Dynamics of glutathione during *Cyclamen persicum* somatic embryogenesis

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Cyclamen persicum (Myrsinaceae), a tuberous ornamental pot plant, is propagated by seeds which requires manual pollination, and results in variability in some cultivar groups. Somatic embryogenesis in Cyclamen could become an alternative vegetative propagation pathway. However, physiological disorders, a low desiccation tolerance, a non-synchronized development of somatic embryos and the lack of a maturation step, still limit the use of somatic embryogenesis for commercial scale propagation. Several studies have reported on the crucial role of a high redox state of glutathione in the early stages of somatic embryo development. Moreover, some authors suggest that the formation of abnormal embryos can be correlated with low levels of GSSG in the late maturation phase. The endogenous glutathione redox state is defined as the ratio of the concentration of the reduced form (GSH) and sum of the concentrations of the oxidized and the reduced form (GSH + GSSG) (BELMONTE ET AL. 2006). In order to better understand the correlation between somatic embryogenesis and glutathione redox state, the aims of this study were to quantify the contents of endogenous GSH, GSSG and cysteine (one of the GSH precursors) during differentiation of somatic embryos of *Cyclamen persicum*.
Tissue was collected after the transfer of embryogenic cells to the plant growth regulator (PGR) free differentiation medium on days 0, 1, 2, 3, 4, 5, 8, 11, 14, 17 and 21. Samples were taken from cultures in liquid medium and cells cultured on solid medium, with three biological replicates in two repetitions of the experiment each. GSH, GSSG and cysteine were extracted using 5% metaphosphoric acid (MPA) and determined by HPLC coupled to ESI-TOF MS following the method described by RELLÁN-ÁLVAREZ ET AL. (2006) with modifications. After 24 hours a peak in GSH and cysteine concentrations was observed. Thereafter, the content of cystein remained stable, while GSH increased slightly over time. GSSG was detected in lower concentrations than GSH and was observed to increase until day 8. However, the results showed a high variation between the repetitions of the experiment which correlated with the variation in the formation of somatic embryos in different experiments. Experiments are in progress to supply GSH or GSSG in different stages of embryo development to support a normal development of SE.

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