



## HiCrome™ Universal Differential Medium

M1600

### Intended Use:

Differential medium recommended for presumptive identification of microorganisms from clinical and non-clinical specimens.

### Composition\*\*

| Ingredients         | g / L   |
|---------------------|---------|
| Peptone             | 15.000  |
| Chromogenic mixture | 2.500   |
| Tryptone            | 4.000   |
| Agar                | 13.500  |
| Final pH ( at 25°C) | 7.2±0.2 |

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 35.00 gram in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

### Principle And Interpretation

HiCrome™ Universal Differential Medium is a modification of the medium formulated on basis of the work carried out by Pezzlo (1), Wilkie et al (2), Friedman et al (3), Murray et al (4), Soriano and Ponte (5) and Merlino et al (6). HiCrome™ Universal Differential Medium is recommended for the presumptive identification of microorganisms from clinical and non-clinical specimens where the medium has broader application as a general nutrient agar for isolation of various microorganisms. This medium helps in the identification of some gram-positive bacteria and gram-negative bacteria on the basis of different colony colours exhibited by them. These colours are formed due to the reactions of genus or species specific enzymes with the two chromogenic substrates incorporated in the medium. *Enterococcus* species, *Escherichia coli* and coliforms produce enzymes which specifically cleave these chromogenic substrates to give characteristically distinctive colony colours. Peptones in the medium serve as sources of amino acids like phenylalanine and tryptophan which aids in indicating tryptophan deaminase activity, thereby facilitating the identification of *Proteus* species, *Morganella* species and

*Providencia* species. One of the chromogenic substrate is cleaved by  $\beta$ -glucosidase enzyme possessed by Enterococci resulting in the formation of bluish green colonies. *Escherichia coli* possesses the enzyme  $\beta$ -galactosidase which specifically cleaves the other chromogenic substrate resulting in the formation of purple coloured colonies. *Escherichia coli* can be differentiated and confirmed from other similar coloured colonies, by performing the indole test.

Coliforms cleave both the chromogenic substrates forming blue to purple coloured colonies. Colonies of *Proteus*, *Morganella* and *Providencia* species appear brown due to tryptophan deaminase activity. Peptone and tryptone provide nitrogenous, carbonaceous compounds, essential growth nutrients and also serve as a source of amino acids.

### Type of specimen

Clinical samples : urine, faeces, etc.; Food and dairy samples, Water samples.

### Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (7,8). For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (9,10,11). For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (12). After use, contaminated materials must be sterilized by autoclaving before discarding.

### Warning and Precautions

In Vitro diagnostic use. For professional use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

## Limitations

1. Since it is an enzyme-substrate based reaction, the intensity of colour may vary with isolates.
2. Final confirmation of suspected colonies must be carried out by serological and biochemical tests.

## Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

## Quality Control

### Appearance

Cream to yellow homogeneous free flowing powder

### Gelling

Firm, comparable with 1.35% Agar gel

### Colour and clarity of prepared medium

Light amber coloured, clear to slightly opalescent gel forms in Petri plates

### Reaction

Reaction of 3.5% w/v aqueous solution at 25°C. pH : 7.2±0.2

### pH

7.00-7.40

### Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

| Organism  | Inoculum (CFU) | Growth    | Recovery | Colour of Colony                              |
|---|----------------|-----------|----------|---|
| <i>Enterococcus faecalis</i> ATCC 29212 (00087*)                      | 50-100         | luxuriant | ≥70%     | blue, small                                   |
| <i>Escherichia coli</i> ATCC 25922 (00013*)                           | 50-100         | luxuriant | ≥70%     | purple  |
| <i>Klebsiella pneumoniae</i> ATCC 13883 (00097*)                      | 50-100         | luxuriant | ≥70%     | blue -green, mucoid                           |
| <i>Pseudomonas aeruginosa</i> ATCC 27853 (00025*)                     | 50-100         | luxuriant | ≥70%     | colourless (greenish pigment may be observed) |
| <i>Proteus mirabilis</i> ATCC 12453                                   | 50-100         | luxuriant | ≥70%     | light brown                                   |
| <i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*) | 50-100         | luxuriant | ≥70%     | golden yellow                                 |
| <i>Salmonella</i> Typhi ATCC 6539                                     | 50-100         | luxuriant | ≥70%     | colourless                                    |
| <i>Salmonella</i> Typhimurium ATCC 14028 (00031*)                     | 50-100         | luxuriant | ≥70%     | colourless                                    |

Key : \*Corresponding WDCM numbers.

## Storage and Shelf Life

Store between 15-25°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

## Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (7,8).

## Reference

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8. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock, D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
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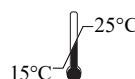
Revision : 04/2024



HiMedia Laboratories Pvt. Limited,  
Plot No.C-40, Road No.21Y,  
MIDC, Wagle Industrial Area,  
Thane (W) -400604, MS, India



**In vitro diagnostic  
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