

## **Asymbiotic Seed Germination and *in vitro* Seedling Development of *Paphiopedilum liemianum* Fowlie, an Endangered Terrestrial Orchid in Northern Sumatra, Indonesia**

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### **ABSTRACT**

*Paphiopedilum liemianum* Fowlie, is a terrestrial orchid species and endemic in Northern Sumatra, Indonesia. However, due to a deep dormancy that the seed display at dispersion and the difficulty to obtain uniform plant in a short time period, micropropagation may be a feasible alternative. Micropropagation by *in vitro* seed germination techniques have been applied to the conservation of endangered orchid. Four months old seeds of *P. liemianum* germinated on five different basal media. All media were supplemented with 2.5  $\mu\text{M}$   $\alpha$ -naphthaleneacetic acid (NAA) and cultures were incubated in the dark for 4 weeks followed by protocorm development at condition a 16/8 h L/D photoperiod. Germination percentage was 78.8% in Vacin and Went (VW) medium were significantly higher than other basal media. To evaluated the effect of organic nutrient additives on seed germination and protocorm development, the seed were cultured on VW medium amended with different of organic nutrient. Additives, especially 10% Coconut Water (CW) to VW medium improved the protocorm development well, with 33.3% the protocorm development to stage 5 (seedling). The seedlings were cultured on VW medium supplemented with different concentrations (0.0, 1.0, 2.0, 3.0 and 4.0  $\mu\text{M}$ ) of thidiazuron (TDZ). Healthy plantlets with developed leaves and roots were planted in pots with sphagnum moss and grown under *ex vitro* condition and the result was 76% survival rate after 4 weeks.

**Key words:** Coconut water, *in vitro* seed germination, seedling development, thidiazuron, vacin and went medium

### **INTRODUCTION**

*Paphiopedilum* is a genus of the flowering plant family Orchidaceae, well known as Slipper Orchids, is characterized by resemblance of the pouch-shaped lip to a lady's slipper (Lin *et al.*, 2000; Chen *et al.*, 2004). This genus is terrestrial orchids with about 70 species that are native to South and South East Asia and a distribution that extends from the ASEAN region up to Papua New Guinea (Teob, 1989). Their flowers are variety of shapes, size, colors and the most popular commercially available orchids among the Orchidaceae family and a highly valued ornamental plant (Cribb, 1998; Ng and Saleh, 2011; Hong *et al.*, 2008).

Although, *Paphiopedilum* produce abundant seeds, this orchid is relatively rare in the wild due to the absence of endosperm in their seed (Arditti and Ernst, 1993; Pierik *et al.*, 1988; Long *et al.*, 2010). In nature, the *Paphiopedilum* are endangered population because of destruction and very