

INTRODUCTION

INTENDED USE

InTray™ DM is an enriched dermatophyte medium used in the detection of dermatophytes from clinical specimens.

DESCRIPTION OF THE SYSTEM AND ITS PRINCIPLES

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.

PRINCIPALS OF THE PRODUCT

The InTray™ DM 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

This product contains soytone, carbohydrate, growth stimulants, antimicrobial agents including: cycloheximide, color indicator and agar in distilled water.

STORAGE

InTray™ DM expires 27 months from the date of manufacture. Upon receipt, store InTray™ DM at room temperature (18-25°C). Avoid refrigeration, freezing or prolonged storage at temperatures greater than 40°C. Do not use an InTray™ DM if the medium shows signs of deterioration or contamination.

KEY NOTES REGARDING SPECIMEN COLLECTION

Specimen collection poses the major uncertainty in using this item.

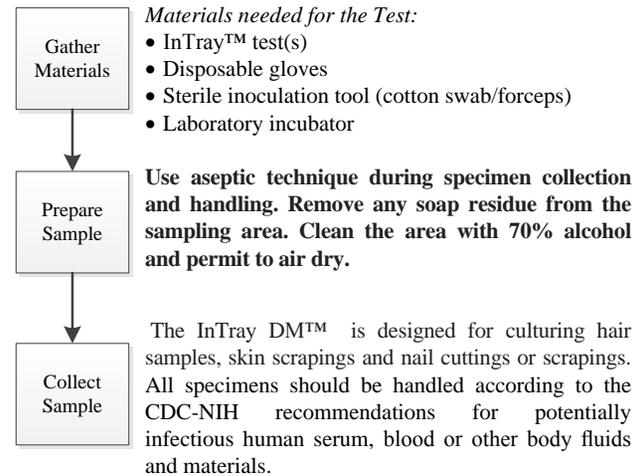
NAILS - Often collecting viable material from infected nails is difficult because the living organisms are well under the nail itself. For best results, cut nails into small pieces.

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.

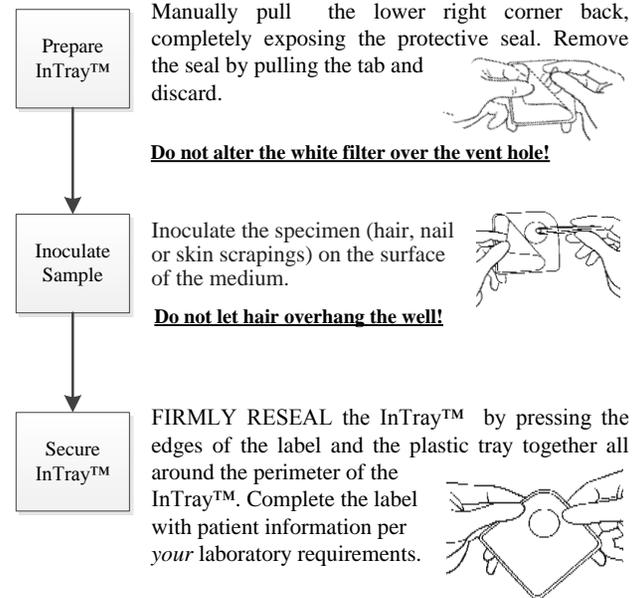
SKIN - scrapings should be taken with an inoculation tool that has been moistened with the medium or a sharp blade from the outer ridge of an active lesion. Vesicular fluid is unacceptable for dermatophyte culture. If vesiculated, skin scrapings should be taken from the surface.

INSTRUCTIONS

SAMPLE COLLECTION



INOCULATION



INCUBATION

Incubate inoculated trays for up to 14 days at room temperature (18°C-25°C), in the dark. Observe the trays daily for color changes, through the clear viewing window.

SAFETY & DISPOSAL

Since InTray™ DM has potential for containing live, infectious materials, it must be handled and destroyed in accordance with BSL-2 requirements. Destroy the InTray™ by autoclaving at 121°C for 20 minutes or other suitable sterilization means.

READING THE RESULTS

EVALUATION

Microscopically observe the medium for growth and color change **without** opening the InTray™ DM by placing the unopened tray under a microscope lens to view the organisms using 100x and 200x power. Staining is not required. See the Identification Chart on the following page.

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 color changes 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 is presumptive positive.

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

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.

Media color change and colony growth will vary depending upon patient's physical condition and medical history. Samples from patients who have been treated for ringworm (prescriptions, OTC products herbs, shampoos, etc.) may not grow macroscopic colonies but may still show media color change and microscopic growth with distortions to hyphae and spores.

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

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. The ability of the media to support growth and demonstrate expected biochemical reactions and morphology is verified.

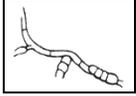
Table 1: Recommended Strains for QC Testing the InTray™ DM

Test Strain	ATCC Number	Expected Results
<i>T. mentagrophytes</i>	9533	Growth
<i>T. rubrum</i>	28188	Growth
<i>T. tonsurans</i>	28942	Growth
<i>M. gypseum</i>	14683	Growth
<i>Aspergillus niger</i>	16404	Significant Inhibition
<i>Staphylococcus</i>	25923	Significant Inhibition
<i>E. coli</i>	25922	Significant Inhibition
<i>C. albicans</i>	60193	Significant Inhibition

DERMATOPHYTE IDENTIFICATION

The dermatophyte medium changes to red in the presence of these and other common, growing dermatophytes 2 to 14 days after inoculation.

Epidermophyton floccosum



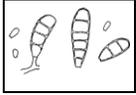
Septate hyphae. Macroconidia: (7-12 x 20-40µm) smooth, thick and thin-walled, club shaped with rounded ends, two to six cells, singly or clusters. Microconidia: none.

Microsporium canis



Septate hyphae. Macroconidia: (10-25 x 35-110µm) numerous, long, spindle-shaped, rough, thick walls are apparent at the knob-like ends. Microconidia: few in number, smooth and club-shaped.

Microsporium gallinae



Septate hyphae. Macroconidia are clavate to cigar shaped (6-8 x 15-50µm) and 2 to 10 celled. Microconidia are unicellular and ovoid to pyriform in shape.

Microsporium gypseum



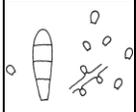
Septate hyphae. Macroconidia: (8-16 x 22-60µm) large numbers, symmetrical, rough and relatively thin-walled with rounded ends, not pointed like *M. canis*. Microconidia: usually present, club-shaped.

Microsporium nanum



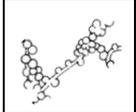
Septate hyphae. Macroconidia (4-8 x 12-18µm) are rough, fairly thin-walled (as in *M. gypseum*), egg-shaped with a truncated base, having usually two cells. Microconidia are club shaped and smooth-walled. Abundance may vary.

Trichophyton equinum



Abundant microconidia which may clavate to pyriform and sessile or spherical and stalked are formed laterally along the hyphae. Macroconidia are rarely produced, but when present are clavate, smooth, thin walled and of variable size.

Trichophyton mentagrophytes



Septate hyphae. Macroconidia: (4-8 x 20-50µm) 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*.

Trichophyton rubrum



Septate hyphae. Macroconidia: (4-6 x 15-30µm) 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 x 3-5µm) lateral, teardrop shaped, form on macroconidia.

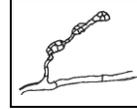
Trichophyton tonsurans



Septate hyphae with variable shaped microconidia all along the hyphae or on short conidiophores that are perpendicular to the parent hyphae. Microconidia are usually teardrop or club-shaped, but may elongate or enlarge to round "balloon" forms. Macroconidia are rare, irregular in form and thick-walled. Many have spiral coils and arthrospores.

SAPROPHYTE (CONTAMINANTS) IDENTIFICATION

Alternaria sp. (Saprophyte)

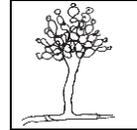


Hyphae are septate and dark. Conidiophores are septate, variable in length and sometimes branched. Macroconidia are large (7-10 by 23-24µm), brown, have both transverse and longitudinal stations, and found singly or in chains. They are usually round at the end nearest the conidiophore, producing a club-like 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. (Saprophyte)

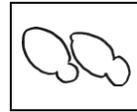


Microscopic morphology - Septate hyphae (2.5 - 8µm in diameter); unbranched conidiophore arises from a specialized foot cell. The conidiophore is enlarged at the tip, forming a swollen vesicle completely or partially covered with flask-shaped phialides. The phialides produce chains of mostly round, sometimes rough conidia (2-5µm in diameter)

Day 10-14: Colony growth with no initial color change.

Formation of white, cottony colonies 10 to 14 days after inoculation that later become yellow, green, black or brown. Reverse side is white, goldish, or brown. The medium will change to red when the colony changes color

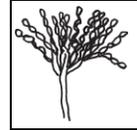
Candida albicans & Candida tropicalis



Microscopic examination shows clusters of bubbles in the media. Yeast is a common contaminant and there are inhibitors in the media to prevent most yeast species from growing. However, yeast species can be pathogenic depending upon the patient's overall condition and medication history.

Day 1-5: Pink to red color change, no visible growth.

Penicillium sp. (Saprophyte)



Microscopic morphology - Septate hyphae (1.5-5µm in diameter) with branched conidiophores that have secondary branches known as metulae. On the metulae, are flask-shaped phialides that bear unbranched chains of smooth or rough conidia (2.5-5µm 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 medium will change to pink/red when the colony changes color.

REFERENCES

1. Kwon-Chung, K.J. and Bennett, J.E., Medical Mycology, Lea and Febiger: Philadelphia, 1992.
2. Murray, P.R., Baron, ET, Pfaller, M.A., Tenover, F.C., Tenover, R.H., Manual of Clinical Microbiology 6th ed., American Society for Microbiology: Washington, D.C. 1995, pp. 709-722.
3. Larone, D.H., Medically Important Fungi: A Guide to Identification, 2nd ed., American Society for Microbiology: Washington, D.C., 1995.

InTray™ DM

Enriched Dermatophyte Medium

Catalog No. 10-4007 5 Test Kit

Catalog No. 10-4001 20 Test Kit

A SELECTIVE CULTURE SYSTEM
FOR THE IDENTIFICATION OF
Dermatophytic fungi

For *In Vitro* Diagnostic Use



BIOMED

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