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OPTIMIZATION OF CONDITIONS FOR *IN VITRO* MICROGRAFTING IN RUBBER (*HEVEA BRASILIENSIS*)

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The conditions for successful *in vitro* micrografting such as the nature of cut and type of support, age and size of stock and scion, the physical nature of the medium and effect of antioxidants were standardised for *Hevea brasiliensis*. The micrografting was carried out using zygotic embryo derived plantlets raised *in vitro*. *Ex vitro* micrografting was standardised using polybag grown seedlings as root stocks and scions from glass house grown plants. Maximum success rate was obtained with saddle type of cut with parafilm as the support. Thirty day-old stocks and 21 day-old scions had considerably enhanced the success rate. Scions of length 1.5 cm were ideal for micrografting. Maximum success rate was observed when 60 g/l sucrose was included in the culture medium. Application of the antioxidant ascorbic acid, at a concentration of 100 mg/l to the cut surface of the explants, prevented browning as well as in increased the survival rate. The technique is useful for combining genetically and physiologically comparable rootstocks and scion of elite clones produced by genetic engineering.

Key words: *Hevea brasiliensis*, *In vitro* micrografting, Micropropagation, Zygotic embryos.

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INTRODUCTION

The cultivated clones of *Hevea brasiliensis* (Muell. Arg.) are commonly propagated through budgrafting. Although for a particular clone the scions selected for budgrafting are uniform, the heterogeneity of seedling root stocks lead to stock-scion interactions resulting in considerable variation among the population of a single clone. The need to have many uniform individuals of a selected genotype led to the development of *in vitro* techniques as a means of vegetative propagation. Micropropagation of *Hevea* through shoot tip culture (Asokan *et al.*, 1988; Seneviratne *et al.*, 1993; Seneviratne and Flegman, 1996) and somatic embryogenesis (Etienne *et al.*, 1993a; Carron *et al.*, 1995; Jayasree *et al.*, 1999) have been successful. However, these technologies have not been commercially adopted due to problems in root induction, long time lag and high mortality during hardening and the higher cost compared to conventional propagation by budgrafting.

Micrografting consists of grafting an apex taken from one plant on to another decapitated young plant (Jonard, 1986). This technique has been successfully used in many horticultural plants to develop virus free clones ((Murashige *et al.*, 1972; Navarro, 1988) and to detect graft incompatibilities at an early stage (Herrero, 1951; Mosse, 1962). This method has been proved to be effective as a means of micropropagation of tea (Prakash *et al.*, 1999) and clone rejuvenation of *Hevea* (Perrin *et al.*, 1994) and pine (Travan and David, 1985). Micrografting has been carried out to eliminate viral diseases on different species of lemon tree (Murashige *et al.*, 1972; Roistacher and Kitto, 1977) and fruit trees like cherry (Ozzambak and Schmidt, 1991), kiwi fruit (Ke *et al.*, 1993) and apple (Richardson *et al.*, 1996). In cocoa, where the rooting of *in vitro* grown shoots and regeneration of somatic embryos were problems, this technique was used as a successful alternative for plantlet production (Aquilar *et al.*, 1992). Since 1953,