Reading the Results

Evaluation

Observe the medium for growth and color change. Without opening the InTray DM-FungID, place the unopened tray under a microscope lens to view the organisms using the 10x objective (100x power) to view distinct fungal structures (i.e. hyphae, micro/marco-conidia). Trays to be used with 10x objective ONLY!. Staining is not required. See the Identification Chart below.

Mixed Growth: Dermatophytes and saprophytes (contaminants) will grow on the same tray. The dermatophytes will start to grow first and will turn the media red around the colony. The saprophytes will grow but there will be no color change around the colony until the colony matures. The colony growth color will change from white to yellow, black, brown, or green.

Positives: If, within 1-14 days, the medium color changes to red at the location of the specimen and whitish colonies grow, the InTray DM-FungID is presumptive positive.

Negatives: Trays that show no colony growth or color change 14 days after inoculation are presumptive negative.

Dermatophyte Identification

This is a selection of commonly encountered organisms. Please consult our DM Wall Chart (Cat. No. 100-000-005; also available online at biomeddiagnostics.com), for a more detailed selection, and the references listed below, as well as other standard mycology and microbiology references.



Trichophyton rubrum Septate hyphae. Macroconidia: $(4-6 \times 15-30 \ \mu m)$ abundant, rare or absent, however can be long, narrow, thin-walled, parallel sides, $2-8 \ cells$, may form on the ends singly or in groups. Microconidia: $(2-3 \times 3-5 \ \mu m)$ lateral, teardrop shaped, form on macroconidia.



Trichophyton mentagrophytes Septate hyphae. Macroconidia: (4-8 \times 20-50 μ m) occasionally present, cigar-shaped, thin-walled, narrow attachments to septate hyphae, 1-6 cells, found in young cultures 5-10 days old. Microconidia: usually present in powdery cultures, very round, clustered on branched conidiophores; in fluffy cultures, smaller, fewer, teardrop shaped and easily confused with those of *T. rubrum*.



Epidermophyton floccosum Septate hyphae. Macroconidia: $(7-12 \times 20-40 \, \mu m)$ smooth, thick and thin-walled, club shaped with rounded ends, two to six cells, singly or clusters. Microconidia: none.

Saprophyte (Contaminants) Identification



Alternaria sp. Hyphae are septate and dark. Conidiophores are septate, variable in length and sometimes branched. Macroconidia are large [7-10 by 23-24 µm), brown, have both transverse and longitudinal stations, and found singly or in chains. They are usually round at the end nearest the conidiophore, producing a club-like shape. Day 10-14: Colony growth with no initial color change. Colony Morphology - Formation of grayish-white, wooly colonies 10 to 14 days after inoculation, that later become greenish black/brown with a light border. It may eventually become covered by short grayish aerial hyphae. Reverse side is black. The medium will change to pink when the colony changes color.



Aspergillus sp. Microsco pic morphology – Septate hypae (2.5-8 µm in diameter); unbranched condidiophore arises from a specialized foot cell. The conidiophore is enlarged at the tip, forming a swollen vesicle completely or partially covered with flask-shaped phialides. The pialides produce chains of mostly round, sometimes rough conida (2-5 µm in diameter). Day 10-14: Colony growth with no initial color change. Formation of white, cottony colonies 10 to 14 days after inoculation that latter become yellow, green, black or brown. Reverse side is white, goldish, or brown. The medium will change to red when the colony changes color



Penicillium sp. Microscopic morphology – Septate hyphae (1.5-5 µm in diameter) with branched conidiophores that have secondary branches known at metulae. On the metulae, are flask-shaped phialides that bear unbranched chains of smooth or rough conidia (2.5-5 µm in diameter). The entire structure forms the characteristic "penicillus" or "brush" appearance. Day 10-14: Colony growth with no initial color change. Colony morphology – Surface is at first white, then becoming very powdery, bluish green with a white border. Some less common species differ in color. Reverse is usually white but may be red or brown. DM-FungID medium will change to pink/red when the colony changes color.

Limitations

If a fungal infection is strongly suspected and the test result is negative, it may be appropriate to retest giving more care to specimen collection.

Some soaps and topical agents may cause an immediate color change. If this occurs, discard the test, wash area and re-sample.

- Optimally, simultaneously inoculate samples on both InTray SAB-FungID w/ CC (Cat. Nos. 11-283-001, 11-283-002) and InTray SAB-FungID (Cat. Nos. 11-263-001, 11-263-002) without antimicrobials (e.g., for some pathogenic fungi inhibited by antimicrobials).
- Transfer of fungal growth to slide examination: Stained slide preparations can be made as
 appropriate based on 10x objective examination. InTray PDA-FungID (Potato Dextrose
 Agar, Cat. Nos. 11-293-001, 11-293-002) can be used to induce sporulation.

The InTray DM-FungID is an agar medium that is susceptible to condensation collection within the inner seal, especially when stored at low temperatures and/or having been exposed to extreme temperature fluctuations. If moisture is visible on the surface of the InTrays, dry them (with the seal removed and InTray label in a position allowing for air flow) under a BSL-2 cabinet just prior to inoculation. There should be no visible droplets of moisture on the surface of the agar when they are inoculated. The surface of the dried medium should be smooth and should not show signs (webbed ribbing pattern on the agar surface) of desiccation.

References

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Symbol glossary: biomeddiagnostics.com/l/symbol-glossary

Document Revision History

Rev. A, September 2019

New format; corrected name InTray DM-FungID as needed; added new catalog numbers, document revision history, reference to online symbol glossary; specified $18-25^{\circ}$ C instead of room temperature; reorganized and retilled some sections; removed some organisms from organism identification table and added reference to DM Wall Chart and listed references; removed Technical notes and some references.



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InTray® **DM-FungID**™

Enriched Dermatophyte Medium

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20

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Introduction

Intended Use

InTray® DM-FungID™ is an enriched dermatophyte medium used to aid in the detection of dermatophytes from clinical specimens with mixed micobiota.

Description and Principle

Dermatophytes are fungi in the genera Microsporum, Tricophyton and Epidermophyton. They are capable of metabolizing keratin found in skin, hair and nails of living hosts. The fungi characteristically may invade the cutaneous tissue of the living host but rarely penetrate the subcutaneous tissue. Tinea and ringworm are two terms commonly used to describe dermatophytes.

InTray DM-FungID is formulated to produce a red color in the presence of growing dermatophytes. Moreover, the medium is formulated to produce distinctive colony growth with typical identifying characteristics both macro- and microscopically. The medium inhibits most gram-positive bacteria, gram-negative bacteria, yeast and saprophytic fungi. It is a single exposure system with dynamic built-in components and features that are designed for user compatibility and ease of detection.

Reagents and Appearance

This product appears transparent with a yellow hue and contains soytone, carbohydrate, growth stimulants, antimicrobial agents including: cycloheximide, color indicator and agar in distilled water. The final pH of the media is 5.6 ± 0.1 at 25° C.

Precautions, Safety and Disposal

For In Vitro Diagnostic Use. For professional use only.

Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing and gloves.

Once the tray has been inoculated and resealed, re-open only in a biological safety cabinet. Because of the potential for containing infectious materials, the tray must be destroyed by autoclaving at 121°C for 20 minutes.

Storage

Upon receipt, store InTray DM-FungID at 18-25°C. Avoid refrigeration, freezing or prolonged storage at temperatures greater than 40°C. Do not use an InTray DM-FungID if the medium shows signs of deterioration or contamination.

Shelf Life

InTray DM-FungID expires 27 months from the date of manufacture.

Procedure

Key notes regarding specimen collection: Specimen collection poses a major uncertainty in using this device.

NAILS - Often collecting viable material from infected nails is difficult because the living organisms are well under the nail itself. For best results, cut nails into small pieces.

HAIR - Samples should be grasped at the uninfected end and several (3-6) small pieces, about 2 cm long, should be cut from the infected portion for inoculation onto the medium.

SKIN - Scrapings should be taken with an inoculation tool that has been moistened with the medium or a sharp blade from the outer ridge of an active lesion. Vesicular fluid is unacceptable for dermatophyte culture. If vesiculated, skin scrapings should be taken from the surface.

Materials Provided

InTray DM-FungID test(s)

Materials Required but Not Provided

- Sterile inoculation tool (e.g., cotton swab/forceps/scalpel blade)
- Laboratory incubator capable of incubation at 18-30°C

Prepare Sample:

Use a septic technique during specimen collection and handling. Remove any soap residue from the sampling area. Clean the area with 70% alcohol and permit to air dry.

Collect Sample:

InTray DM-FungID is designed for culturing hair, skin and nail samples (i.e., cuttings/scrapings). All specimens should be handled according to CDC infectious materials isolation guidelines;

cdc.gov/infectioncontrol/guidelines/isolation

1 Prepare InTray



Immediately label the tray with patient/sample information and date. Pull back the lower right corner adjacent to the clear window of the tray label until the protective seal is completely visible.

Remove the seal by pulling the tab. Discard the seal.

DO NOT REMOVE OR ALTER THE WHITE FILTER STRIP OVER THE VENT HOLE!

2 Inoculate Sample



Inoculate the specimen on the center surface of the medium. A sterile inoculating loop that has been moistened by touching the surface of the medium may be used for inoculation of solids or scrapings.

Re-seal all around the tray to ensure a complete seal by pressing the edges of the label against the plastic tray.

DO NOT COVER THE VIEWING WINDOW. Complete re-seal prevents dehydration!

Incubation

Incubate inoculated trays for up to 14 days at 18-30°C, in the dark. Observe the trays daily for color changes, through the clear viewing window.

Quality Control

This product has been tested and meets the CLSI (formerly NCCLS) Approved Standard for commercially prepared media (M22-A3). At the time of manufacture, quality control testing is performed on each lot of the InTray DM-FungID. The ability of the media to support growth and demonstrate expected biochemical reactions and morphology is verified by lot.

Recommended strains for QC testing of InTray DM-FungID

Test Strain	ATCC	Expected Result
T. mentagrophytes	9533	Growth
T. rubrum	28188	Growth
T. tonsurans	28942	Growth
M. gypseum	14683	Growth
A. brasiliensis	16404	Significant Inhibition
S. aureus	25923	Significant Inhibition
E. coli	25922	Significant Inhibition
C. albicans	60193	Significant Inhibition