

Freezing of *Diplonema papillatum*

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Abstract

Citation: Binnypreet Kaur^{1,2}, Drahomíra Faktorová^{1,2}, , Priscila Peña-Díaz¹ and Julius Lukeš^{1,2} Freezing of *Diplonema papillatum*. **protocols.io**

dx.doi.org/10.17504/protocols.io.hfzb3p6

Published: 11 Jul 2018

Protocol

Step 1.

The culture should be in an actively growing state (log phase or exponential growth) to ensure optimum health and good recovery. Ideally, the culture medium should be changed 24 hours prior to harvesting

Step 2.

Label the 1.5ml cryogenic vials and add 200ml of 50% (v/v) glycerol

Step 3.

Add 800ml of active sterile cell culture to this 200ml of 50% (v/v) glycerol, so that overall storage is 10% (v/v) glycerol

Step 4.

Transfer the vials to the controlled freezing unit of at least 1°C/min to retain optimal viability and put the box in -80°C for short storage and in liquid nitrogen for long term storage.