JOURNAL OF CLINICAL MICROBIOLOGY, July 1975, p. 74-75
Vol.2, No. 1

Printed in U.S.A.

Evaluation of *Aspergillus* Differential Medium

I. F. SALKIN* AND M. A. GORDON

Division of Laboratories and Research, New York State Department of Health, Albany, New York 12201

Received for publication 21 March 1975

The new *Aspergillus* differential medium distinguishes effectively between members of the *Aspergillus flavus* group and other *Aspergillus* species of interest in medical mycology.

Bothast and Fennell (1) have recently devised a culture substrate, *Aspergillus* differential medium (ADM), to distinguish members of the *Aspergillus flavus* group from other species of *Aspergillus* and common storage molds.

TABLE 1. *Production of yellow-orange reverse pigment on ADM*

Species of *Aspergillus* (and culture numbers)[a]	Production of pigment[b]
A. fischeri: 5 cultures (M 34 C, NRRL 181, NRRL 5034, QM 1983, UAMH 464)	−
A. flavus: 8 cultures (M 32 C, M 32 D, M 32 E, NRRL 627, NRRL 1820, QM 380, UAMH 266, UAMH 2679)	+
A. fumigatus: 22 cultures (M 26, M 26 B, M 26 C, M 27, M 27 A, M 27 B, M 27 C, M 27 D, M 28, M 28 A, M 28 B, M 28 C, M 29, M 29 A, M 29 B, M 29 C, M 30 A, M 30 B, M 30 C, M 33, M 33 A)	−
A. nidulans: 7 cultures (M 31 B, NRRL 187, NRRL 5218, QM 1985, QM 9637, UAMH 1656, UAMH 1782)	−
A. niger: 3 cultures (M 31, M 31 A, M 31 Aa)	−
A. oryzae: 4 cultures (M 1340, QM 82 i, QM 1273, NRRL 493)	+
A. oryzae: 1 culture (NRRL 482)	−
A. terreus: 10 cultures (M 35 A, NRRL 265, NRRL 1960, QM 72 f, QM 82 j, QM 1991, QM 1992, QM 8010, UAMH 327, UAMH 1897)	−
A. ustus: 1 culture (M 34 A)	−
A. versicolor: 1 culture (M 34 D)	−
A. wentii: 1 culture (M 35 C)	−

TABLE 1—Continued

[a] Sources of cultures: M, Laboratories for Mycology, Division of Laboratories and Research, New York State Department of Health, Albany, N.Y.; NRRL, Northern Regional Research Laboratory, U.S. Department of Agriculture, Peoria, Ill.; QM, NLABS Culture Collection of Fungi, Department of Botany, University of Massachusetts, Amherst, Mass.; UAMH, Mold Herbarium and Culture Collection, University of Alberta, Edmonton, Alberta, Canada.

[b] Assessed 6 days after inoculation.

Their study encompassed 65 strains of several species of *Aspergillus* and other molds. All 5 strains of *A. flavus* tested, 5 of 11 representative strains of additional species and varieties in the *A. flavus* group, and a few species in other *Aspergillus* groups produced a yellow-orange pigment on the reverse of ADM cultures. However, the medium was tested on few zoopathogenic species other than *A. flavus;* in particular, *A. fumigatus* was omitted. This prompted us to investigate the usefulness of the new medium for differentiation of these two species as well as others of interest to medical mycology. The group of organisms chosen for study comprised 63 strains in 10 species of *Aspergillus*, most of which contain isolates pathogenic for either humans or lower animals. Except for 1 isolate (noted below), none of the 63 we studied was employed by Bothast and Fennell.

ADM was prepared in slants of 7 ml in glass culture tubes (16 by 150 mm), according to the formula of Bothast and Fennell. Each slant was inoculated with a 1-mm cube of agar with mycelium from the advancing edge of a 2-week-old stock culture, incubated at 27 C, and observed daily for color change.

All 8 strains of *A. flavus* tested produced the yellow-orange pigment on the reverse of ADM slants (Table 1), as did 4 of 5 strains of *A. oryzae*, a member of the *A. flavus* group (2). After 6 days of incubation, no color change was

observed with any of the other species, including 22 clinical isolates of *A. fumigatus*.

Our results indicate that ADM can serve as an aid in differentiating members of the *A. flavus* group from other species of *Aspergillus* of particular interest to medical mycology. The bright yellow-orange pigment greatly facilitates the identification of *A. flavus* and would, as noted by Bothast and Fennell, permit the effective use of ADM by technicians untrained in mycology. However, as with other primary diagnostic procedures, ADM should not be employed as the sole method of identifying isolates as members of the *A. flavus* group.

Pigment production by some isolates of *A. oryzae* may present a diagnostic problem, especially since the morphology of this species is very similar to that of *A. flavus*. However, *A. oryzae* rarely becomes pathogenic and is less commonly encountered than most of the other species of interest to medical mycology. We have no ready explanation for the fact that pigment was produced by 5 of our 6 isolates but by only 2 of the 7 strains tested by Bothast and Fennell. However, the 1 strain common to both test groups (NRRL 482) was nonreactive.

ADM may prove to be especially useful in geographic regions such as the Sudan, where *A. flavus* is one of the most frequently isolated species of this genus (3, 4).

LITERATURE CITED

1. Bothast, R. J., and D. I. Fennell. 1974. A medium for rapid identification and enumeration of *Aspergillus flavus* and related organisms. Mycologia 66:365–369.
2. Raper, K. B., and D. I. Fennell. 1965. The genus *Aspergillus*. The Williams and Wilkins Co., Baltimore.
3. Sandison, A. T., J. C. Gentles, C. M. Davidson, and M. Branko. 1967. Aspergilloma of paranasal sinuses and orbit in Northern Sudanese. Sabouraudia 6:57–69.
4. Veres, B., O. A. Malik, A. A. El Tayeb, S. El Daqud, E. S. Mahgoub, and A. M. El Hassan. 1973. Further observations on the primary paranasal *Aspergillus* granuloma in the Sudan. Am. J. Trop. Med. Hyg. 22:765–772.