INVESTIGATION OF SIZE AND EXPOSURE TIME DEPENDENT BIOACCUMULATION OF SILVER NANOPARTICLES IN PLANTS BY LIBS

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1. INTRODUCTION

Environmental and food safety is a very important issue in our world. Since nowadays nanoparticles are produced each year in the amount of thousands of tons for industrial purposes, they easily end up in the environment and can contaminate the food chain too. Thus, fast and reliable measuring tools are needed to monitor the bioaccumulation of NPs.

The aim of the project was to investigate the accumulation of silver nanoparticles in plants. The adverse effects of silver nanoparticles on the environment have been discussed in numerous publications, but no size-dependent bioaccumulation studies of silver nanoparticles in plant samples by LIBS have been performed so far. Earlier studies on animal and human tissues [Peng 2012] have found that smaller nanoparticles are more harmful than larger ones. The toxicity effect and the bioaccumulation of the nanoparticles and cadmium-containing quantum dots on plant samples were measured by LIBS [Modlitbová 2020a, Modlitbová 2020b].

2. EXPERIMENTALS

2.1. Instrumentation

A LIBS Discovery system were used in this study, the measurements were made with the following parameters: the laser pulse energy was set to 20 mJ, the gate delay was 0.75 μs, the number of shots per point was 1, the repetition frequency was 20 Hz and the distance between each measurement point was chosen to be 100 μm, this gave us the 100 μm spatial resolution. The measurements were performed in air. During the measurements the Ag (I) 328.062 nm line was monitored. A Czerny-Turner Shamrock

spectrometer equipped with an ICCD detector iStar 734 (both from Andor) was used for measurements.

2.2. Sample preparation

White mustard (*Sinapis alba*) was chosen for the model plants because they germinate relatively quickly in about three days. In the first phase of the experiment, the mustard seeds are placed in a Petri dish lined with filter paper soaked in deionized water. The seeds were placed at a sufficient distance (15 mm) to allow space for growth and germination. The seeds were kept for 3 days at 24 °C in a covered Petri dish.

After the third day, when the germs had grown to a sufficiently large size, the seeds were relocated into Eppendorf tubes containing the nanodispersion (**Figure 1.**). A 10 µmol/L nanodispersion of citrate-stabilized silver nanoparticles of different sizes, 10 and 40 nm, were used. Samples were grown in the nanoparticle solution for 48 and 72 hours, respectively. In the preliminary experiments, 5-5 samples grew for 48 hours in 10 nm and 40 nm particle solution, and 5-5 samples grew in 72 hours also in 10 and 40 nm particle solution and 5 samples grew in 72 hours in deionized water as a control group.

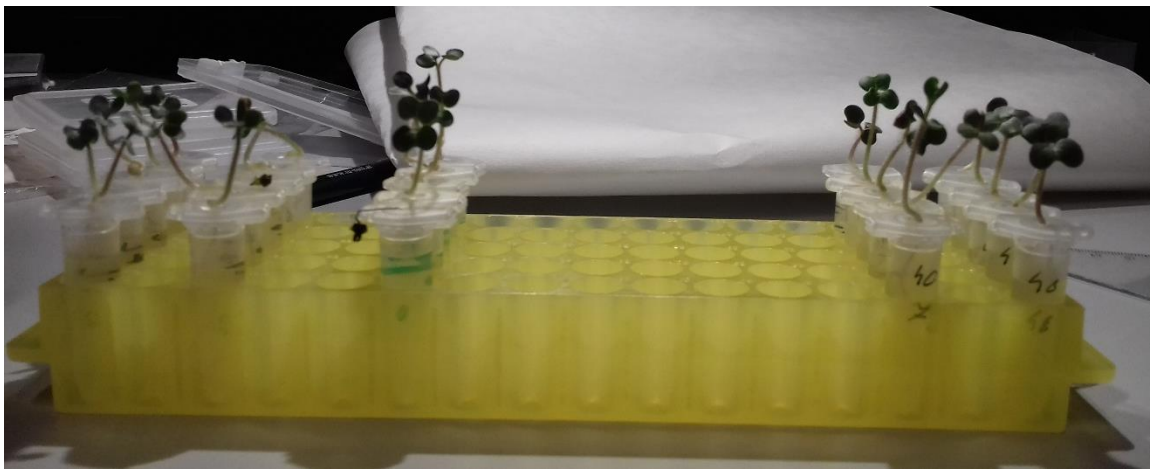

Figure 1. In the post-germination phase, plants were transferred to the Eppendorf tubes containing the nanodispersion, for 48 and 72 hours, respectively.

After each exposure time had elapsed, the samples were carefully removed from the tubes with tweezers due to their extreme fragility, and the roots were rinsed with deionized water to wash away any nanoparticles that were not absorbed but adhered to the root surface. The length of the root, stem, and total length were measured of each plant. Prior to the start of LIBS measurements, the pressed and dried plants were mounted on a glass slide with epoxy resin and allowed to dry for 24 hours.

3. RESULTS AND DISCUSSION

In this research the size and exposure time dependent bioaccumulation of silver nanoparticles were examined. Two, differently sized (10 nm and 40 nm) silver nanoparticles were used and the exposer times were chosen to 48 and 72 hours. As a control group plants were grown in ultra-pure water for 48 and 72 hours.

3.1 Size-dependent bioaccumulation

In **Figure 2.**, the silver distribution maps can be seen of those plants which were grown in 10 nm and 40 nm silver NPs containing nanodispersions for 48 hours. The colour scale ranges from blue to red, where blue indicates low silver intensity and red indicates high intensity. In **Figure 2. A**, the distribution of 10 nm, in **2. B**, the distribution of 40 nm sized silver nanoparticles is presented. The comparison of these maps reveal that the smaller nanoparticles are present both in the roots and in the plant's stem, but the larger particles are only enriched mostly in the roots of the plant. A possible explanation of this phenomenon is that smaller nanoparticles can easier pass through the intercellular barriers and membrane bilayers of the plant than larger ones.

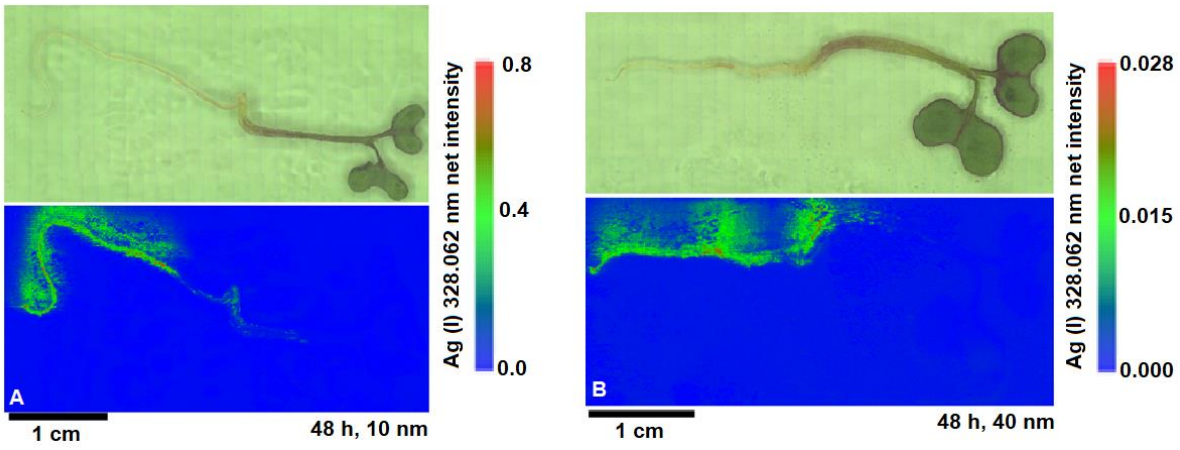

Figure 2. Optical microscopic image (on the top) and LIBS map of Ag (I) 328.062 nm of one of the plants grown in an aqueous dispersion of 10 nm (A) and 40 nm (B) silver nanoparticles for 48 hours.

It is also apparent that the concentration of the silver detected is also different, judged by the signal intensity. In those plants, which were grown in 10 nm NP-containing dispersions, the detected LIBS signals are more than twenty times higher, than in those which were grown in the 40 nm NPs containing nanodispersions. Since the exposition time was the same, this indicates a stronger accumulation of the smaller particles.

3.2 Time-dependent bioaccumulation

Another parameter that influences the accumulation of NPs is the duration of exposition. This effect was tested using two different exposition times: 48 and 72 hours. The result show that detected LIBS signals are twice as high for the 72 hour batch than for the 48 batch. Along with the expectations, this suggests that a longer exposition time produces stronger accumulation.

4. CONCLUSIONS

In this research we show that LIBS is a promising tool for this task. We successfully detected silver singes from plant samples grown in silver nanodispersions. Two aspects were examined one was the size dependence, in this part 10 nm and 40 nm sized NPs accumulation was monitored and we found that the smaller nanoparticles are more easily penetrate in the plants compared to the bigger ones. The second influencing factor which was examined was the exposer time. As we thought previously the longer exposer time cause higher NP deposition in the plants.

5. ACKNOWLEDGEMENTS

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6. References

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