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Preservation of amycelial-aconidial Neurospora cultures using anhydrous silica gel

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Abstract

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The Fungal Genetics Stock Center routinely maintains conidial strains of Neurospora stocks on anhydrous silica gel. This method of preserving aconidial-amycelial strains has proved uncertain. Using a modified version of this method described in NN #1 :13 (1962) aconidial-amycelial strains can be preserved. Sterile anhydrous silica gel (Grace-Davison Chemical Corporation, Baltimore, MD, refrigerator grade PA-400), which is a less granular and better grade of silica gel from that described in NN #1, is saturated with a prepared sterile solution of a 1 :1 concentration of each medium: Bacto corn meal ago, with dextrose (Difco #00114-01), Bacto Neurospora culture ago, (Difco #0321-5), Bacto Neurospora minimal (Difco #0817-01), and reconstituted powdered milk (7 grams/100 ml. distilled water).

A sample of the culture is transferred onto the soft agar-silica gel slant and allowed to grow in an incubator at 30-32° C (25° C for temperature sensitive mutants) for 3-5 days. The culture tube (FGSC pre-freezes the culture tube in crushed dry ice) is placed in a vacuum desiccator containing a dish of powdered P_2O_5 as a desiccant and evacuated with a vacuum pump. The desiccato, is stored in a deep freeze overnight to permit desiccation. If the tube is not dry on inspection, it may be necessary to repeat the step. When the culture tube has dried, a layer of sterile anhydrous silica gel is added to the tube and sealed with a sterile screw cop for storage. Submerge the cap completely in molten paraffin for indefinite storage. The extra layer of silica gel is added to take up any moisture that might seep into the tube during storage. The dehydrated culture tuber are stored in a plastic container to which a layer of tel-tale silica gel has been added to absorb any moisture seeping into the storage container. Cultures preserved by this method have been tested after 3-15 months of storage and of the 5, cultures tested, viability was 96 %.

Sampler from the above culture tube during the growth stage hove been used for lyophilization with very reliable results.

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