

KEY TO ALL LABORATORY ORGANISMS

ORGANISM IS **GRAM-POSITIVE**:
 ORGANISM IS **GRAM-NEGATIVE**:

GO TO SECTION I
 GO TO SECTION II

I. ALL GRAM POSITIVE ORGANISMS

A. ALL GRAM-POSITIVE ORGANISMS STUDIED

1. Organism is a **coccus**: Go to Section B.
2. Organism is a **bacillus**: Go to Section E.

B. ALL GRAM-POSITIVE COCCI

1. **Catalase positive**; some arranged in grape-like clusters and others in regular arrangements of 2,4,6 or 8: Go to Section C.
2. **Catalase negative**; most characteristic arrangement is in chains although chain length can be as small as 2; some members are not true cocci appearing as cocco-bacilli: Go to Section D.

C. MICROCOCCACEAE

1. **Random arrangement in grape-like clusters**; nutrient agar colonies with flat generally white in color with even margins; luxurious growth on **6.5% salt media**; characteristically commensals or pathogens of vertebrates including man:
Go to Section C.3.
2. **Regular arrangement** in groups of 2,4,6, or 8; moderate to light growth on **6.5% salt media**; nutrient agar colonies moderate to small domed, round, entire colonies; most are gray but some can be highly pigmented when cultured at room temperature; characteristically saprophytic organisms:
Go to Section C.4
3. *Staphylococcus*
 - a. Ferments **mannitol**: Organism: *Staphylococcus aureus*
 - b. Does not ferment **mannitol**: Organism: *Staphylococcus epidermidis*
4. *Micrococcus*
 - a. **Urease positive**: Ferments maltose and mannitol; gelatinase positive. Arrangement usually in groups of 4; Organism: *Micrococcus ureae*
 - b. **Urease Negative**: Does not ferment mannitol; gelatinase negative. Arrangement most often observed is groups of 4, 6 & 8 from colonies on nutrient media.
Organism: *Micrococcus luteus*

D. STREPTOCOCCACEAE

1. **Blood hemolysis**
 - a. β -hemolytic: Go to D.2
 - b. α -hemolytic: Go to D.3
 - c. γ -hemolytic: Go to D.4
2. **β -hemolytic Streptococci**
 - a. Ferments **mannitol**: Organism: *Streptococcus pyogenes*
 - b. Does not ferment **mannitol**: Organism: *Streptococcus galactiae*

3. α -hemolytic Streptococci

- a. Almost always observed in Gram stains as diplococci; ferments **inulin** & **raffinose**
(Ask instructor for results of this test if media is not available):

Organism: *Streptococcus pneumoniae*

- b. Appears in short to medium numbered chains; does not ferment **inulin** & **raffinose**
(Ask instructor for results of this test if media is not available).

Organism: *Streptococcus salivarius*

4. γ -hemolytic streptococci

- a. Salt tolerant; ferments **mannitol**: **Organism:** *Enterococcus faecalis*

- b. Salt intolerant; does not ferment **mannitol**:

Organism: *Streptococcus mutans*

E. ALL GRAM-POSITIVE BACILLI STUDIED**1. Cellular Morphology**

- a. Cells regular shaped looking like rectangles: **Go To Section E.2**
b. Cells irregular in shape: **Go To Section E.5**

2. Catalase Test

- a. **Catalase** and **amylase** positive: **Go To Section E.3**
b. **Catalase** and **amylase** negative: **Go To Section E.4**

3. *Bacillus*: All members form endospores that are best observed in Gram stains of older cultures.

- a. Ferments **mannitol**: **Organism:** *Bacillus subtilis*
b. Does not ferment **mannitol**: **Organism:** *Bacillus thuringiensis*

4. *Lactobacillus*

- a. Ferments **raffinose**: **Organism:** *Lactobacillus casei*
b. Does not ferment **raffinose**: **Organism:** *Lactobacillus acidophilus*

5. Cellular Morphology

- a. Cells observed in a variety of polymorphic forms: X, Y and V shaped very common.
Go To Section E.6
b. Cells observed as long slender pointed rods in chains resembling long strands like fungal mycelia. **Go To Section E.7**

6. *Corynebacterium*

- a. **Urease** positive **Organism:** *Corynebacterium pseudodiphtheriticum*
b. **Urease** negative **Organism:** *Corynebacterium xerosis*

7. *Mycobacterium*

- a. Ferments mannose and rhamnose. (Ask instructor for results of this test if media is not available.) **Organism: *Mycobacterium phlei***
- b. Does not ferment mannose and rhamnose. (Ask instructor for results of this test if media is not available.) **Organism: *Mycobacterium smegmatis***

II. ALL GRAM NEGATIVE ORGANISMS

A. ORGANISM IS A COCCUS: go to Section C.

B. ORGANISM IS A BACILLUS OR COCCOBACILLUS: Go to Section D.

C. *NEISSERIA***1. Lactose fermentation**

- a. Does not ferment lactose: **Organism: *Neisseria gonorrhoeae***
- b. Ferments lactose: **Go To C.2**

2. Sucrose fermentation

- a. Does not ferment sucrose **Organism: *Neisseria meningitidis***
- b. Ferments sucrose **Organism: *Neisseria sicca***

D. ALL GRAM-NEGATIVE BACILLI

1. Ferments Lactose: **Go to Section E**
2. Does not ferment Lactose: **Go to Section F**

E. ALL LACTOSE FERMENTING BACILLI**1. Indole test:**

- a. Indole positive; citrate negative **Organism: *Escherichia coli***
- b. Indole negative: **Go to Section E.2**

2. Hydrogen Sulfide Production

- a. H₂S produced; citrate positive **Organism: *Citrobacter freundii***
- b. H₂S not produced: **Go to Section E.3**

3. Urease Test

- a. Urease positive **Organism: *Klebsiella pneumoniae***
- b. Urease negative **Organism: *Enterobacter aerogenes***

F. LACTOSE NONFERMENTERS

1. Produce hydrogen sulfide: **Go to Section G**
2. Do not produce hydrogen sulfide: **Go to Section J**

G. HYDROGEN SULFIDE PRODUCERS

1. Urease positive: **Go to Section H.**
2. Urease negative: **Go to Section I.**

H. *PROTEUS*

1. Indole positive **Organism: *Proteus vulgaris***
2. Indole negative **Organism: *Proteus mirabilis***

I. SALMONELLA

1. Citrate negative: **Organism: *Salmonella typhi***
2. Citrate positive: **Organism: *Salmonella typhimurium***

J. HYDROGEN SULFIDE NEGATIVE ORGANISMS

1. Oxidase negative; ferments glucose: Go to **Section K.**
2. Oxidase positive; does not ferment glucose or other carbohydrates. Go to **Section N.**

K. OXIDASE NEGATIVE GLUCOSE FERMENTING ORGANISMS

1. Citrate positive: **Go to Section L.**
2. Citrate negative: **Go to Section M.**

L. SERRATIA **Organism: *Serratia marcescens***

M. SHIGELLA

1. Indole positive: **Organism: *Shigella flexneri***
2. Indole negative: **Organism: *Shigella sonnei***

N. OXIDASE POSITIVE NONFERMENTING ORGANISMS

1. Gelatinase positive: **Organism: *Pseudomonas aeruginosa***
2. Gelatinase negative: **Organism: *Alcaligenes faecalis***