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Surface sterilization and culture establishment of Araucaria (*Araucaria columnaris*) explants excised from shoot tips of secondary shoot branches

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The current study was carried out to identify an *in vitro* micro propagation protocol for mass scale production of this species as conventional propagation through seeds is slow and inadequate while difficulties in root formation when using cuttings and production of plagiotropic plants if lateral shoots used. The importance of this plant is for reforestation, timber production and as an ornamental.

Shoot tips derived from secondary branches were excised from four year old tree and surface sterilized using three different concentrations of Clorox (5.25% Sodium Hypochlorite –NaOCl) (10%, 15% and 20%) with three different exposure time durations (10, 15 and 20 minutes) with 70% ethanol for two minutes. Explants were established in modified MS (Murashige and Skoog's medium, 1962), WPM: McCown's Woody Plant (Lloyd and Mc Cown, 1981), Anderson (Anderson, W.C. 1984) and B5 (Gamberg, 1968) media with and with out using 1g/L activated charcoal. Experiments were replicated 20 times and analyzed using SAS computer software. Contaminations, browning and growth performances were recorded in weekly intervals and data were analyzed using SAS computer software. Results revealed that 10% NaOCl for 20 minutes exposure time with 70% alcohol for two minutes exposure time recorded the highest survival percentages where contaminations reduced to 30% without deaths due to chemical burning. Modified WPM with 1g/L activated charcoal, was the best establishment medium for shoot tips with minimum browning occurred in the medium as well as on explants with a height increment (1 cm/ explant) of explants. Fungal contaminations were controlled by subsequent sub culturing to the same medium. It can be concluded that Araucaria shoot tips detached from four year old tree can be successfully surface sterilized using commercially available Clorox and ethanol solutions and they can be established *in vitro* in modified WPM medium containing 1 g/L activated charcoal.

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