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Long term storage of Podospora anserina

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Abstract

Until now, maintenance of *Podospora anserina* strains was tedious, requiring yearly sexual crosses, spore isolation and germination, verification of the genotype and transfer of mycelium to stock tubes maintained at 4 C. We have circumvented these difficulties by storage of ascospores at -80 C.

Long term storage of *Podospora anserina*

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Until now, maintenance of *Podospora anserina* strains was tedious, requiring yearly sexual crosses, spore isolation and germination, verification of the genotype and transfer of mycelium to stock tubes maintained at 4 C. We have circumvented these difficulties by storage of ascospores at -80 C. Sexual crosses were made on synthetic medium, either by confrontation or spermatization, to reach high fertility. When perithecia began to shoot ascospores, plates containing agar/salt medium were put upside down over plates containing perithecia. Ascospores were recovered after 24 h by scraping the surface of the agar with a sterile spatula. More than 10(4) asci were recovered from each plate. They were transferred from the spatula to a 1.2 ml Nalgene cryovial containing 0.3 ml sterile storage medium (3 parts 50% glycerol: 7 parts liquid corn meal medium). The vials were vortexed 30 sec and placed at -80 C.

After two years storage, the germination rate of ascospores was only slightly lower than that of freshly isolated ones and several cycles of freeze-thawing did not decrease it significantly (Table 1). This method has been successfully applied to wild-type strains of *Podospora anserina* and to mutant strains displaying ascospores of wild-type phenotype. We are now testing with strains which have more fragile ascospores. For methods and media used in growing and crossing *Podospora anserina*, see Esser, K. 1969. Neurospora Newsletter 15:27-31.

Table 1. Survival of wild-type ascospores after -80 C storage

	Time of storage			
strain*	2 months	14 months	24 months	24 months+
s	20/20	60/60	52/87	71/92
S	8/8	nd	77/83	nd
A	17/17	79/80	nd	72/82

* : s, S and A are three wild-type strains of Podospora

+ : ascospores after 3 cycles of freeze-thawing