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Somaclonal Variation and RAPD Analysis in Schisandra chinensis

V. R. Anbazhagan & J.I. Park*

Lab of Forest Genetic Resources, College of Agriculture, Life and Environment Science
Chungbuk National University, 361-763 Republic of Korea

Objectives

Schisandra chinensis (Turcz) Baill., is native to east Asian countries and possess high pharmaceutical value (Krussmann,1978). The present investigation was carried out to develop a protocol for mass propagation of plants through somatic embryogenesis via suspension culture from cotyledonary somatic embryos and determination of somaclonal variation (Larkin and Scowcroft, 1986) in *S. chinensis* by adopting RAPD technique.

Materials and methods

Explants and medium: Cotyledonary somatic embryos, Callus induction-MS (Murashige and Skoog, 1962) medium with 2,4-D $(4.52\mu\text{M})$ and L-glutamine (1.71.mM).

Somatic embryo development and germination - PGR-free MS medium

RAPD analysis: Genomic DNA isolation from leaves of somatic embryo derived plantlets (wizard® genomic DNA purification kit ,Promega, USA), Primer - 5'GAC GCC ACA C3' (20 pmol), dNTPs (0.25mM), Taq DNA polymerase (2.5U),10X reaction buffer, Denaturation temperature-94°C for 4 min, Annealing temperature-55°C, extension temperature-72°C, Reaction cycle-35, PCR were carried out in-Biometra UNO-Thermoblock (U.K). Amplification products were separated on 1.2% agarose (Sigma, USA) gel in 1X TAE buffer (PH 8.4), stained with ethidium bromide and photographed under UV light using Kodak 1D program.

Results and Discussion

Somatic embryos were developed from suspension culture. Induction of embryogenic callus occurred on MS semi-solid medium with 2,4-D (4.52 μ M) and L-glutamine (1.71.mM). Somatic embryos were developed on PGR-free MS liquid medium. The germination of cotyledonary somatic embryos occurred PGR-free MS semi-solid and liquid medium. RAPD analysis confirmed that genetic fidelity of somatic embryo derived plant differed with field grown control plant, thus proving somaclonal variants.

References

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^{*} Corresponding author: Jae In Park, Tel: 043-261-2535, E-mail: jipark@chungbuk.ac.kr