Biochemical test for identification and differentiations of Enterobacteriaceae

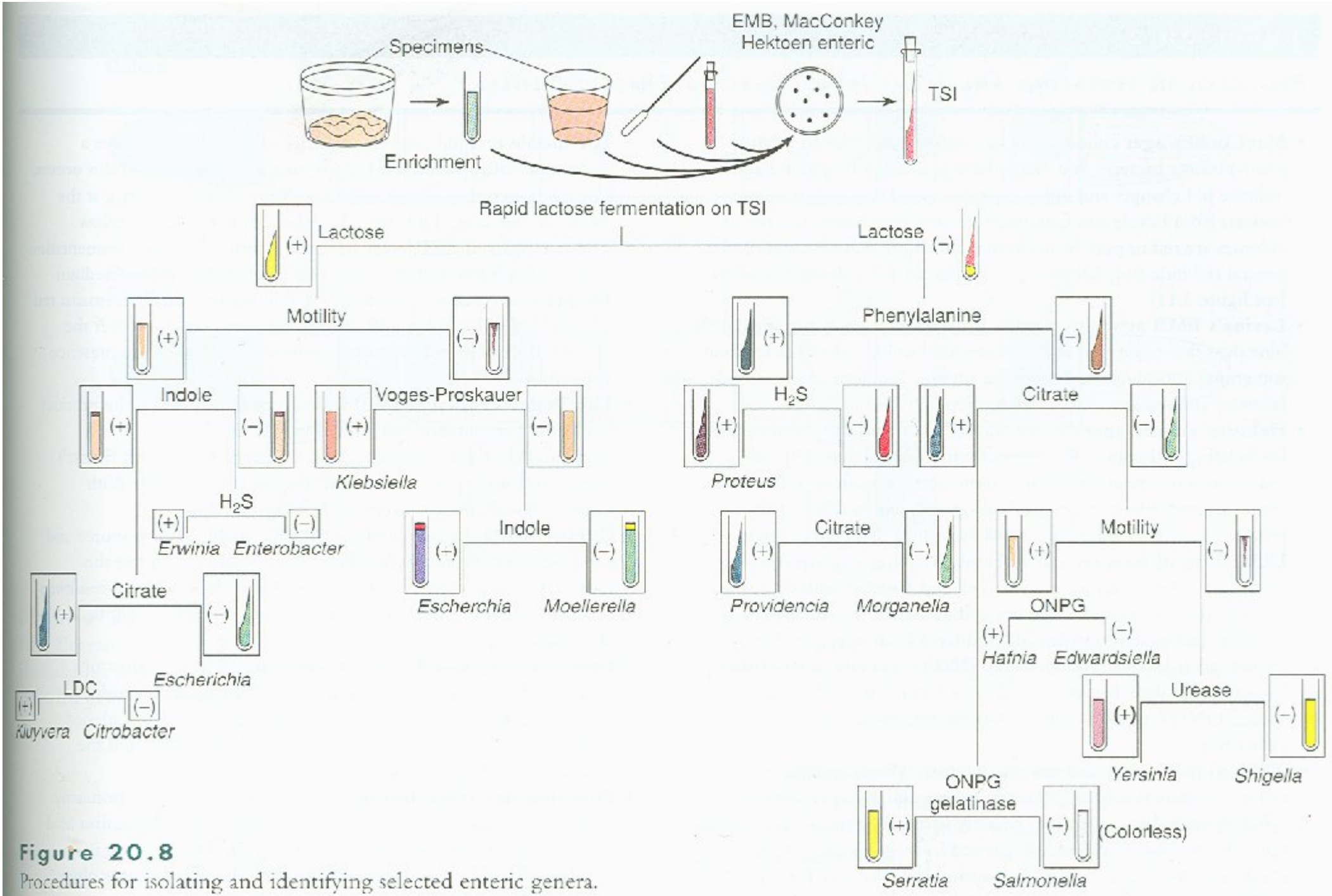
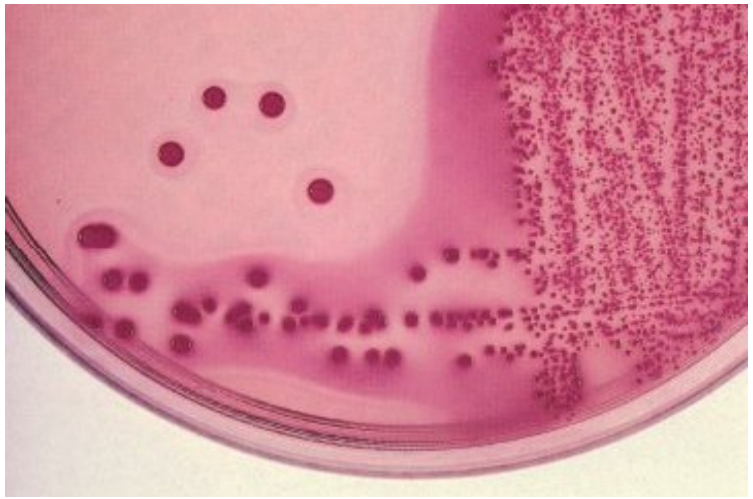


Figure 20.8
Procedures for isolating and identifying selected enteric genera.

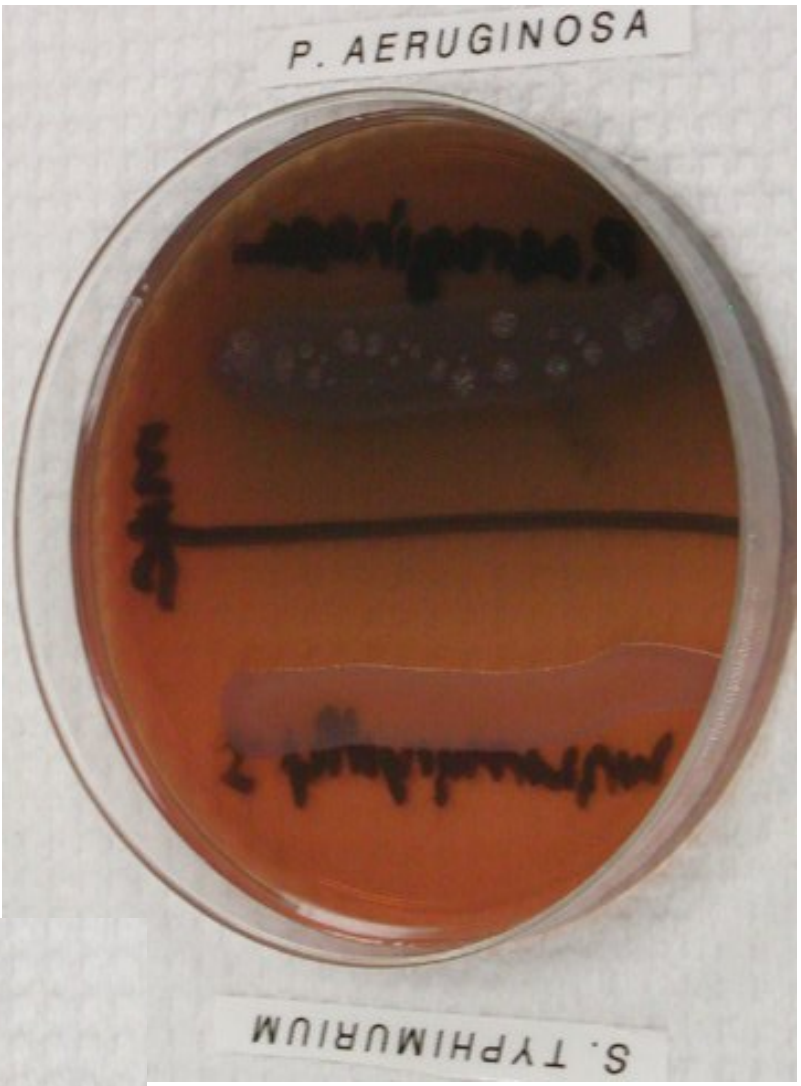
MacConky agar

- Bile salts
- Crystal violet
- Lactose
- Neutral red

Lac – not colored



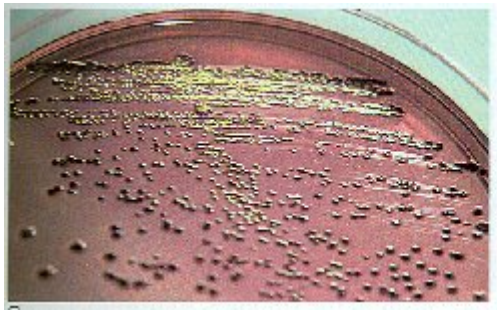
Lac + pink or red

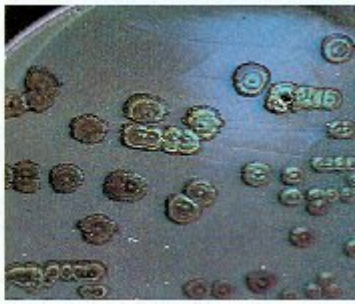

P. AERUGINOSA
S. TYPHIMURIUM

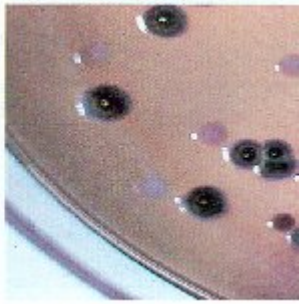

M. LUTEUS
K. PNEUMONIAE

Levines's EMB agar

- Bile salts
- Eosin and methylene blue precipitate at low PH
- Lactose + dark nucleus
- Lactose – pale







Hektoen enteric agar

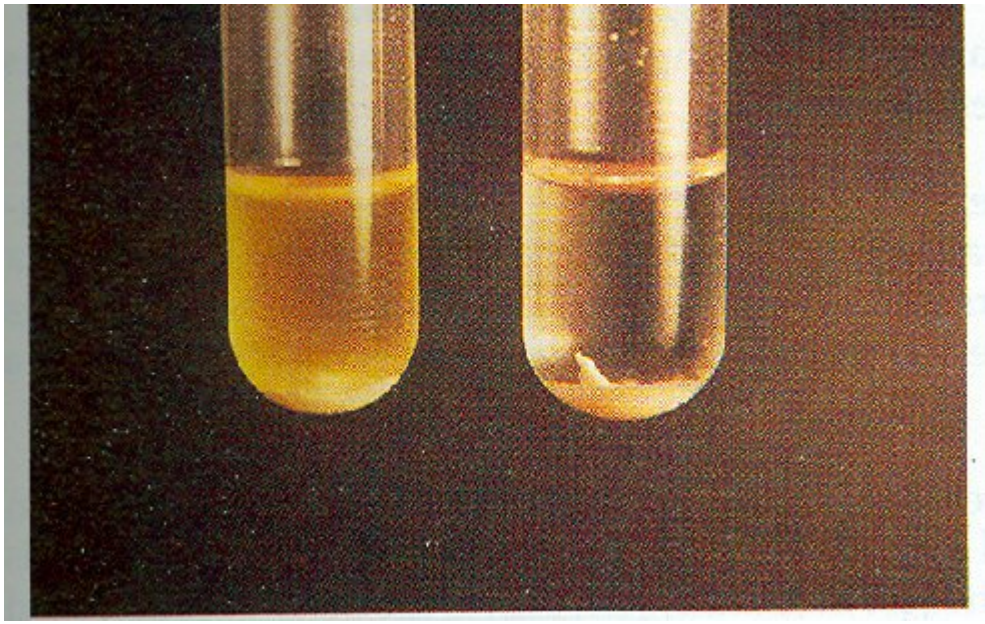
- Selective for sal. & sh.
- Bile salts
- Bromthymol blue
- Acid fuchsin
- Sodium thiosulfate
- Ferric ammonium citrate
- Lac + pink to orange
- Lac – blue green
- $H_2S \rightarrow$ ferric sulfide
  (black precipitate)

ONPG test

- 0-nitrophenyl-ß-galactopyranoside
- Tow enzyme : permease and ß-galactosidase
- Lactose _____ glucose + galactose
- Slow fermenters only have ß-galactosidase

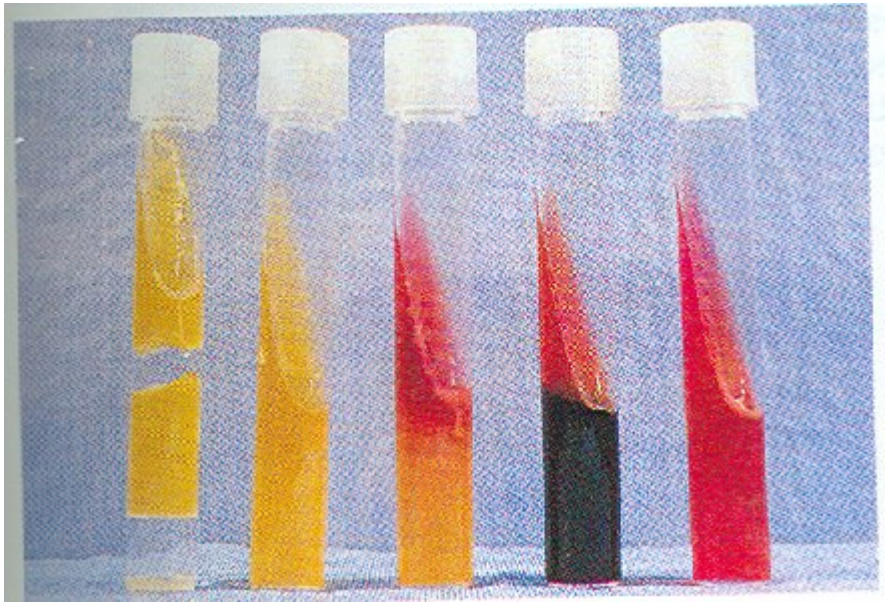
ONPG+ ß-galactosidase(hydrolysis) →

O-nitrophenol(yellow)



Triple sugar iron agar(TSI)

- Lactose, glucose and sucrose
- Ferrous sulfate
- Phenol red
- Reveal gas and H2S production
- H2S+ iron salts_____ black precipitate (ferric sulfide)



Indole test

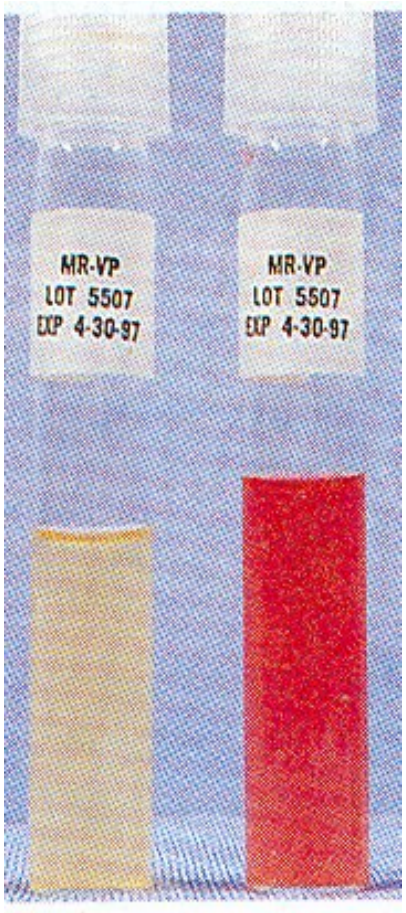
- Tryptophan $\xrightarrow{\text{tryptophanase}}$ acid pyruvic +ammonia+Indole
- Kovac's reagent (HCl+ p-dimethylaminobenzaldehyde+n-amyl alcohol)

Methyl red (MR) test

- Glucose fermentation(PH=4.2) acid formation
- Methyl red dye remain red when adding in the tube
- MR negative bacteria yellow to orange

Voges-Proskauer(VP)

- Determine whether the product of glucose is acetylmethylcarbinol(acetoin) neutral metabolite
- Acetoin reacts with Barrit's reagent(KOH & creatinine) to form a pink to rosy red tinge in the medium
- In negative result the tube remains brown to yellow

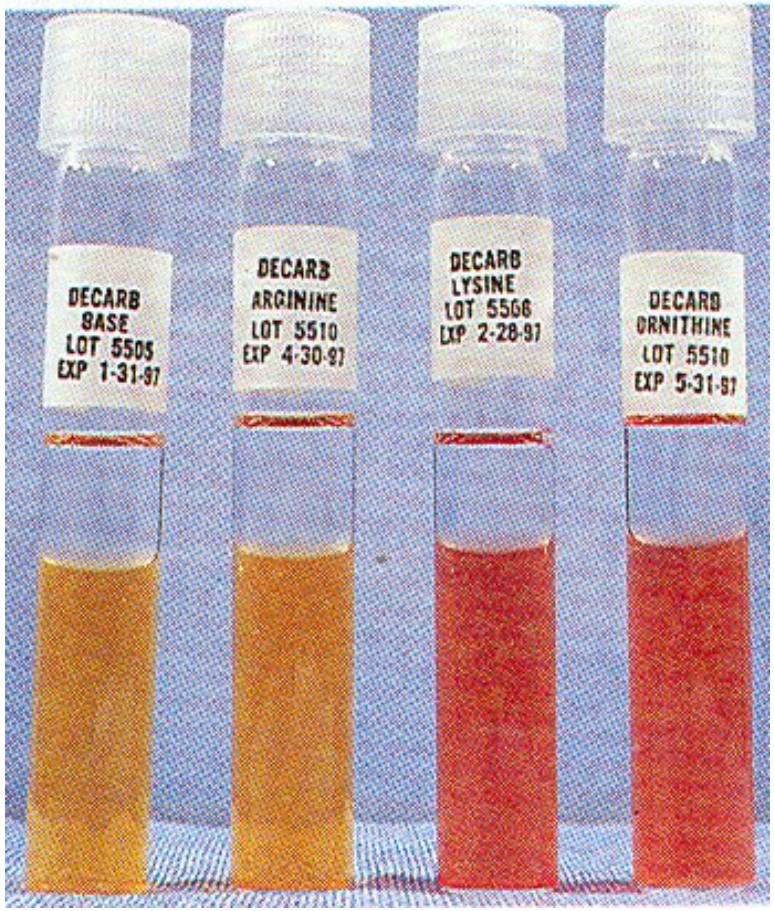


Citrate media

- Contain citrate as the only usable carbon source
- Ammonium dihydrogen phosphate
- Bromthymolblue(indicator)
- By using citrate alkaline by-products
- The color of medium from green(neutral) to blue(alkaline)

Lysine and ornithine decarboxylase(LDC, ODC)

- Enzymes that remove carboxyl from aminoacids
- The end products are alkaline amines
- The raise of PH convert bromcresol purple to purple color
- Negative results are yellow



Urea medium

- 2% Urea
- Phenol red

Urea $\xrightarrow{\text{urease}}$ ammonia +CO_2

The indicator turns bright pink

Phenylalanine(PA) deaminase

- Remove an amino group from PA
- Produce phenylpyruvic acid
- Visualized by adding ferric chloride10%
- Indicator turns olive green

Gelatinase test

- Below 32 semisolid
- Above 32 viscous

Motility

- Hanging drop slide
- Motility test medium