Thesis Title

Preservation of Some Thai Native Orchid Species by Synthetic Seed Technique

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Abstract

Protocorms size 1 ± 0.2 mm of Doritis pulcherrima Lindl. and Dendrobium cariniferum Rchb. f. were precultured onto modified VW (1949) medium comprising 0.058, 0.3 and 0.6 M sucrose for 2 days, then subsequently encapsulated in calcium alginate beads to form synthetic seeds. The results showed that the non - dehydrated synthetic seeds of both species had significant higher germination percentages i.e. 85.00 - 97.50 and 80.00 - 95.00 per cent respectively than that obtained from the 0.9 M sucrose treatment. Longer dehydrating period resulted in decreasing germination percentages, but 0.6 and 0.9 M sucrose promoted the 4 and 6 hrs dehydrated Doritis synthetic seed to have higher germination percentages than from other sucrose concentrations. The 4 hrs - dehydrated Dendrobium synthetic seeds yielded highest germination percentage when sown onto the 0.058 M sucrose medium, but the 6 hrs dehydrated synthetic seeds gave highest germination on the 0.6 M sucrose medium. The encapsulated protocorms in calcium alginate beads before preculturing onto modified VW (1949) medium comprising 0.6 M sucrose for 2 days , and dehydrated for 4 hrs yielded highest germination in the Doritis , Dendrobium , Spathoglottis plicata Bl. , and Phaius tankervilliae (Banks in L' Herit.) Bl.). Increasing dehydrating time to 6 hrs , the first 3 species still had high germination percentages, but not significantly differed from the 4 hrs treatment.

Storage of dehydrated synthetic seeds of *Doritis*, *Dendrobium*, and *Spathoglottis* species for 2 hrs at 4 \pm 1, 8 \pm 2, and 25 \pm 2 °C showed that all synthetic seeds could not survive longer than 10 weeks. The non-dehydrated synthetic seeds of the *Dendrobium* could

be preserved at 4 \pm 1 °C for 16 weeks , but germination is declined significantly when the time increased. Storage of the non - dehydrated synthetic seeds of all the 3 species at 25 \pm 2 °C for 16 weeks , yielded high germination percentages i.e. 80.00 - 100.00 , 62.50 - 80.00 and 87.50 - 100.00 respectively.

In vitro germination of the Doritis and Spathoglottis species yielded higher germination than those from the *in vivo* germination. Modified VW (1949) medium + 4 per cent sucrose used as synthetic endosperm helped the synthetic seeds to produced highest germination percentage. Synthetic endosperms comprising 0.0 - 1.5 g/l Agrimycin did not significantly improve *in vivo* germination of both species. But , sucrose at 4 per cent level gave highest germination , but only at 8.20 and 18.30 per cent respectively.

Doritis and Spathoglottis synthetic seeds were precultured onto modified VW (1949) medium containing 0.6 M sucrose and glycerol at 0.0 , 0.5 and 1.0 M for 2 days , prior to dehydrating for 6 hrs , or dehydrating synthetic seeds of the Doritis , Spathoglottis and Phaius species by PVS2 or PVS3 solutions at 25 \pm 2 $^{\circ}$ C for 2 hrs , before cryopreserving in liquid nitrogen showed that the synthetic seeds could not survive. However , very low germination was also obtained from those treatments without cryopreservation.