

Evaluation of Nuclear DNA Content / Ploidy Level of the Rootstocks of Genus Prunus Used for Plum Cultivation

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For breeding purposes, evaluation of nuclear DNA content was performed in order to determine ploidy level of rootstocks of genus Prunus for plums. In the first stage of the research, the conditions of flow cytometry analysis (with application of propidium iodide for DNA staining) (FCM/PI) were optimized: type of buffer for nuclei extraction and incubation time as well as selection of internal standards (genotypes with known nuclear DNA content). Repeatable results and relatively good-quality histograms were obtained using Partec extraction buffer with 1 % PVP addition, with incubation time longer than 50 min. The best internal standard for FCM analysis of Prunus rootstocks with a nuclear DNA content from about 0.5 to 1.4 pg proved to be Solanum lycopersicum (2C = 1.96 pg), which peaks 2C and 4C did not coincide with the peaks of the genotypes tested, but overlapped partially with the peaks of Prunus genotypes with larger genomes tested. For rootstock genotypes with a nuclear DNA contents from 1.8 to 2.3 pg, Glycine max (2.91 pg) or Zea mays (2C = 5.44 pg) were selected as the internal standards. In the second stage, the optimized method and well-chosen internal standards allowed to assess with high precision the nuclear DNA content / ploidy level of Prunus genotypes of the rootstocks for plums. For the standard Prunus genotypes of the known chromosome number, 2C DNA values was 0.68 pg for diploid P. cerasifera var. divaricata Led. 'Anna', the value of 1.18 pg for triploid 'Gisela 3' (P. cerasus 'Schattenmorelle' × P. canescens) and 2.16 pg for hexaploid P. domestica 'Eruni'. In four rootstock genotypes ('Ferciana Ishtara', 'VVA-1', 'Druzba' and 'GF 667') 2C values ranged

from 0.61 to 0.67 pg indicating their diploid chromosome number. Three rootstocks were identified as hexaploids ('St. Julien A', 'Pixy' and 'GF 655/2') owing to their DNA contents from 2.07 to 2.23 pg. The rootstock 'P 8.13' was considered as pentaploid due to their 2C value of 1.64 pg which was approximately two and a half times more than in diploid Prunus sp. (2C DNA = 0.66 pg). The ploidy level of genotypes of plum rootstocks was discussed in relation to morphological and agronomical traits.

Keywords: genome size, rootstock, Prunus domestica, flow cytometry.

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