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Poster Abstract – A.76

MUTATIONAL EFFECTS OF SYNTHETIC CYTOKININS DIPHENYLUREA-DERIVED ON PLANTS REGENERATED BY SOMATIC EMBRYOGENESIS

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Style-stigma explants from calamondin (*Citrus madurensis* Lour.) were cultured in media supplemented with three synthetic phenylurea derivatives, N-(2-chloro-4-pyridyl)-N-phenylurea (4-CPPU), N-phenyl-N'-benzothiazol-6-ylurea (PBU) and N,N'-bis-(2,3-methilendioxyphenyl)urea (2,3-MDPU). A cytokinin adenine-derived N^6-benzylaminopurine (BAP) supplemented medium and an hormone-free (HF) medium were used as negative controls. Somatic embryos were regenerated in all the conditions used and plantlets were obtained. Among the phenylurea analogous compounds, the 2,3-MDPU showed the highest embryogenic potential (68.0, 49.3 and 43.3% of responsive explants in 2,3 MDPU, PBU and 4-CPPU, respectively) and all of them showed a significantly higher percentage of responsive explants than that obtained with BAP and HF conditions (33.7 and 10.0, respectively). In order to verify the presence of somaclonal variability of the regenerants, twenty-seven somaclones, coming from different embryogenic events, were randomly selected from each different culture condition and analyzed by using inter-simple sequence repeat (ISSR) and random amplified polymorphic DNA (RAPD) analyses. We observed that plantlets regenerated in media supplemented with 2,3-MDPU and PBU gave 3.7% of somaclonal mutants, whereas plantlets from medium supplemented with 4-CPPU gave 7.2% of mutants. No somaclonal variability was observed when plantlets were regenerated in BAP or HF medium. Our results suggest that, although diphenylurea derivatives show an higher embryogenic potential as compared to BAP, they induce higher levels of somaclonal variability. This finding should be taken into consideration when new protocols for clonal propagation have to be set up. On the other side, the regeneration of somatic embryos in presence of diphenylurea derivatives, that induce somaclonal variability, can contribute to plant genetic improvement programs.