Method for freezing slime

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Recommended Citation

Creighton, M. D., and J.R. Trevithick (1973) "Method for freezing slime," *Fungal Genetics Reports*: Vol. 20, Article 32. <https://doi.org/10.4148/1941-4765.1840>

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Abstract

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Method for freezing slime.

To overcome the necessity of constant **passage** in either liquid or solid medium of the strain **FGSC#326** (fz; sg; as-1, arg-1, cr-1, aur), a slime variant of *N. crassa*, freezing **was** studied as a **holding** method.

Eight-day cultures, grown on Vogel's agar medium (1.5% agar) with arginine (50 mg/100 ml medium) was **washed** down with a stream of Vogel's arginine SSP medium (Vogel's arginine medium plus 2% sucrose, 10% **sorbose**, Penicillin G Sodium 100U/ml of medium, dihydrostreptomycin sulphate 100µg/ml of medium) with a sterile Pasteur pipette. The milky liquid **was** transferred to a **sterile tube** and centrifuged 15 minutes at 2500 rpm.

The **supernatant was** discarded **and** the residue resuspended in the freezing fluid (Vogel's arginine SSP plus **dimethyl sulfoxide**, 4:1), until a four-fold dilution of this suspension gave on absorbance **reading** of 1.0 at 280 nm on a Beckman **spectrophotometer**. 2 ml of the concentrated suspension **was** placed in sterile vials, sealed **and** quick-frozen, using liquid nitrogen. The vials were stored **at -70°C** until used. To date they **have** been held up **to** three months, but longer periods of holding time are being tested. **Thawing was done** in warm **water and** the contents of the vial were emptied into a liter of Vogel's medium plus arginine and antibiotics (Penicillin **and** Streptomycin in the concentrations noted above). No retardation of growth has been noted when the frozen and thawed cultures **have** been compared **to** others **maintained** by continual passage.

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