost, it should be noted that the treatment with copper nanoparticles has a **high cost.** About 450 euros per hectare.

Doses of copper in the vineyard have a legal limitation, but nanoparticles allow to work with doses well below those allowed (15 mg / l).

Source of information

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More information on

www.winetwork-data.eu

Technical datasheets:

- Good pruning practices
- Global vineyard strategy to prevent GTDs

Video clips:

- <u>Epidemiology and symptomatology of GTDs</u> (Dr. Vincenzo Mondello, URCA)
- <u>Scientific overview on Grapevine Trunk Diseases</u> (Dr. Vincenzo Mondello, URCA)

