Time-dependent modulation of genetic stability and phytochemical profiles in chrysanthemum leaves and inflorescences by iron oxide nanoparticles and IAA auxin

Dariusz Kulus[a], Alicja Tymoszuk[a], Mateusz Cichorek[b], Katarzyna Gościnna[c], Magdalena Osial[d]

[a]*Laboratory of Horticulture and Landscape Architecture, Department of Biotechnology, Faculty of Agriculture and Biotechnology, Bydgoszcz University of Science and Technology, Bernardyńska 6, 85-029 Bydgoszcz, Poland*
[b]*Department of Plant Physiology, Genetics and Biotechnology, Faculty of Biology and Biotechnology, University of Warmia and Mazury in Olsztyn, Oczapowskiego 1a, 10-719 Olsztyn, Poland*
[c]*Department of Agronomy and Food Technology, Faculty of Agriculture and Biotechnology, Bydgoszcz University of Science and Technology, Kaliskiego 7, 85-796 Bydgoszcz, Poland*
[d]*Institute of Fundamental Technological Research, Polish Academy of Sciences, Pawińskiego 5b, 02-106 Warsaw, Poland*

The increasing role of nanoparticles (NPs) is revolutionizing horticultural practices by enhancing plant growth, improving nutrient absorption, and enabling precise delivery of agrochemicals. However, little is known about the use of NPs in the production of synthetic seeds - a propagation technique particularly valuable for seedless species, such as chrysanthemum. This research studied the impact of pure iron oxide nanoparticles (Fe_3O_4 NPs), citrate-stabilized iron oxide nanoparticles (Fe_3O_4CA NPs), and indole-3-acetic acid (IAA) on the genetic stability and metabolic activity of *Chrysanthemum* × *morifolium* (Ramat.) Hemsl. plants obtained from synthetic seeds. For this purpose, axillary buds of chrysanthemum 'Richmond' were embedded in 3% calcium alginate supplemented with NPs and IAA, either singularly or in combination. Next, the synthetic seeds were stored at 4°C in the dark (for eight weeks) on an agar-water medium and then transferred to room temperature for 30 or 60 days. Next, the germinated seeds were transplanted to the greenhouse until the plants were fully flowering. The content of total polyphenols was determined in the leaves and inflorescences of the plants. Moreover, the content of anthocyanins was measured in the inflorescences. RAPD markers were used to assess the genetic stability of plants. It was found that NPs and IAA significantly affected the content of total polyphenols (TCP) in the leaves of chrysanthemum. Most treatments stimulated the accumulation of these compounds but in a time-dependent manner. No decline in the value of this parameter was reported compared with the untreated control. Conversely, Fe_3O_4NPs and IAA + Fe_3O_4CA NPs stimulated the biosynthesis of polyphenols and anthocyanins in the inflorescences after 30 days of treatment, however, a decline in the content of these compounds was reported after 60 days in most experimental objects, except for Fe_3O_4CA NPs and IAA + Fe_3O_4CA NPs. The inflorescences of plants treated with nanoparticles usually exhibited a larger diameter than the control, but only after a shorter exposure to the analyzed factors. In contrast, prolonged treatment resulted in the opposite effect. The genetic uniformity of the plants was confirmed with 2160 RAPD markers. This study expands the knowledge of the application of nanoparticles in plant biotechnology, particularly synthetic seeds.