University of AL-Mustansiriyah College of Sciences Biology Department Bacterial Taxonomy

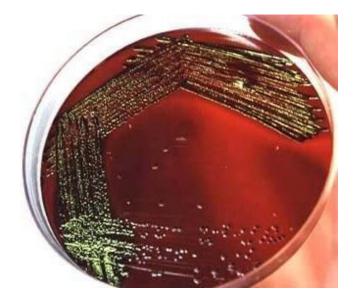
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Lab. 2: The differential media

Differential media contain compounds that allow groups of microorganisms to be visually distinguished by the appearance of the colony or the surrounding media, usually on the basis of some biochemical difference between the two groups i.e. (contains substances that if used by the organism cause a visible change in the medium or in the colony, examples include:

1. Eosin Methylene Blue agar (EMB agar)

Contains the dyes eosin and methylene blue. They inhibit Gram-positive organisms; such a medium is selective for Gram-negative species. Lactose-fermenting organisms such as *Escherichia coli* appears as large, blue-black colonies, often with a green metallic sheen. *Enterobacter* spp. present as brown to blue-black, mucoid colonies with no sheen. Non-lactose-fermenting colonies such as *Shigella* spp. and *Salmonella* spp., appear transparent and colorless. Thus, the medium is considered differential with respect to lactose fermentation.



E. coli on EMB agar

2. MacConkey agar

MacConkey agar is similar to EMB agar in that it is also selective for Gram-negative species and differential with respect to lactose fermentation. MacConkey agar is used for the detection of coliforms and enteric pathogens based on their ability to ferment lactose. Lactose-fermenting bacteria appear red to pink (*Escherichia coli, Enterobacter* and *Klebsiella*) while non-lactose fermenting bacteria appears as colorless or transparent colonies (*Salmonella, Proteus* species, *Yersinia* and *Shigella*).



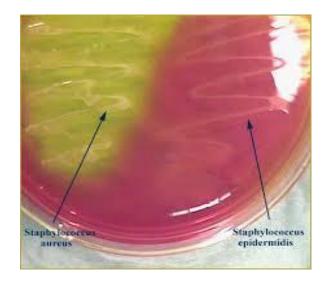
MacConkey Agar

3. Mannitol salt agar

Selective medium (7.5% NaCl) for staphylococci and differential with respect to mannitol fermentation. Growth of most bacteria other than staphylococci, is inhibited by the high concentration of salt in the medium. Fermentation of mannitol is only seen in the pathogenic species of *Staphylococcus* and is signaled by the production of acidic products leading phenol red in the media to change from a neutral red-orange to bright yellow.

** Staphylococcus: fermenting mannitol, medium turns yellow (e.g. S. aureus)

** Staphylococcus: not fermenting mannitol, medium does not change color (e.g. S. epidermidis)



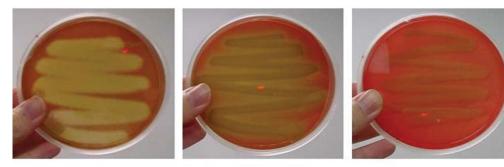
4. Blood agar

Is a nutritive medium with differential properties in respect to heamolysis. **Heamolysis is the destruction of erythrocytes (RBCs).** The degree to which the erythrocytes are destroyed can be recognized by the effects of heamolysin on the cells in the medium.

When **alpha heamolysis** (α-heamolysis) (Partial destruction of the RBCs) is present, the agar under the colony is dark and greenish (such as *Streptococcus pneumoniae*).

Beta heamolysis (β -heamolysis) sometimes called complete hemolysis, is a complete <u>lysis</u> of RBCs in the media around and under the colonies: the area appears lightened (yellow) and transparent (such as *Streptococcus pyogenes*).

Camma (γ) hemolysis is usually the term applied to growth on blood agar that causes no damage to the RBCs (no hemolysis) and no change in the medium (such as *Staphylococcus epidermidis*).



Beta Hemolysis

Alpha Hemolysis

Gamma Hemolysis