

was further confirmed as follows. The 10-day-old cultures which were maintained in complete darkness produced no conidiophores, but when exposed to light for 48 h formed abundant conidiophores. Moreover the cultures which had been maintained in continuous light for 10 days, producing abundant conidiophores but no conidia, produced conidia when placed in the dark for 48 h.

The 'light and dark' requirement for conidium formation had also been reported in *Drechslera teres* (Onesirosan & Banttari, 1969), *Helminthosporium dictyoides* (Vargas & Wilcoxson, 1967), *Alternaria solani* (Lukens, 1963), *A. dauci* and *A. tomato* (Leach, 1967). According to Leach's (1967) classification *D. tritici-repentis* would be regarded as a diurnal sporulator on the basis of the above results, since the inductive phase of sporulation requires light, and the terminal phase is accomplished in darkness, under certain conditions. It has been suggested (Lukens, 1963) that in a similar diurnal sporulator, *A. solani*, the inhibition of conidium formation in light is due to the inactivation of flavine, which appeared to be essential for conidium formation.

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DEDIKARYOTIZATION OF *COPRINUS LAGOPUS* FOLLOWING GROWTH WITH 2-DEOXY-D-GLUCOSE

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In the basidiomycete dikaryon two compatible haploid nuclei coexist in the same cell. The condition is an extremely stable one; conjugate nuclear divisions and the elaborate apparatus of the clamp connexion ensuring its continuance as dikaryotic hyphae grow. Dedikaryotization, i.e. the resolution of the dikaryon into its monokaryotic components, is a technique which has particular significance to studies of the effects of dikaryotic cytoplasm on monokaryotic mycelia and of somatic recombination, but

would also be of use in the identification of unknown dikaryons by fertility testing (Miles & Raper, 1956).

Dedikaryotization is easiest in species like *Collybia velutipes*, which produce uninucleate spores on the dikaryon. In work with species like *Coprinus lagopus* and *Schizophyllum commune* which do not normally produce such spores, use must be made of microsurgical operations which kill the terminal cell and its clamp connexion (Harder, 1927; Fries & Aschan, 1952; Papazian, 1955) or which isolate rare monokaryotic outgrowths from unusually large dikaryotic cells such as chlamydo-spores (Lewis, 1961), cystidia (Papazian, 1956) or veil cells (Cowan, 1964). Less exacting techniques, however, make use of chemicals which induce dedikaryotization by interfering with the completion of clamp connexions. Cholic acid and sodium taurocholate are known to be effective (Miles & Raper, 1956). In the course of experiments with the glucose analogue 2-deoxy-D-glucose a new chemical treatment has been developed which gives regular and rapid resolution of *Coprinus* dikaryons.

Table 1. *Resolution of a wild-type dikaryon of Coprinus lagopus induced by 2-deoxy-D-glucose*

Medium	Incubation time (h)	No. of isolates	No. of monokaryotic isolates
5 mm D-glucose	20	57	4
5 mm D-glucose + 10 mm 2-deoxy-D-glucose	44	58	47
5 mm D-fructose	20	51	0
5 mm D-fructose + 0.05 mm 2-deoxy-D-glucose	48	52	48
5 mm sodium acetate	20	54	0
5 mm sodium acetate + 0.05 mm 2-deoxy-D-glucose	48	52	52

The method depends on the initial preparation of a suspension of dikaryotic hyphal fragments. This is readily obtained by scraping dikaryotic aerial mycelium into suspension in sterile water and, after adding a few glass beads, shaking vigorously with a wrist-action shaker. Filtration through cotton yields a suspension of hyphal fragments consisting, in our experience, of from nil to thirty viable cells, with a mean of about six. Appropriate quantities of this suspension are best added to 100 ml volumes of the basal medium of Moore (1969), held molten at 46° and then each dispensed into four Petri dishes. Colonies can be isolated after about 48 h incubation.

Representative results for the resolution of a wild-type dikaryon are shown in Table 1 with the concentrations of analogue and of normal carbon source which have been found most satisfactory.

The indications are that, as with the bile acids used by Miles & Raper (1956), dedikaryotization results from the prevention of the completion of clamp connexions. Under the influence of 2-deoxy-D-glucose, outgrowths destined to become clamp connexions continue to grow away from the parent hypha and become established as monokaryotic branches.

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A SELECTIVE MEDIUM FOR THE
ISOLATION OF BASIDIOMYCETES FROM DISEASED
ROOTS, MYCORRHIZAS, AND SOIL

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Basidiomycetes are an important component in the soil flora, particularly in mycorrhizas. Isolation of slow-growing basidiomycetes from roots or soil is made difficult by competition from other organisms when standard media are used. This note describes a medium which has been developed for the selective isolation of basidiomycetes.

The basal medium is glucose, 10.0 g; KH_2PO_4 , 1.0 g; $(\text{NH}_4)_2\text{SO}_4$, 1.0 g; Mg SO_4 , 0.5 g; peptone, 1.5 g; Davis agar, 15.0 g; water, 1000 ml; pH 5.5. The selective agents used are benomyl (1-butyl carbamyl-2-benzimidazole carbamic acid methyl ester) 5 $\mu\text{g/l}$; neomycin sulphate 50 $\mu\text{g/l}$ and streptomycin sulphate 50 $\mu\text{g/l}$. These are added from stock solutions of 10000 or 1000 $\mu\text{g/l}$ suspended in 0.1% water agar (Nash & Snyder, 1962) kept in a refrigerator, and added to 100 ml aliquots of the basal medium. Thiamine HCl is also added in the same manner to the medium at 100 $\mu\text{g/l}$. The following range of basidiomycetes grew on the medium: Agaricaceae - *Armillariella elegans* Heim, *A. mellea* (Vahl. ex Fr.) Kummer., *A. novo-zelandiae* Stevens., *Agaricus bisporus* (Lange) Pilát, *Collybia* sp., *Flammulina velutipes* (Curt. ex Fr.) Karst., *Hygrophorus* sp., *Marasmius oreades* (Bolt. ex Fr.) Fr., *Merulius americanus* Burt, *Paxillus involutus* (Batsch ex Fr.) Fr., *Russula* sp.; Polyporaceae - *Fomes annosus* (Fr.) Karst., *F. pini* (Thore ex Pers.) Lloyd, *Polyporus adustus* Willd. ex Fr.,