Protocol development for the sterilization technique and germination process for the *Aponogeton natans* seeds

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Aponogeton natans, *A. crispus*, *A. rigidifolius* and *A. jacobsonii* are *Aponogeton* species native to Sri Lanka. As the industry has a high demand for these species for aqua-scaping, vegetative propagation method could not achieve the prevailing market demand. Hence it is very important to develop proper micro-propagation techniques. *A. natans* seeds were selected as ex-plant and were thoroughly washed with liquid soap and then with 70% ethanol for 1 minute. Then the seeds were washed thrice with distilled water taken as the control (C) and 8%, 10% 15% NaOCl (Clorox) solution were taken as T1, T2 and T3 respectively. Seeds which had undergone all treatments were washed for 10 minutes with Distilled water, thrice. The seeds were cultured in Murashige and Skoog (MS) medium with some modifications: without sugar (T1), 15g/L Sucrose (T2) and MS medium with no modification (T3) and sterilized distilled water (C) as the control. After selecting the best medium, Ms Medium with Different growth hormone concentrations were tested for Benzyl Adenine Purine (BAP) 1 mg/L and Indole Butyric Acid (IBA) 0.5 mg/L (T_1), BAP 2 mg/L and IBA 0.5 mg/L (T_2), BAP 3 mg/L and IBA 0.5 mg/L (T_3) and BAP 4mg/L and IBA 0.5 mg/L (T_4) were taken as the treatments and the medium without hormone used as the control (C). All the treatment had ten replicates and each treatment consisted of one seed. All the experiments conducted separately. The results showed that T2 obtained the highest survival percentage (91.0±1.0%) in sterilization experiment and it was significantly different with other treatments. When choosing the best medium T3 obtained the highest survival (78.7±0.7%) which used only MS medium. When selecting best hormone combination T2 showed the highest survival percentage of 89.3±1.2% which was significantly different among all treatments. According to the results obtained the seeds of *A. natans* gave high percentage of germination by sterilizing with 10% NaOCl for ten minutes and better growth in MS medium with 2mg/L BAP and 0.5 mg/L IBA hormone combination.

Keywords: *Aponogeton natans*, germination, micro-propagation, seed, sterilization

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