

Changes in the antioxidant status of tobacco plants upon exposure to silver nanoparticles and silver nitrate

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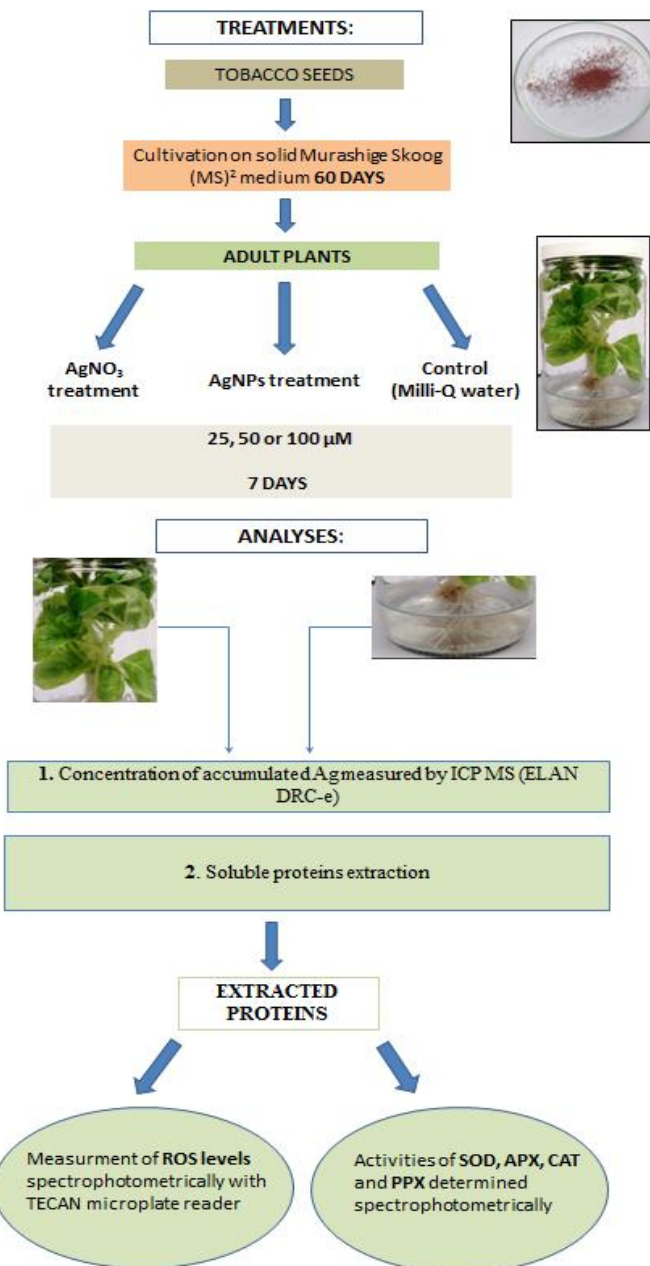
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INTRODUCTION

Silver nanoparticles (AgNPs) have a wide application in consumer products due to their antimicrobial properties. However, their increasing release into water or soil represents a potential environmental hazard. In this work, *in vitro* grown tobacco (*Nicotiana tabacum*) plants were exposed to AgNPs stabilized with cetyltrimethylammonium bromide (CTAB) or polyvinylpyrrolidone (PVP) coating, and to ionic silver (AgNO₃) of the same concentrations (25, 50 and 100 μM). The aim of the study was to investigate generation of reactive oxygen species (ROS) and changes in activities of antioxidant enzymes in regard to different treatments, thereby revealing if plants were exposed to oxidative stress. Silver uptake, ROS formation and activities of antioxidant enzymes catalase (CAT), ascorbate (APX) and pyrogallol peroxidase (PPX) and superoxide dismutase (SOD) were determined in leaves and roots extracts and compared to non-exposed, control plants.

MATERIALS AND METHODS



RESULTS

Table 1. Ag content measured in roots and leaves of adult tobacco plants. The results represent the mean value of 6 replicates ± standard error. Values marked with different letters represent significant difference ($p \leq 0.05$) according to Duncan test. Value $< 0.0001 \mu\text{g g}^{-1}$ represents instrument quantification bound.

Treatment	ADULT PLANTS	
	ROOT Ag concentration ($\mu\text{g g}^{-1}$ fresh weight)	LEAF Ag concentration ($\mu\text{g g}^{-1}$ fresh weight)
Control	$< 0.0001^a$	$< 0.0001^a$
25 μM AgNPs	1247.4 ± 122.0^b	12.1 ± 2.6^b
50 μM AgNPs	1395.2 ± 351.5^b	13.9 ± 2.6^b
100 μM AgNPs	1742.2 ± 192.8^{bc}	36.3 ± 3.4^c
25 μM AgNO ₃	1121.5 ± 136.2^b	18.2 ± 4.0^b
50 μM AgNO ₃	1450.9 ± 436.2^b	21.4 ± 7.3^b
100 μM AgNO ₃	1747.4 ± 150.0^{bc}	38.3 ± 5.4^c

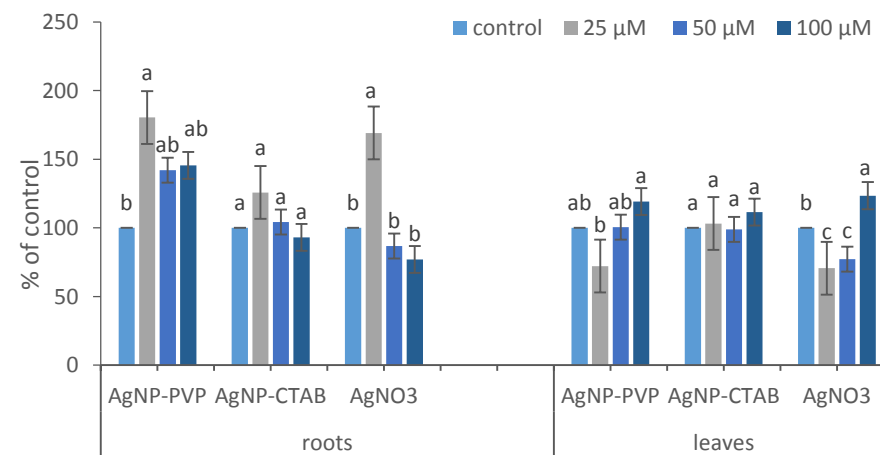


Figure 1. Levels of ROS in tobacco roots and leaves after exposure to 25, 50 and 100 μM AgNPs and AgNO₃. The presented results show mean values of 6 replicates ± standard error. Values marked with different letters represent significant difference ($p \leq 0.05$) according to Duncan test.

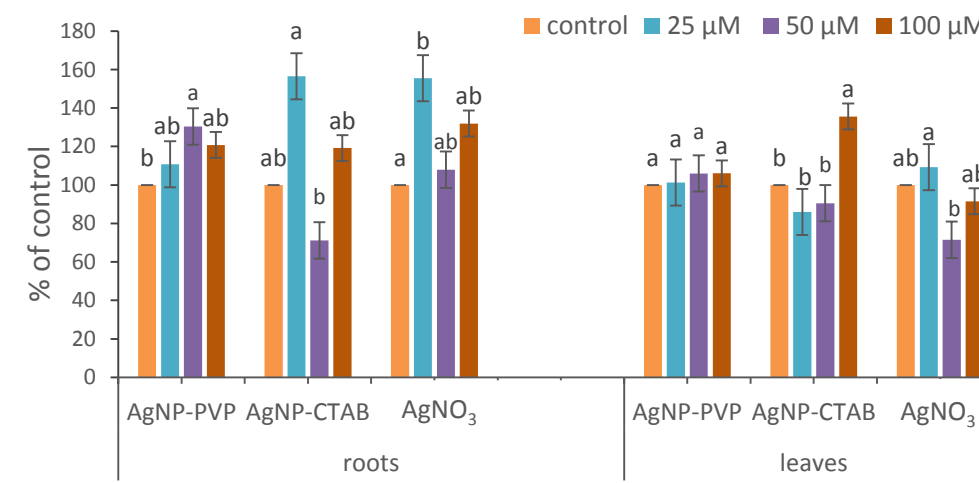


Figure 2. Activity of SOD in tobacco roots and leaves after exposure to 25, 50 and 100 μM AgNPs and AgNO₃. The presented results show mean values of 6 replicates ± standard error. Values marked with different letters represent significant difference ($p \leq 0.05$) according to Duncan test.

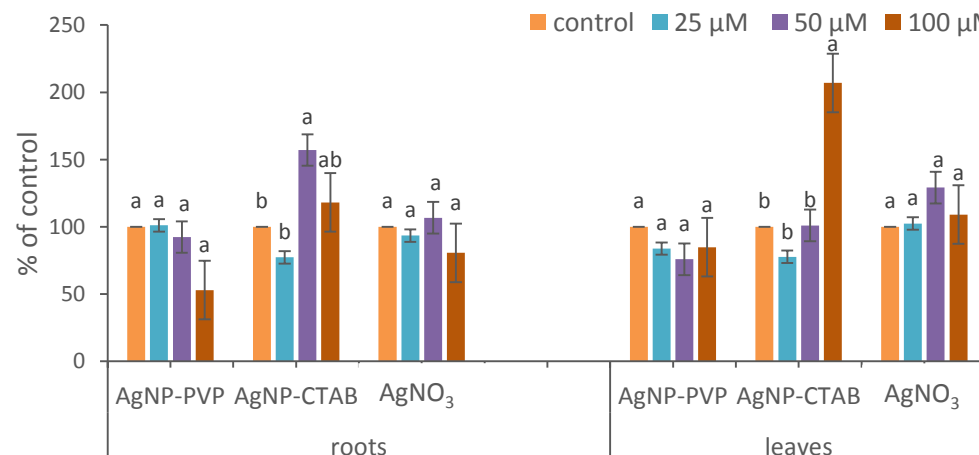


Figure 4. Activity of CAT in tobacco roots and leaves after exposure to 25, 50 and 100 μM AgNPs and AgNO₃. The presented results show mean values of 6 replicates ± standard error. Values marked with different letters represent significant difference ($p \leq 0.05$) according to Duncan test.

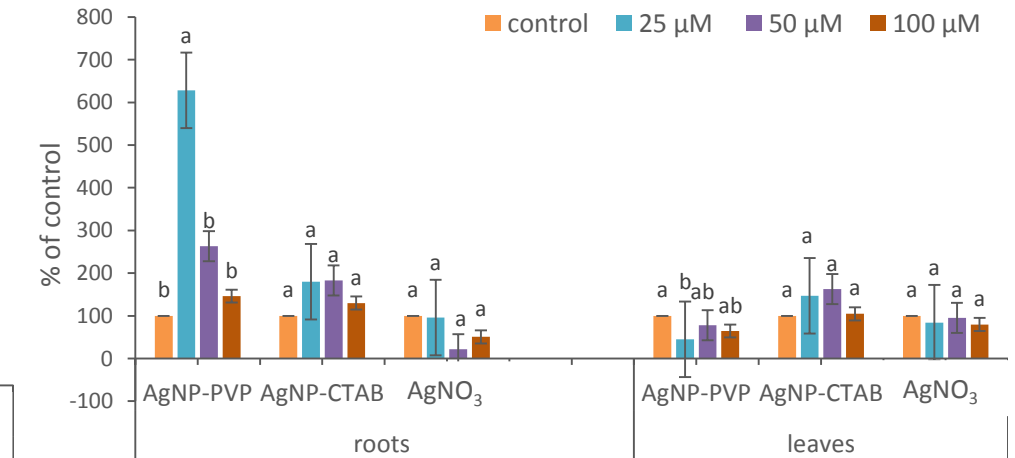


Figure 3. Activity of APX in tobacco roots and leaves after exposure to 25, 50 and 100 μM AgNPs and AgNO₃. The presented results show mean values of 6 replicates ± standard error. Values marked with different letters represent significant difference ($p \leq 0.05$) according to Duncan test.

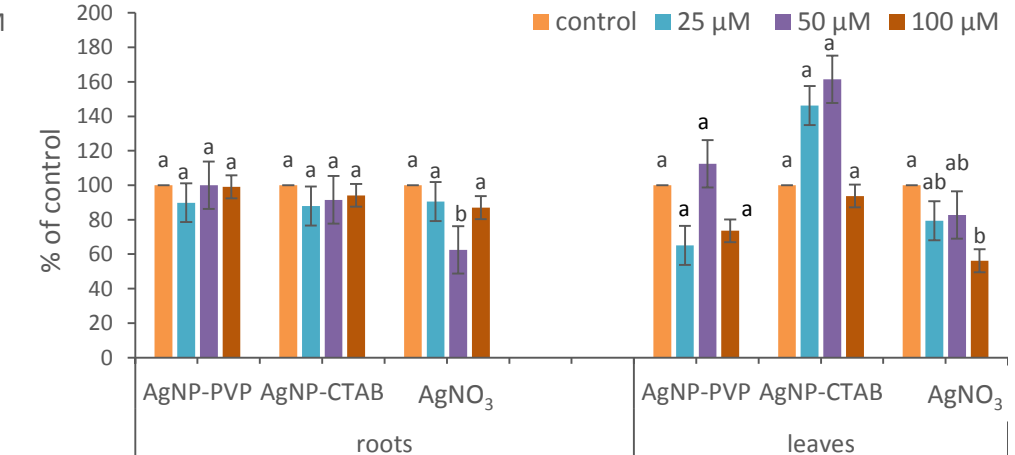


Figure 5. Activity of PPX in tobacco roots and leaves after exposure to 25, 50 and 100 μM AgNPs and AgNO₃. The presented results show mean values of 6 replicates ± standard error. Values marked with different letters represent significant difference ($p \leq 0.05$) according to Duncan test.

CONCLUSIONS

- After both types of treatments similar accumulation of Ag was obtained, which was significantly higher in roots compared to leaves
- Increase in ROS level was observed in roots after AgNP-PVP and 25 μM AgNO₃ treatment. In leaves, significant changes in ROS level compared to control were obtained after AgNO₃ exposure
- In roots, significant changes in SOD activities were observed after all treatments, while CAT activity exhibited significant increase after exposure to AgNP-CTAB; in leaves the most prominent changes in activities of SOD and PPX were observed after AgNO₃ treatment, while 100 μM AgNP-CTAB showed the highest increase in both CAT and SOD activities
- Obtained results suggest that Ag treatments induced oxidative stress, while different antioxidant enzymes were activated in roots compared to leaves depending on the form in which Ag was applied