

Establishment of *in vitro* culture to produce friable callus from leaf of *Camellia sinensis* (L.)

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Abstract: Attempts were made to produce *in vitro* callus from tea (*Camellia sinensis* L.) leaves. Unfolded leaves were collected and surface sterilized in various concentrations of CloroxTM (15% -75%) in combination with various exposure times (15-60 min) to obtain optimal concentration and exposure time for sterilization of field grown leaves. Results indicated that 50% and 58% aseptic cultures were achieved in 60% and 75% solutions of CloroxTM in a soaking period of 30 min respectively. Further, sterilized mature zygotic embryos were cultured on MS media containing 1 to 10 mg/l BAP in combination with 0.1 mg/l NAA to obtain the suitable concentration of BAP for the establishment of *in vitro* micro shoots. The result showed that 5 mg/l concentration of BAP would be suitable for the initiation of *in vitro* micro shoot cultures. At 12th week, plantlets regenerated in BAP at 5 mg/l were subcultured in the presence of 3 mg/l BAP and 0.1 mg/l NAA. Multiplication rate of first two subcultures was 3.6 ± 0.2 . Further leaf segments at 2nd, 3rd and 4th subculture periods were cultured on callus medium to determine the competence of friable callus initiation on leaves of newly establishing *in vitro* micro shoots. Results revealed that initiation of friable callus was fairly better on leaves obtained at 4th subculture among the tested treatments. Moreover, *in vitro* and field grown leaves were compared on the efficiency of callus initiation. A significant high frequency of callus induction (79.2%) was achieved from *in vitro* leaf explants, which were collected at 5th subculture.

Key words: Friable callus, leaves, *in vitro* aseptic culture, micro shoots.