

CYTOLOGICAL AND FLOW CYTOMETRIC CHARACTERIZATION OF AN *ARUNDO DONAX* L. MUTANT

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Arundo donax L. (common name Giant Reed) is a rhizomatous perennial plant native to Asia and the Great Middle East but now diffused all over the world. The interest in this species is increasing due to its high biomass production, resistance to biotic and abiotic stresses, and great adaptability to marginal land, and is therefore a promising energy crop. *A. donax* is strictly sterile and therefore cross-breeding strategies cannot be applied. To overcome this constraint and to generate genetic variability, physical mutagenesis based on gamma-ray irradiation was formerly applied starting from an Emilia-Romagna ecotype of Giant Reed. Among c. 900 clones, one ('Unibo1') was selected showing high tillering capacity and easy propagation. In this study we characterized 'Unibo1' in terms of chromosome number and DNA content. *A. donax* genome is characterized by a very high number of chromosomes (ranging from 102-110, caused by high and still unknown polyploidy level) of small size (total DNA amount $2C = 5,46$ pg), making its cytogenetic characterization a daunting task. An optimized protocol was developed for obtaining good metaphases spreads from root tip cells. Nuclei in suspension were also isolated from the same organ for flow cytometry DNA content evaluation. Root tips were obtained from sprouts or cane nodes grown under hydroponic condition, and mitotic cells were at first synchronized with 8 hydroxyquinoline, then tissues were fixed with Carnoy and enzymatically digested using a selected enzymatic solution. From a number of metaphase plates, twenty ones were

analyzed and the chromosome number was scored. Differences among chromosome counts were observed between 'Unibo1' and wild type, also enforced by flow cytometry analysis of DNA content, with 'Unibo1' showing $2n = 102 \pm 2$ chromosomes, while $2n = 108 \pm 2$ chromosomes were counted in wild type. Our work provided a protocol to streamline further cytogenetic investigations in *A. donax* and shed light on the effects of physical mutagenesis on plant genomes.