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Induction of Somatic Embryos from Cotyledonary and Leaf Tissues of Tea [*Camellia sinensis* L.]

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Abstract

This study was carried out to achieve efficient somatic embryogenesis from cotyledonary and leaf tissues of tea. Small cotyledon segments were taken from sterilized cotyledon (proximal to zygotic embryonic zone) of four tea cultivars (TRI 2043, TRI 2025, TRI 777 and DT1). These explants were placed on MS basal medium with 2.0 mg L⁻¹ BAP and 0.2 mg L⁻¹ NAA to express their embryogenic capacity. The results revealed that the cotyledon explants of high yielding cultivars, DT1 and TRI 2025 performed better in embryogenic potential. As a preliminary study to induce somatic embryos from cotyledonary petioles, small petiole segments excised from *in vitro* seedlings of TRI 2025 were placed on the above medium. Results showed that somatic embryos were directly formed on cotyledonary petioles. It has potential to produce a large numbers of propagules from hybrid seeds. Further work was aimed to induce somatic embryos indirectly from leaf explants. Leaf segments excised from *in vitro* shoots of TRI 2043 were cultured on MS basal medium with 2 mg L⁻¹ BAP and 3 mg L⁻¹ NAA. After 16 weeks of culture, primary calli were cultured on MS medium contained some selected combinations of growth regulators (BAP, kinetin, NAA, GA₃ and adenine sulphate) to produce somatic embryos. Results showed that formations of mature somatic embryos (11.7% and 10%) were high in MS medium with BAP (1 mg L⁻¹) and NAA (0.1 mg L⁻¹) in combination with GA₃ or adenine sulphate at 0.1 mg L⁻¹ respectively. This protocol will be useful to achieve new somatic variants from seedling explants and also to use in transformation technique.

1. Introduction

In Sri Lanka, tea is still high priority plantation crop and still makes a significant contribution to the economy of our nation. It is grown under various climate conditions and is faced with pest and disease problems. For commercial planting of tea, newly elite cultivars within the existing genotypes have been mainly developed by Tea Research Institute, Sri Lanka through conventional breeding methods. High yielding tea cultivars (TRI 2024, TRI 2025 and DT1) gave low values (1 - 6%) for seed set after hand pollination with several other cultivars (Anandappa *et al.*, 1988).

Germination of hybrid materials plays an important role even in a vegetatively propagated crop such as tea. Dadd (1928) suggested that cracking the seed gives easy access of water and oxygen to the embryos and results in a higher percentage of germination in a shorter time. Crossing materials may fail to germinate in the nursery beds (Kato, 1989). Therefore, it is necessary to obtain efficient germination of hybrid seeds under *in vitro* conditions to help tea breeders. Further, vigorous and abundance plants require for efficient selection process at initial phase of plant improvement. Propagation of tea by somatic embryogenesis has the potential to produce a large numbers of propagules from cotyledonary tissues.