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초록 제목: (국문) 엽록소 형광의 분석을 통한 형질전환체의 선별

(영문) Screening of transgenic tobacco plants by using a chloroplyll fluorescence assay

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Efficiency of chlorophyll (Chl) fluorescence assay was tested for screening transgenic tobacco plants (Nicotiana tabacum cv. Xanthi) carrying a selection marker gene, a kanamycin(Km) resistant gene(nptII) or a herbicide phosphinothricin(PPT) resistant gene(bar) coding PPT-acetyl transferase(PAT). By the treatment of 200 mg/L Km to leaf discs from wild-type plants. (Fv)m(Fm minus Fo) was decreased, and Fo was increased significantly. Variations among samples could be reduced by using (Fv)m/Fo ratio as a screening parameter. By using this parameter, Km-treated samples could be distinguished from the control within 4 days after the treatment. After cocultivation of tobacco plants with Agrobacteria containing nptII gene, seventy-five shoots were selected in 200 mg/L Km containing media and leaf discs were taken from them for Chl fluorescence assay. Among them, 40 plants were Km sensitive ('escaped'). All the sensitive plants did not contain nptII gene in their chromosome as shown by polymerase chain reaction amplification of this gene and did not show any neomycin phosphotransferase activities also. All the resistant plants were proved as positive by these two tests. By the treatment of PPT under low light intensity (30  $\mu$ mol·m<sup>-2</sup>·s<sup>-1</sup>). Fm/Fo value from wild type tobacco leaf discs were decreased in half by the treatment of 100 mg/L PPT for 12 hrs. However, similar effects could be observed only 1-3 hrs after the treatment under high light intensity(60 µmol· m<sup>-2</sup>·s<sup>-1</sup>). All the transformed and screened plants showed PAT activities and did not show similar change in fluorescence induction parameters. Within 48 hrs of the treatment, symptoms of PPT toxicity in the leaf discs were hardly detectable by naked eyes.

These results prove the efficiency of this non-destructive Chl fluorescence assay in screening transgenic tobacco plants carrying selection marker genes. Especially *bar* gene containing plants could be screened very quickly by employing Chl fluorescence assay under high light condition.