(05-2-14)

Histological analysis by developmental stages of zygotic embryos and somatic embryogenesis in *Pinus rigida* × *P. taeda*

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Objectives

We have tried to develop an effective *in vitro* propagation system of *Pinus rigida* × *P. taeda* through somatic embryogenesis and to examine the potential of zygotic embryos at different developmental stages in the initiation of embryogenic tissue by histological analysis.

Materials and Methods

1. Material

Plant - immature seeds of *Pinus rigida* × *P. taeda*, embryogenic tissue

2. Methods

Immature seeds collected during the period from June 11 to July 30 were cultured. And the seeds were fixed weekly to observe the developmental stages of zygotic embryo. Embryogenic tissues were initiated on the *Pinus taeda* basal medium (P6) supplemented with 2,4-D and BA. Somatic embryos were matured on the medium containing ABA and a high concentration of gellan gum. Matured somatic embryos germinated on the medium without plant growth regulators(PGRs).

Results and Discussion

Out of over 3,400 seeds cultured, 5 embryogenic tissue lines were obtained (initiation rate of 0.14% totally). Embryogenic tissues were extruded from the micropylar end of megagametophyte and had translucent and mucilaginous appearance. The highest tissue initiation (0.55%) was observed with the seeds collected on July 3. Histological analysis revealed that those seeds collected on July 3 were at the transition from proembryos to precotyledonary stages with majority showing the former stage. In the presence of 80-150 M ABA, somatic embryos were matured on the media containing a high concentration of gellan gum (1.0%). Matured somatic embryos germinated on the medium with low concentration of gelrite (0.2%) and activated carbon (0.05%).

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