

PROBLEMS ASSOCIATED WITH ACCLIMATIZATION OF *IN VITRO* REGENERATED PLANTLETS

P. Kumari Jayasree

Rubber Research Institute of India, Kottayam-686 009, Kerala, India

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Tissue culture derived plantlets are usually very delicate, and have weak root system and exhibit high mortality rates upon their transfer from *in vitro* culture vessel to *ex vitro* conditions. These plantlets therefore need to undergo a gradual adaptation from *in vitro* to natural conditions through a process of hardening / acclimatization. Controlled hardening procedures are therefore necessary to ensure high survival rates of plants and to correct abnormalities. Under such conditions, plants rapidly get converted from a heterotrophic state to an autotrophic mode along with the development of a fully functional root system and better control on stomatal and cuticular transpiration. The present review describes the characteristic variations commonly observed in *in vitro* developed plantlets like limited photosynthesis, reduced epicuticular wax, dysfunctional stomata, etc. Moreover, some methods for the successful acclimatization of *in vitro* generated plantlets are also discussed.

Keywords: Acclimatization, Cuticular transpiration, Epicuticular wax, Hardening, Stomatal transpiration

INTRODUCTION

For many plant species, considerable research has been conducted to optimize the nutrient medium and culture conditions for different stages of plant development. However, the advantage of any *in vitro* culture system may be fully accomplished only by the successful transfer of plantlets from *in vitro* to the *ex vitro* conditions and survival thereafter. A large number of *in vitro* grown plants do not survive upon transfer from culture vessels to greenhouse or field environments, which still remain as a major bottleneck in many plants, including rubber. Compared to *in vitro* conditions, greenhouse and field possess highly variable

humidity, higher light intensity and favourable atmosphere which are stressful to the *in vitro* plants (Hazarika, 2003). Usually, *in vitro* regenerated plants are much smaller, grown on very precise conditions of nutrient media containing exogenous sugars and plant growth regulators and under higher relative humidity (100%). These culture conditions contribute to abnormalities in morphology, anatomy and physiology of plants (Desjardins, 1995; Kozai and Smith, 1995). As a result, many plants wilt or dry up rapidly when transferred directly to glasshouse or field. A period of acclimatization to the *ex vitro* environment is necessary for the successful establishment and survival of plantlets (Chandra *et al.*,