IECPS 2024
The 3rd International Electronic Conference on Plant Sciences
15–17 January 2024 | Online

HELLENIC REPUBLIC
National and Kapodistrian University of Athens
EST. 1837

Is MAP65-1 phosphorylation related to Cr(IV) effects on the microtubules of *Arabidopsis thaliana*?

Ioannis-Dimosthenis S. Adamakis

*Section of Botany, Department of Biology, National and Kapodistrian University of Athens, Greece, * iadamaki@biol.uoa.gr*

INTRODUCTION & AIM

Chromium (Cr) is a heavy metal, occurring in both terrestrial and aquatic habitats [1]. It is found in two oxidation states, the trivalent Cr(III) and the hexavalent Cr(VI) [1]. Cr (VI) is a toxic and non-essential element for plants. When encountered at high concentrations, almost all physiological, biochemical and cellular processes of plants are negatively affected [2]. Microtubules (MTs) in particular, have been found to be prone to Cr(VI) in plant cells, and constitute a universal target of Cr(VI) toxicity [3]. MAP65-1 (the most abundant plant structural microtubule-associated protein) modulates microtubule stability since, it binds and bundles them by forming stabilizing cross-bridges between neighboring MTs. This ability is affected by its phosphorylation status, and when MAP65-1 becomes phosphorylated it becomes unbound from MTs during the cell cycle phases [4]. In the present study, the effects of Cr(VI) on MAP65-1 presence on cortical MTs of *Arabidopsis thaliana* roots and hypocotyls were investigated.

METHOD

A. thaliana lines expressing GFP:TUA, GFP:AtMAP65-1 and the line expressing the non-phosphorylatable AtMAP65-1, AtMAP65-1^{9A} (GFP:AtMAP65-1^{9A}) [5] have been used. Four-day-old seedlings were transplanted to Petri dishes with ½ MS solid medium supplemented with 100 μM potassium dichromate ($K_2Cr_2O_7$, Cr(VI) for brevity), and left to grow for 24 or 48 h. All specimens have been examined under a Nikon D-Eclipse C1 confocal laser scanning microscope (CLSM, Nikon Inc., Tokyo, Japan). Special care was taken to retain the laser beam gain equal among the different treatments. Image recording was conducted with the EZ-C1 3.20 software (Nikon Inc.) according to the manufacturer's instructions.

RESULTS & DISCUSSION

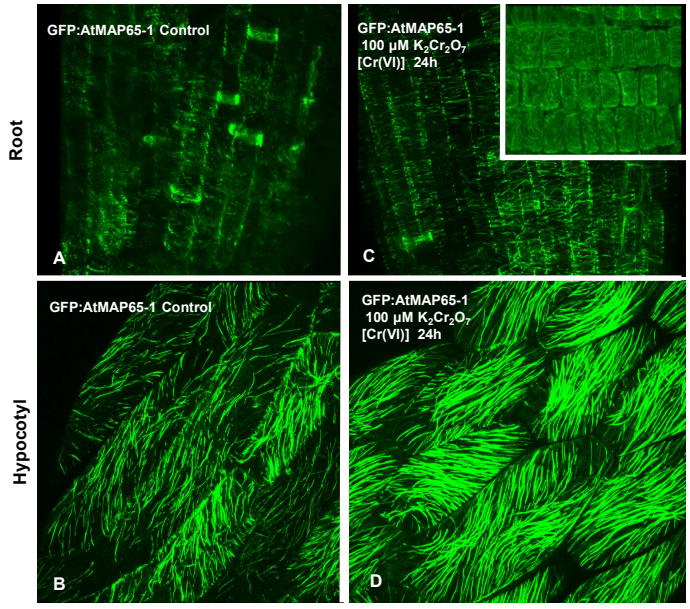

Figure 3. CLSM images of root (A,C) and hypocotyl cells (B, D) of *A. thaliana* lines expressing the non-phosphorylatable AtMAP65-1, AtMAP65-1^{9A} (GFP:AtMAP65-1^{9A}) in control (A, B) or Cr(VI) (C, D) treated seedlings. Upon Cr(VI) treatment GFP:AtMAP65-1^{9A} signal appears to be retained (C) or is intensified, especially in hypocotyl cells (D), contrast to the GFP:AtMAP65-1 signal (Figure 2). Root microtubules of this line, seemed not to be severly affected (inset in C).

CONCLUSION

Cr(VI), affected microtubules (MTs) of root and hypocotyl cells. Except MTs, AtMAP65-1 seemed to be also a target of Cr(VI) toxicity. However, Cr(VI), had no effect on the non-phosphorytable AtMAP65-1^{9A} , while MTs of this line, also seemed to be unaffected by Cr(VI). The above observations show that the influence of Cr(VI) on MTs is related to MAP65-1 phosphorylation.

https://iecps2024.sciforum.net/

RESULTS & DISCUSSION

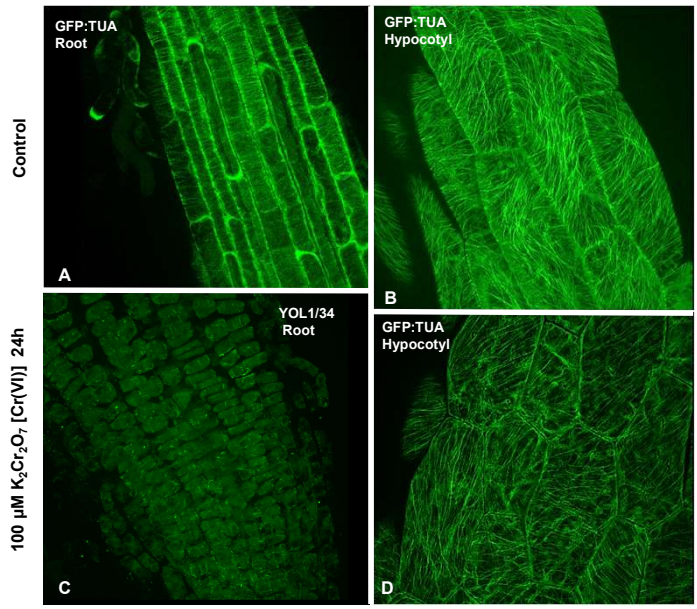

Figure 1. CLSM images of root (A,C) and hypocotyl cells (B, D) of *A. thaliana* lines expressing GFP:TUA (A, B, D) or after YOL1/34 immunolabeling (C), in control (A, B) or Cr(VI) (C, D) treated seedlings. Upon Cr(VI) treatment microtulubes appear fragmented (C), especially in root cells, or GFP:TUA signal is reduced (D).

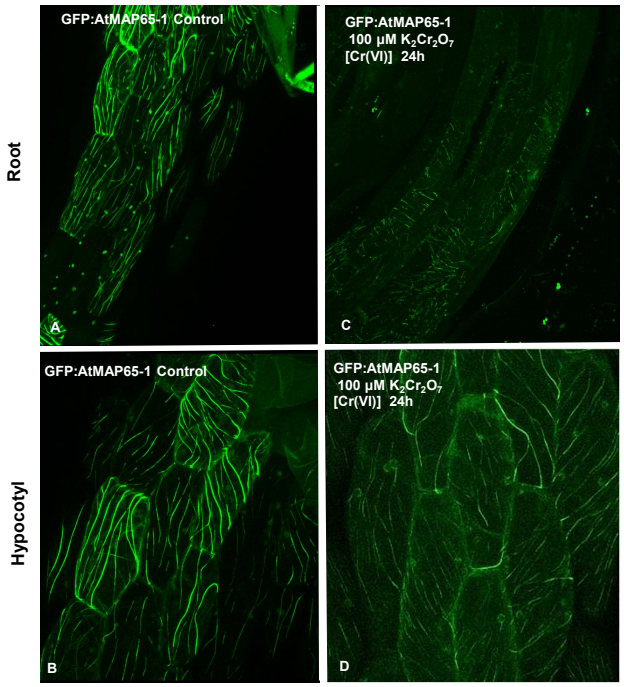

Figure 2. CLSM images of root (A,C) and hypocotyl cells (B, D) of *A. thaliana* lines expressing AtMAP65-1 (GFP:AtMAP65-1) in control (A, B) or after Cr(VI) (C, D) treated seedlings. Upon Cr(VI) treatment GFP:AtMAP65-1 signal appears reduced both in root (C) and hypocotyl cells (D).

REFERENCES

[1]. Dotaniya, M. L., et al. "Chromium pollution: a threat to environment-a review." *Agricultural Reviews* 35.2 (2014): 153-157.
[2]. Ali, Sajad, et al. "Chromium toxicity in plants: signaling, mitigation, and future perspectives." *Plants* 12.7 (2023): 1502.
[3]. Eleftheriou, P. Eleftherios., et al. "Hexavalent chromium-induced differential disruption of cortical microtubules in some Fabaceae species is correlated with acetylation of α-tubulin." *Protoplasma* 253 (2016): 531-542.
[4]. Mao, Tonglin, et al. "Two microtubule-associated proteins of the Arabidopsis MAP65 family function differently on microtubules." *Plant Physiology* 138.2 (2005): 654-662.
[5]. Smertenko, Andrei P., et al. "Control of the AtMAP65-1 interaction with microtubules through the cell cycle." *Journal of cell science* 119.15 (2006): 3227-3237.