ONE BIG FAMILY



A simple guide for the identification of bacteria

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INTRODUCTION

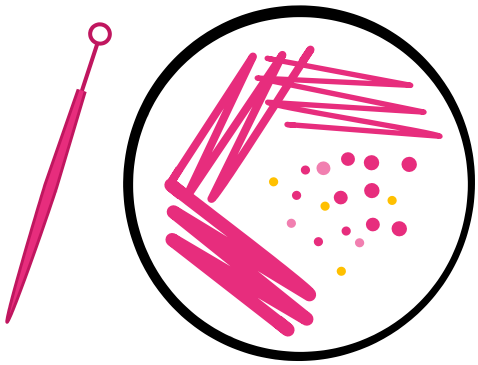
The purpose of this booklet is to provide a **quick** and **easy** guide to the classification of clinically important bacteria.

The classification is principally based on the **GRAM STAIN** result of the organism. Further diagnostic tests include: **BIOCHEMICAL** reactions AND sensitivity to **ANTIBIOTICS**.

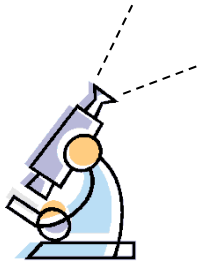
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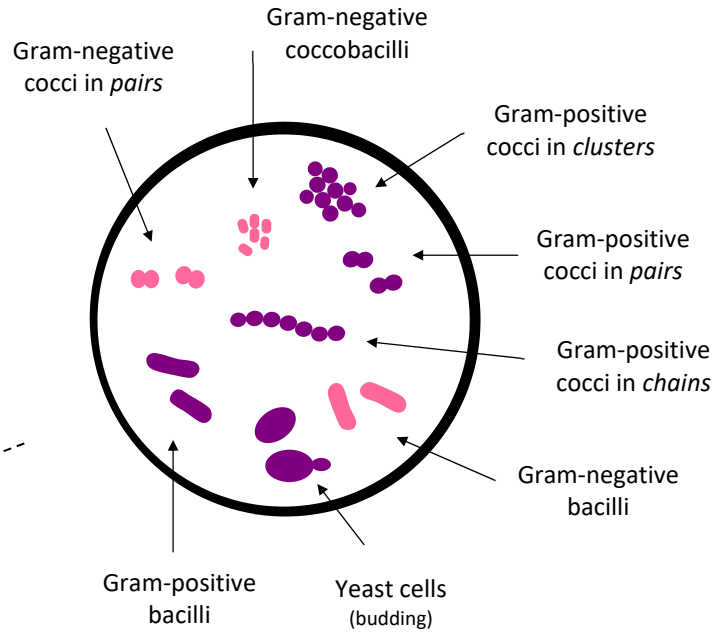
REMEMBER that a good **IDENTIFICATION** always begins with **SEPARATED COLONIES** and a clear **GRAM STAIN** result.

1 **CULTURE**



2 **STAIN**





ABBREVIATIONS & SHAPES

The following **ABBREVIATIONS** and **SHAPES** are used throughout this booklet:

GPB	Gram Positive Bacilli
GPC	Gram Positive Cocci
GNB	Gram Negative Bacilli
GNC	Gram Negative Cocci
GNCB	Gram Negative Coccobacilli
AFB	Acid Fast Bacilli
S	Sensitive
R	Resistant
O_2	Strict aerobic (O_2 present)
CO_2	Capnophilic (5% CO_2)
MO_2	Microaerophilic (Low O_2)
$Ø_2$	Strict anaerobic (O_2 absent)
	Spore-forming
	Motile
	Diplococci
	May produce β-lactamases
	Curved bacilli
	DNase positive
	Bile Esculin positive
	Lactose fermenter
	Non-lactose fermenter
	Low virulence organism

THE FAMILY STONE

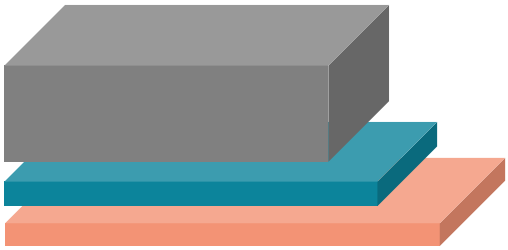
The following diagrams compare the **CELL WALL** structure of **Gram-positive**, **Gram-negative** and **Acid-fast** bacteria:

GRAM-POSITIVE CELL WALL



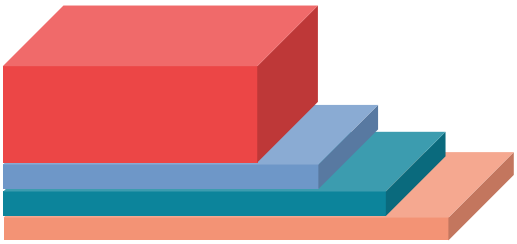
Peptidoglycan (>60%)
Cytoplasmic membrane

GRAM-NEGATIVE CELL WALL

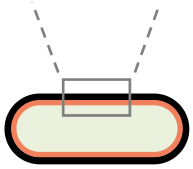


Outer membrane → Lipopolysaccharide
Periplasmic space
Peptidoglycan (5-10%)
Periplasmic space
Cytoplasmic membrane

ACID-FAST CELL WALL

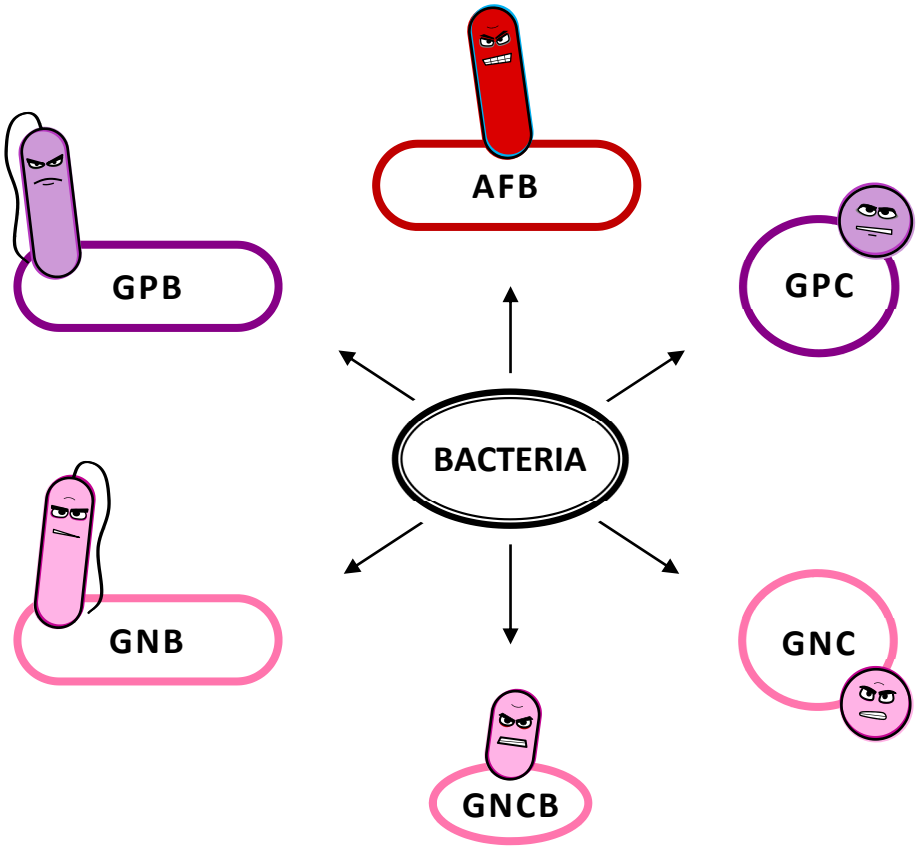


Glycolipids → Mycolic acid (60%)
Arabinogalactan
Peptidoglycan
Cytoplasmic membrane



Cell wall
Cell membrane
Cytoplasm

THE BIG FAMILY

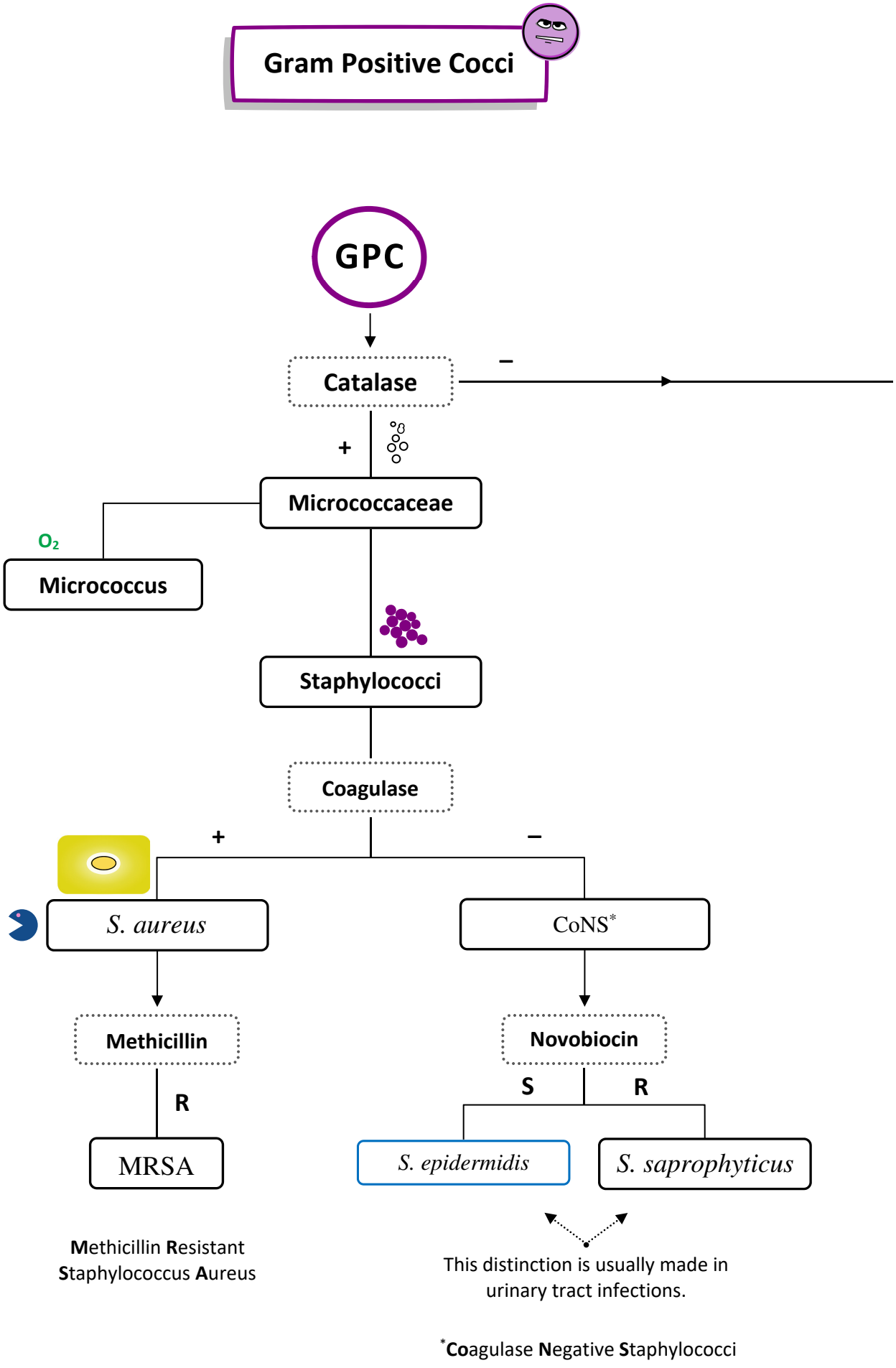


NOTES

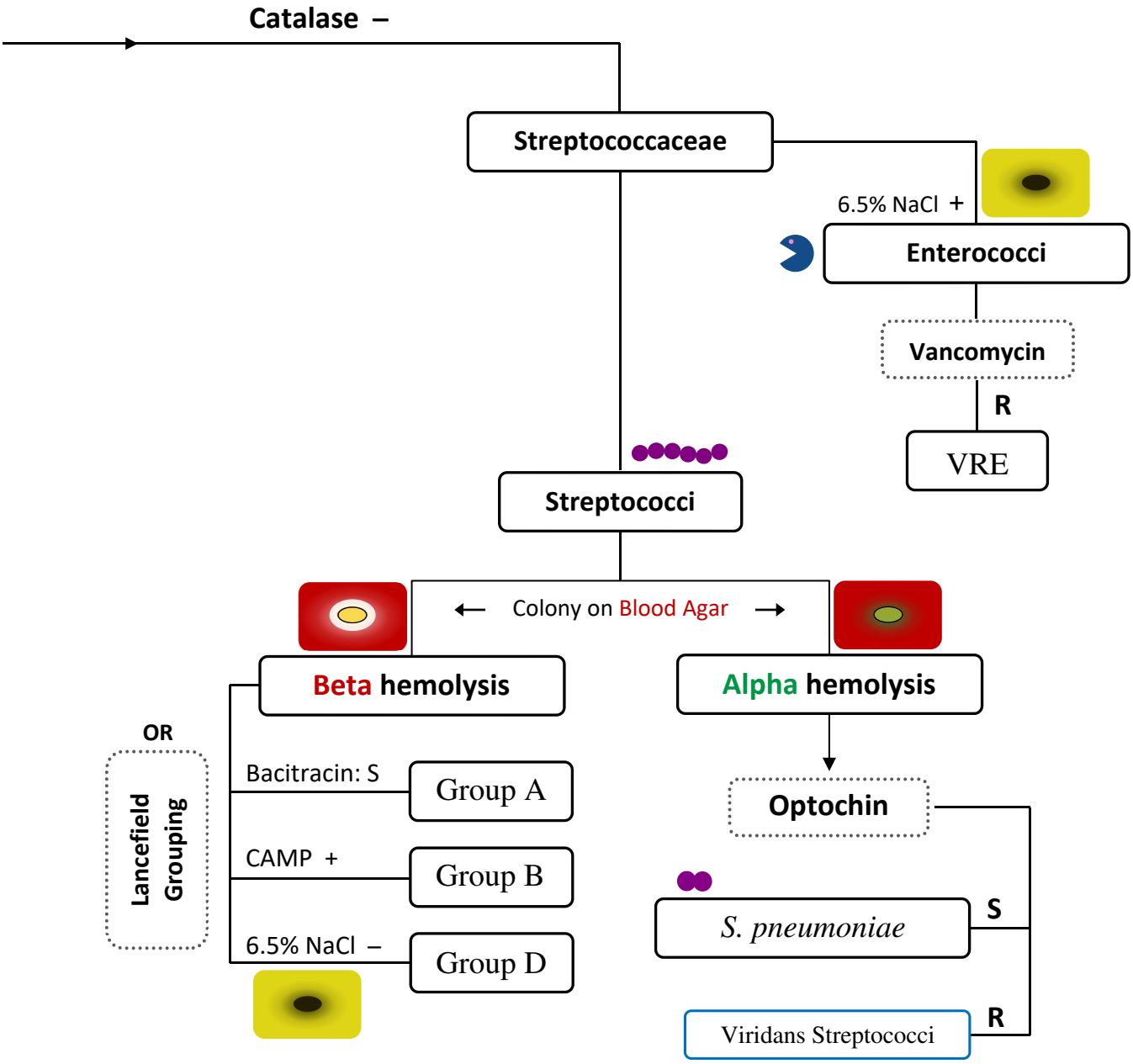
When stained using the Gram stain: **GRAM-POSITIVE** bacteria appear **PURPLE**, while **GRAM-NEGATIVE** bacteria appear **PINK**.

ACID FAST BACILLI (AFB) are stained using special dyes such as the Ziehl-Neelson stain and they appear as **RED** bacilli against a *blue* background.

In addition, there are other types of bacteria that belong to different groups such as the ***Spirochetes***, ***Chlamydia*** and ***Mycoplasmas***. These are not included in this guide, as their diagnosis is based on different detection or culture techniques.


Gram Positive Cocci
GPC
Catalase
–
+
Micrococcaceae
O_2
Micrococcus
Staphylococci
Coagulase
+
–
S. aureus
CoNS*
Methicillin
R
MRSA
Novobiocin
S
R
S. epidermidis
S. saprophyticus
Methicillin Resistant Staphylococcus Aureus
This distinction is usually made in urinary tract infections.
*Coagulase Negative Staphylococci

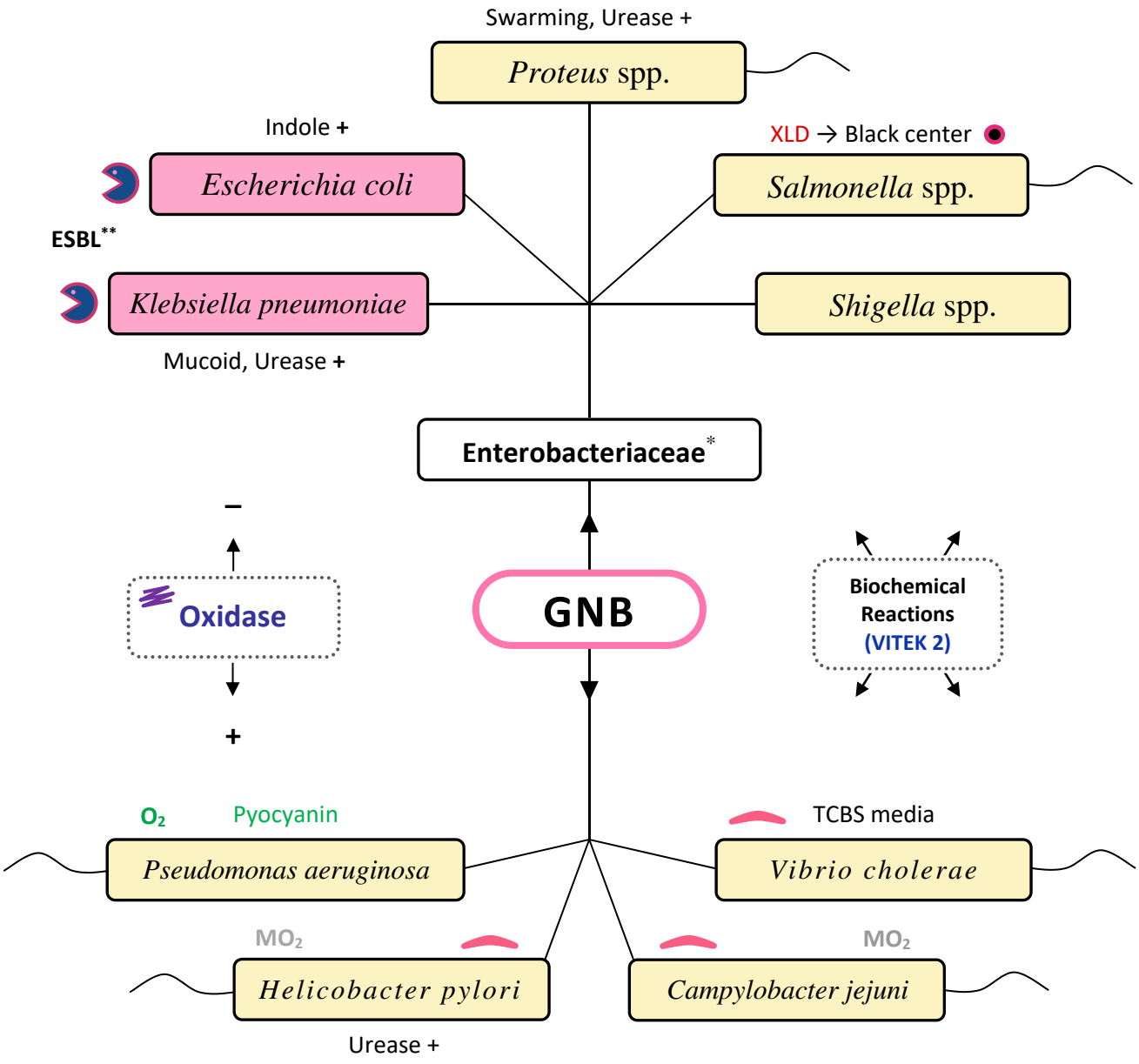
...continued



Group A = *Streptococcus pyogenes*
Group B = *Streptococcus agalactiae*
Group D = Non-Enterococci

Enterococci include *E. faecalis* & *E. faecium*
VRE: **V**ancomycin **R**esistant **E**nterococci
Viridans Strept include *S. mitis* & *S. mutans*

Gram Negative Bacilli

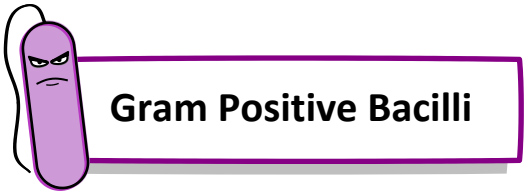


May produce **ESBL enzymes
(Extended Spectrum Beta-Lactamases)
=> Resistance to cephalosporins

***Other important Enterobacteriaceae include:**

- *Citrobacter freundii*
- *Enterobacter cloacae*
- *Morganella morganii*
- *Serratia marcescens*
- *Yersinia enterocolitica*

Identify using VITEK 2



Psychrophile

B. cereus

L. monocytogenes

B. anthracis

Listeria

Bacillus

All

Catalase +

Lactobacilli

Catalase –

GPB

Corynebacterium

Diphtheroids

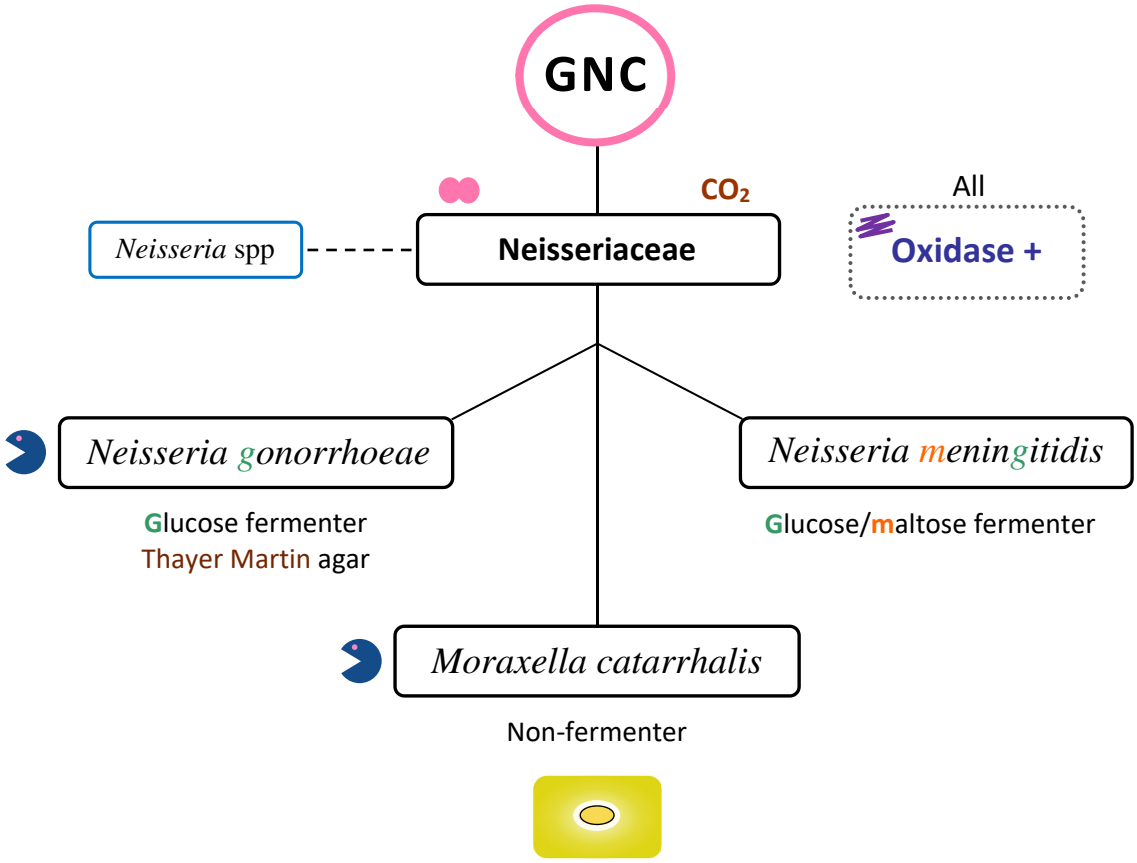
C. diphtheriae

Chinese letters

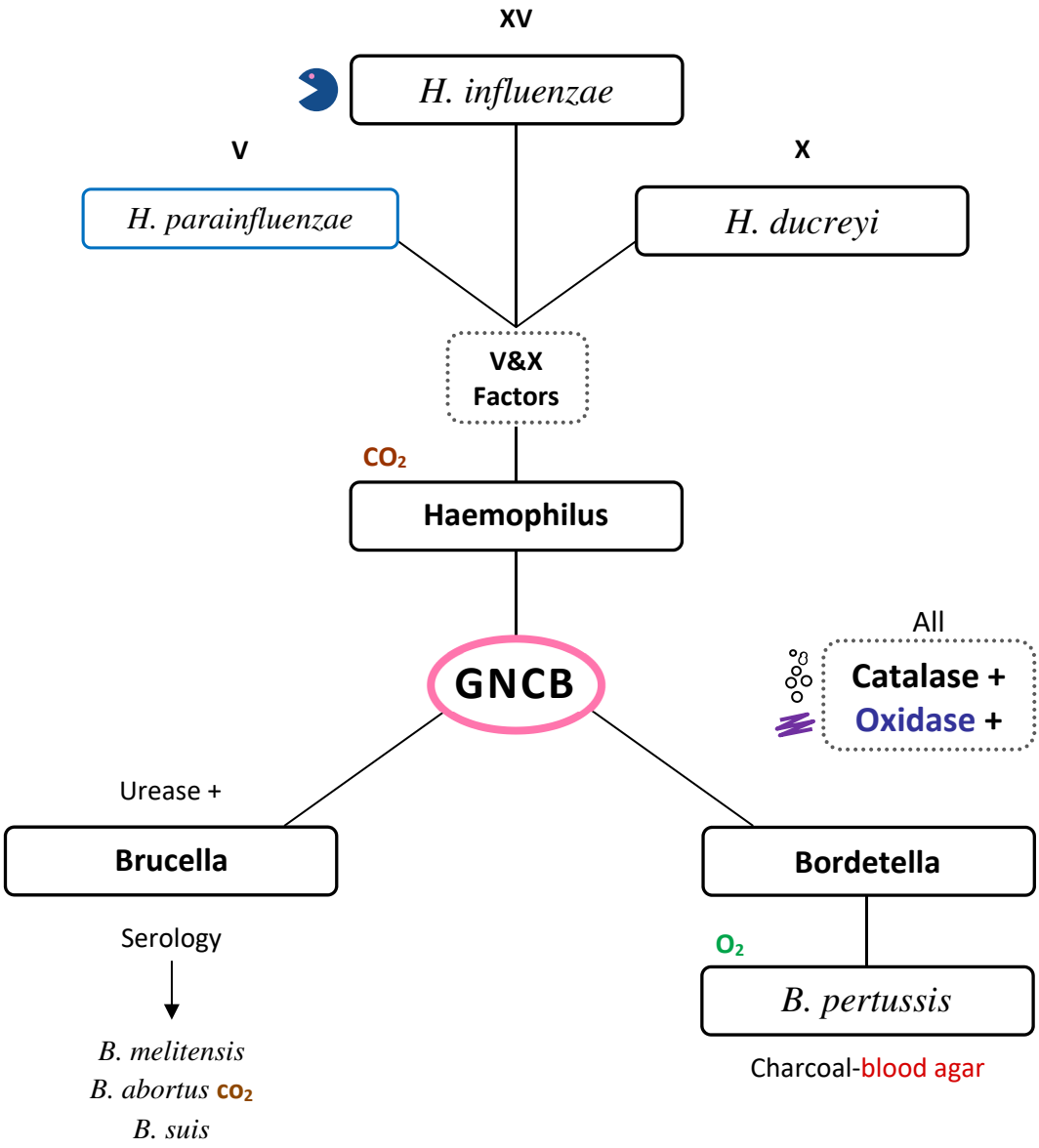
Tellurite-blood agar

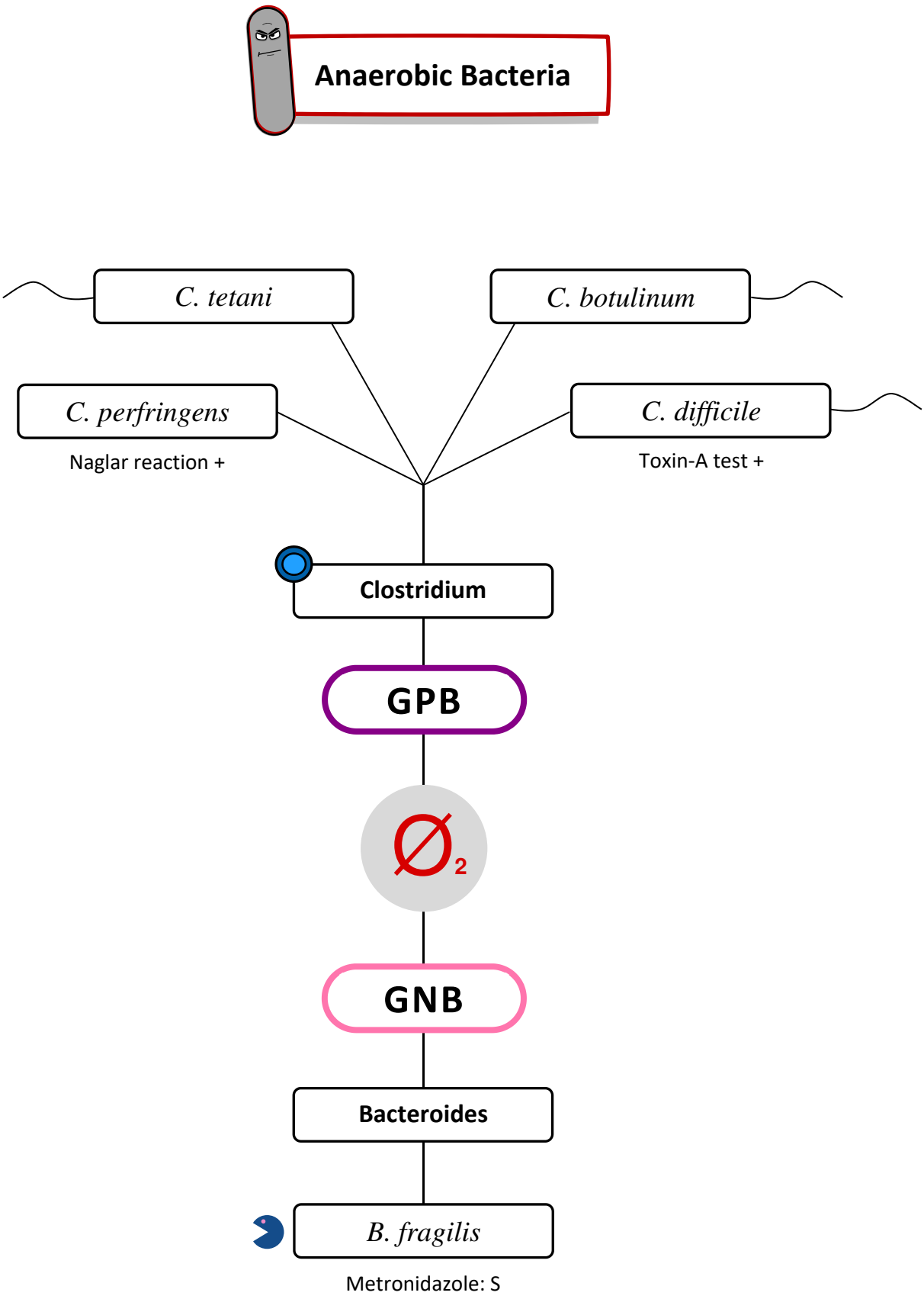
(Toxigenic) Elek test +

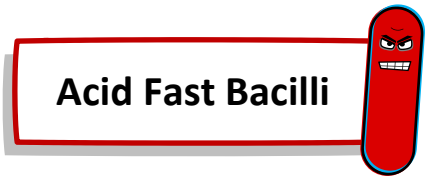
Gram Negative Cocci

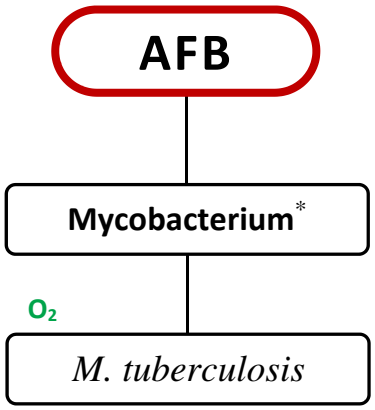


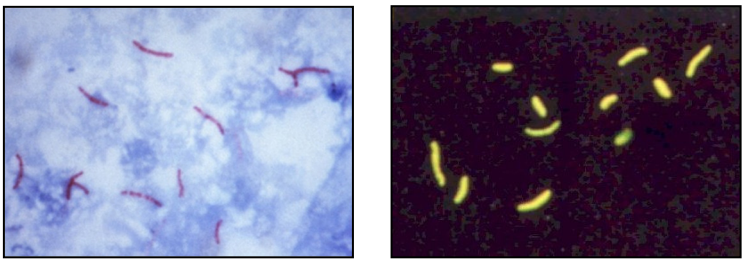
Gram Negative Coccobacilli




Anaerobic Bacteria
C. tetani
C. botulinum
C. perfringens
Naglar reaction +
C. difficile
Toxin-A test +
Clostridium
GPB
∅2
GNB
Bacteroides
B. fragilis
Metronidazole: S







TB rods stained using Ziehl-Neelsen (**ZN**) stain
& Auramine-Rhodamine (**AR**) fluorescent stain

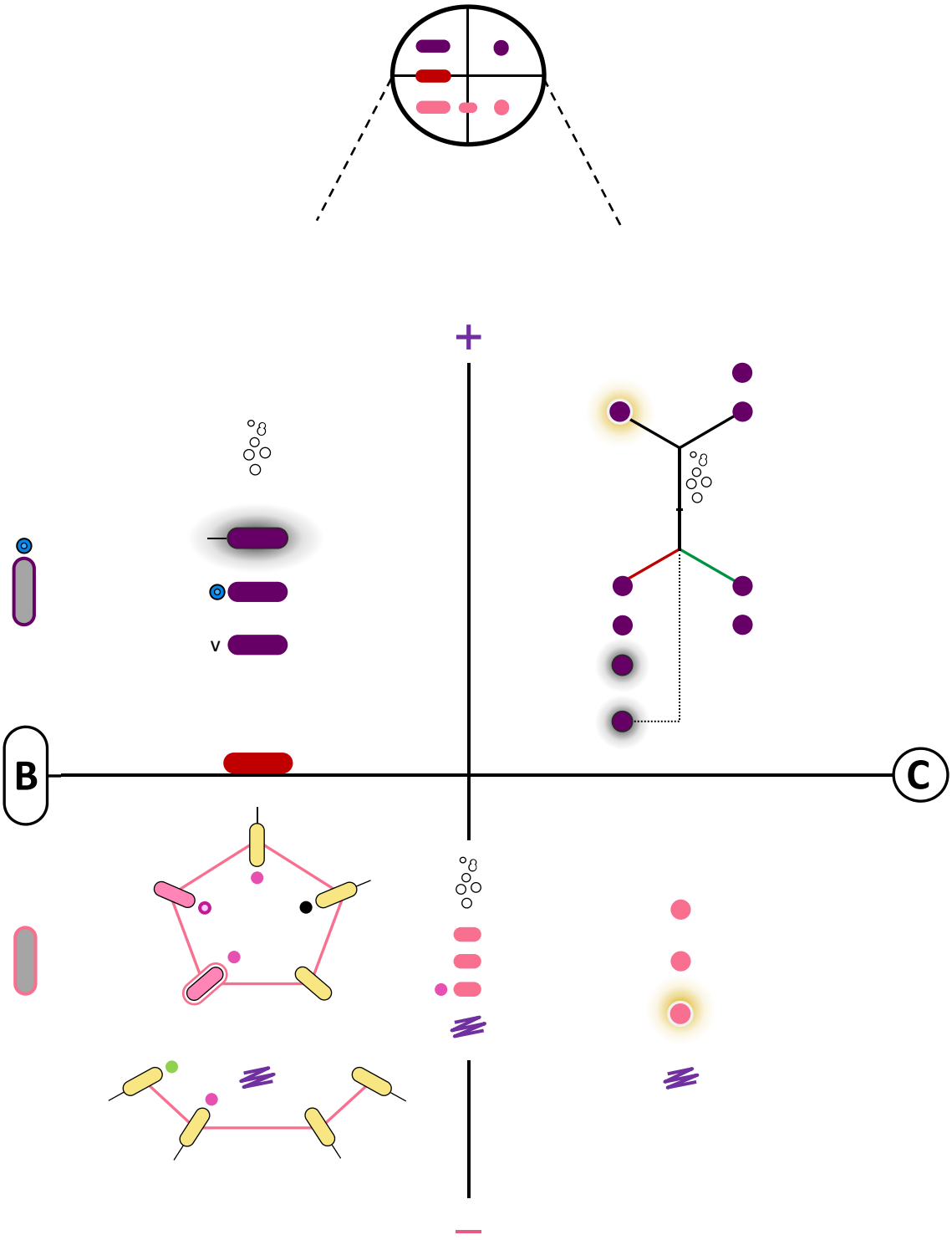


TB colonies grown on
Lowenstein Jensen (**LJ**) medium

*Mycobacterium species include:
Mycobacterium tuberculosis (**MTB)** => TB infection
Mycobacteria Other Than Tuberculosis (**MOTT**) => TB-like infection

THE BIG GATHERING

(Summary map of organisms)



MEMBER PROFILES

(Growth requirement & colony appearance)

	ORGANISM	MEDIA*	INCUBATOR	COLONY MORPHOLOGY
GPC	*S. aureus*	BAP & MSA	37°C	β-hemolysis, yellow on MSA
	S. pyogenes	BAP	$Ø_2$	β-hemolysis
	S. pneumoniae	BAP	CO_2	α-hemolysis, doughnut shape
	Enterococcus spp	BAP	37°C	Small, white
GNB	*E. coli*	MAC	37°C	Pink, dry
	K. pneumoniae	MAC	37°C	Pink, mucoid
	Proteus spp	MAC	37°C	Pale yellow
	Salmonella spp	DCA & XLD	37°C	Pale yellow/pink, black center
	Shigella spp	DCA & XLD	37°C	Pale yellow/pink
	P. aeruginosa	MAC	37°C	Pale yellow, pyocyanin
	V. cholerae	TCBS	37°C	Yellow (sucrose fermenter)
GNCB	*H. influenzae*	CAP	CO_2	Small, translucent
	Brucella spp	BAP	37°C	Gray-white
	B. pertussis	Charcoal-BAP	37°C	Mercury-like
GPB	*C. diphtheriae*	Tellurite-BAP	37°C	Gray-black
	L. monocytogenes	BAP	37°C	β-hemolysis
GNC	*N. meningitidis*	BAP	CO_2	Shiny, grayish
	M. catarrhalis	BAP	CO_2	Crumble, slide across
Others	*C. perfringens*	BAP	$Ø_2$	Double β-hemolysis
	B. fragilis	BAP	$Ø_2$	Small, sensitive to MTZ disc
	M. tuberculosis	LJ medium	37°C	Rough, tough and buff

* To show characteristic morphology, but the organism may also grow on other media.

- **ABBREVIATIONS:**

BAP = Blood Agar Plate
MSA = Mannitol Salt Agar
MAC = MacConkey agar
LJ = Lowenstein Jenson
CAP = Chocolate Agar Plate
DCA = Deoxycholate Citrate Agar
XLD = Xylose Lysine Desoxycholate
TCBS = Thiosulfate Citrate Bile salts Sucrose



(Normally present in healthy individuals)

BODY SITE		NORMAL FLORA
	Skin, Eye & Urethra	CoNS* Viridans Streptococci Diphtheroids
	Oropharynx	CoNS* Viridans Streptococci Diphtheroids Neisseria spp
	Vagina	GBS CoNS* Lactobacilli Candida spp
	Large intestine	Bacteroides Clostridium Coliforms** Proteus Pseudomonas
BODY FLUIDS	**CSF** **Blood** **Urine** (in bladder) **Synovial Fluid** **Ascitic Fluid**	Normally Sterile

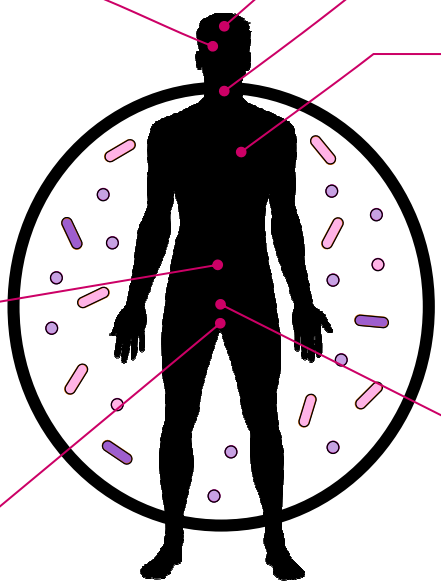
* **CoNS:** Coagulase Negative Staphylococci (e.g. *S. epidermidis*).

** **Coliforms**: are lactose-fermenting Enterobacteriaceae, such as *Escherichia*, *Klebsiella*, *Citrobacter* and *Enterobacter*.

THE USUAL SUSPECTS



(Common causative agents)



Meningitis
N. meningitidis
S. pneumoniae
H. influenzae
E. coli
GBS

Eye infection
H. influenzae
S. pneumoniae
P. aeruginosa

Pharyngitis
Strept Group A,C,G
H. influenzae (<6yrs)

Septicemia
Staphylococci
Streptococci
GNB
C. albicans
Anaerobes

Pneumonia
S. pneumoniae
H. influenzae
M. catarrhalis
GNB

Skin & Wound
S. aureus
S. pyogenes
GNB
Anaerobes

Gastroenteritis
Salmonella
Shigella
Campylobacter
Helicobacter
Vibrio

Urinary Tract Infection
E. coli
Proteus
K. pneumoniae
P. aeruginosa
Enterococcus
S. saprophyticus
C. albicans

Genital infection
N. gonorrhoeae
H. ducreyi
G. vaginalis
C. albicans

Other Body Fluids
Staphylococci
Streptococci
GNB
Anaerobes